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Oral Presentation



Hematology



Amino Acids Profile in Patients with Myocardial Infarction

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Background and Aim: Myocardial infarction (MI) is one of the preeminent causes of death universal. Altered amino acids (AAs) metabolism has been linked to cardiovascular complications, signifying their role in the pathogenesis of the cardiovascular disease (CVD). A cross-sectional case-control study was set to profile AAs in MI investigate their potential as diagnostic and prognostic.

Methods: A total of 60 subjects was selected, including 40 MI patients and 20 controls. Ninhydrin derivatization-High pressure liquid chromatography was employed to quantify the plasma AAs. The GRACE risk score was calculated for the cases. The receiver operating characteristic curve (ROC) was calculated to assess potential markers for the diagnosis ability of MI.

Results: Fourteen AAs exhibited discriminative values between cases and controls. Of these, BCAA (leu, ile), methionine, glycine, threonine, serine, ornithine, arginine, histidine, and tyrosine exhibited low levels in MI cases compared to healthy controls ($p \leq 0.001$). Meanwhile, MI cases represented a higher abundance of cysteine, taurine ($p \leq 0.05$), total aromatic AAs ($p \leq 0.01$), and asparagine ($p \leq 0.001$). Only leucine, isoleucine, and asparagine were correlated to the GRACE risk score. Methionine and glycine scored highly on ROC analysis ($AUC > 0.95$), sensitivity ($>97\%$), and specificity ($>95\%$) followed by histidine ($AUC > 0.92$), sensitivity ($>82\%$), and specificity ($>85\%$).

Conclusion: Our study designated a panel of 14 plasma circulating AAs perturbed in MI. Methionine, glycine, and histidine were potential markers to recognize MI cases from healthy Individuals. Eventually, plasma AAs profiling could help diagnose but is less potent as a prognostic marker for MI.

Keywords: Amino acids; Myocardial infarction; Cardiovascular disorders; Pathogenesis; Diagnosis.

Combined Mild Hypoxia and Bone Marrow Mesenchymal Stem Cells Enhances Expansion and Stemness of Human Cord Blood CD34+ Stem Cells

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Background and Aim: Cord blood (CB) is a rich source of Hematopoietic stem cells (HSCs) that has been used successfully to treat a variety of disorders. Beside the advantage of CB, the main disadvantages of CB are the limited number of stem cells available for transplantation and delayed engraftment. Identifying strategies to enhance expansion, and maintain stemness of HSCs can improve transplant efficiency. The goal of this study was to examine different culture conditions on ex vivo expansion and stemness capacity of CB-HSCs.

Methods: In this study, human cord blood CD34+ HSC, cultured in the serum-free media supplemented with cytokines (TPO, FLT3L, SCF) with/without Bone marrow Mesenchymal stem cell (BM-MSC) feeder layer in normoxia (21% O₂) and mild hypoxia (5% O₂) for 7 days. At 0 and 7 days, total nucleated cell count (TNC), CD34+ cells count, CFC assay, *c-MYC*, *SOX2* and *HOXB4* expression, as a stemness markers by Real time PCR were evaluated. The data analyzed using the t-test and ANOVA. P Value < 0.05 were considered statistically significant.

Results: At the end of 7 days of culture, highest number of total nucleated cell (TNC), CD34+ cells, colony forming units (CFUs), *c-MYC*, *SOX2* and *HOXB4* levels were seen in coculture of HSC with MSC feeder layer at 5% O₂. Our findings demonstrated statistically significant (1.4-1.68 fold) increase of *SOX2* gene expression (1.4-1.6 fold) increase of *c-MYC* gene expression and (1.25-1.8 fold) increase of *HOXB4* gene expression in hypoxia versus normoxia.

Conclusion: BM-MSC and mild hypoxia combination not only improves HSC expansion but also enhanced stemness capacity of HSC and better mimicked the niche microenvironment. These findings are expected to contribute to the development of more efficient culture systems for the ex vivo expansion of CB HSCs.

Keywords: Cord blood; Hematopoietic stem cell; CD34+ cells; Mesenchymal stem cell; hypoxia; *SOX2*; *c-MYC*; *HOXB4*

Genotyping of Blood Groups in Alloimmunized Patients with β -Thalassemia Major by T-ARMS-PCR and Multiplex-aso-pcr

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Background and Aim: Beta-thalassemia major is severe hemolytic anemia requiring life-long blood transfusion. Planned random donor blood transfusion is associated with alloimmunization against incompatible antigens. Determination of the minor blood group systems phenotype or genotype, and administration of the compatible blood components can significantly reduce the rate of alloimmunization. The present study aimed to determine the prevalence of alloimmunization, and genotype/phenotype characteristics of the minor blood groups systems in patients with β -thalassemia major.

Methods: This study was conducted on 1147 β -thalassemia major patients. Initially, antibody screening and antibody identification were performed. Then, phenotyping and genotyping for the Rh, Kell, Kidd, and Duffy blood groups were done in alloimmunized patients using monoclonal antibodies and Multiplex-Allele Specific Oligonucleotide-Polymerase Chain Reaction (Multiplex-ASO-PCR) and Tetra-primer amplification refractory mutation system-PCR (T-ARMS-PCR), respectively. Any phenotype/genotype discrepancy was assessed by direct sequencing.

Results: Ninety-seven (8.5 %) out of 1147 patients had alloantibodies against the minor blood group antigens (44 males, 45.4 %, and 53 females, 54.6 %). The most common alloantibodies were against the RH (n: 47, 48.5 %), and the Kell (n: 23, 23.7 %) blood groups systems. Twenty-three (2.1 %) genotype/phenotype discrepancies out of 1067 tests, including 9 in the Rh (9.3 %), 8 in Duffy (34.8 %), and 6 in Kidd (26.1 %) blood groups were detected. No discrepancy was found in the Kell blood group system. Direct sequencing revealed that the results of molecular methods were correct.

Conclusion: Multiplex-ASO-PCR and T-ARMS-PCR molecular methods are fast, reliable, and cost-benefit molecular methods for the minor blood group genotyping in multi-transfused β -thalassemia major patients.

Keywords: Alloimmunization; Blood transfusion; β -thalassemia major; Minor blood groups; Multi-transfusion

Renal Function in Patients with Thalassemia Major Receiving Exjade® Dispersible Tablets and a New Film-Coated Tablet Formulation of Deferasirox (Nan jade®)

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Background and Aim: In patients with β -thalassemia major (TM), one of the long-term complications of regular blood transfusion is renal dysfunction. The purpose of the current study was to evaluate the renal function in TM patients receiving Exjade® dispersible tablets and a new film-coated tablet formulation of deferasirox (Nan jade®).

Methods: In this descriptive cross-sectional study, a total of 80 TM patients aged 11 to 48 years old entered the study. Patients received 20 to 30 mg/kg/d (single dose) Exjade® (Exjade group, n=40) and Nan jade® (Nan jade group, n=40) orally. To evaluate renal function, serum creatinine (SCr), estimated glomerular filtration rate (eGFR), blood urea nitrogen (BUN), 24-h urine protein (UPro), UCa/UCr, spot UPro/UCr ratio and serum ferritin were calculated at baseline and every 3 months to 9 months.

Results: There was no significant difference in SCr, BUN, eGFR, 24-h UPro, UPro/UCr ratio, UCa/UCr ratio and serum ferritin between groups at baseline and end of study ($P_{\text{baseline}} > 0.05$, $P_{\text{end of study}} > 0.05$). There was no significant difference in proteinuria between groups at baseline and end of study ($P_{\text{baseline}} > 0.05$, $P_{\text{end of study}} > 0.05$).

Conclusion: The proportion of SCr, BUN, eGFR, 24-h UPro, UPro/UCr ratio, and UCa/UCr ratio were not significantly different in TM patients treated with Nan jade® Compared to patients received Exjade®. Nan jade® had similar effects to Exjade®, and therefore, the use of Nan jade® is safe in TM patients and does not seem to be associated with increased renal failure, proteinuria and hypercalciuria.

Keywords: Thalassemia major; Renal function; Deferasirox



Immunology



Imipenem Alters Systemic and Liver Inflammatory Responses in CLP- Induced Sepsis Mice in a Dose-Dependent Manner

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Background and Aim: Considering the role of inflammation in the outcome of sepsis and the widespread use of imipenem in the disease, this study was designed to assess the effect of imipenem on the dynamics of inflammatory responses in sepsis mouse model.

Methods: Cecal Ligation and Puncture (CLP) model was used to induce sepsis in mice. C57BL/6 mice were divided into sham, CLP-induced sepsis mice, CLP-induced sepsis mice receiving 25 mg/kg, and 125 mg/kg imipenem. Blood and liver samples were obtained and bacterial load, endotoxin level, and liver enzymes were evaluated. The concentration and mRNA expression of cytokines were also determined.

Results: Sepsis mice treated with a high dose (125 mg/kg) of imipenem showed a significant reduction in bacterial load while increasing liver enzymes, endotoxin level, and inflammatory cytokine production in plasma and liver. In contrast, a significant reduction in the liver enzymes, bacterial load, endotoxin levels, and inflammatory cytokine levels were observed in the mice treated with a low dose (25 mg/kg) of imipenem compared with other mice groups. Liver tissue pathology of mice indicated little tissue destruction in the sepsis mice treated with 25 mg/kg of imipenem compared to other groups. Mice receiving 25 mg/kg of imipenem had a better survival rate.

Conclusion: Our results demonstrated the dose-dependent effect of subcutaneous administration of imipenem on the inflammatory responses in sepsis mice. A dose of 25 mg/kg imipenem resulted in better pathology, lower inflammatory mediators, and increased survival rate in sepsis mice.

Keywords: Imipenem; Sepsis; Liver; Inflammation; Immune responses

Potential Effect of the Accredited Vaccines on Body Protection against SARS-CoV-2 Infection, in Iran

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Background and Aim: Analyzing antibodies against spike-receptor binding domain (S-RBD) represented valuable tools to assess potential individual body protection against the current pandemic disease, COVID-19. There are still limited data available evaluating anti-RBD level in either affected or vaccinated individuals, particularly in Iran. In this study, we aimed to compare level of anti-RBD antibody in Iranian patients and normal individuals before and after vaccination.

Methods: A total of 181 individuals were classified into five different groups, including (A) those who were neither disease affected nor administrated vaccine, (B) those who were disease affected but not administrated vaccine, (C and D) non-affected individuals who have injected one or two vaccine shots against SARS-CoV-2 and (E and F) affected individuals who were administrated one or two vaccine shot(s). Peripheral blood sample was obtained from different groups and serum IgA/IgG anti-RBD antibody was analyzed using an ELISA kit (Razi Insititute, Iran). A cut-off was considered for anti-RBD level (≥ 1.15).

Results: Data analyses showed anti-RBD level of ≥ 1.15 in only 21% of the group (A) individuals. This antibody level was detected in 78% of the group (B) individuals. Curiously, level of the male antibody was significantly higher than female in this group. Compared to this group, the antibody level was determined in 91% of the individuals in groups (C) and (D). The highest range of anti-RBD antibody level (98%) was observed in groups (E) and (F).

Conclusion: Analyses of anti-RBD antibody level suggested that potential of body protection against SARS-CoV-2 was significantly higher in the affected male compared to the affected female. Additionally, the administration of accredited vaccines, in Iran, might increase body protection level rather than the patients with no vaccine administration.

Keywords: SARS-CoV-2; Anti-RBD Antibody; ELISA; Vaccine

The Prognostic Value of PD-1/PD-L1 Expression on Tumor Cells and Tumor-Infiltrating Immune Cells in Patients with Colorectal Cancer: Systematic Review and Meta-Analysis

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Background and Aim: Colorectal cancer (CRC) is one of the most common cancers worldwide. However, the role of immune checkpoint molecules, especially Programmed cell death protein-1 (PD-1) and Programmed cell death-ligand1 (PD-L1), in the progression of CRC remains unclear. This systematic review and meta-analysis investigate the prognostic significance of PD-1/PD-L1 expression on tumor-infiltrating immune cells and tumor cells in patients with CRC.

Methods: The protocol of this study has been prospectively registered in the PROSPERO (registration NO. CRD42020156233). A comprehensive electronic search on PubMed/MEDLINE, Scopus, Web of Science (WOS), Embase, and ProQuest was conducted using a combination of MeSH terms of “programmed cell death 1”, “programmed cell death ligand 1”, “colorectal”, and “cancer” between 1 January 1990 and 28 February 2020 with no language limitation. Two independent reviewers performed study selection, data extraction, and risk of bias assessment. The Newcastle-Ottawa Scale (NOS) for cohort studies is used to assess the risk of bias. Due to the high methodological heterogeneity between studies, a random-effect model was used for meta-analysis. Statistical heterogeneity was evaluated by the Chi² test with the I² statistic. “Egger’s statistical test and graph” was used to assess publication bias. Stata V.14 software employed for meta-analysis.

Results: Finally, 18 studies were included in this study. Among them, 12 studies evaluated the relationship between PD-L1 expression on tumor cells (tPD-L1) and prognosis, 7 examined the association between PD-L1 expression on tumor-infiltrating immune cells (iPD-L1) and prognosis, and three studies evaluated the prognostic value of PD-1 expression on tumor-infiltrating immune cells. Meta-analysis showed no relation between tPD-L1 and OS, DFS, and RFS. However, there is a small association between iPD-L1 and OS (HR=0.47; 95%CI: 0.37, 0.59; p less than 0.001), and patients with high expression of iPD-L1 had a better prognosis than others. There is no significant association between iPD-1 and OS. Expression of PD-L1 on both tumor cells and immune cells had no relationship with clinicopathological characteristics of patients except microsatellite instability (MSI) status.

Conclusion: The results of this study showed iPD-L1 might be a biomarker for a good prognosis of CRC.

Keywords: PD-1; PD-L1; Colorectal cancer; Prognosis; Survival



Medical mycology



Invasive Aspergillosis and Mucormycosis Co-Infection in COVID-19 Patients; Case Series

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Background and Aim: Secondary infection with other fungus can develop in patients with COVID-19, especially for severe or critically ill patients. Mucormycosis is an angioinvasive infection caused by fungi Mucorales, mainly in immunocompromised patients. Aspergillosis is also an opportunistic fungal infection caused by *Aspergillus* species. Coinfection with mucormycosis and aspergillosis is very rare, and very few cases have been published in the literature till now. There is an increase in the incidence of mucormycosis infection in post-COVID-19 patients.

Methods: blank

Results: Here, we are going to report a case series of four cases of combined infection of mucormycosis with *Aspergillus*. These four patients were elderly with a mean age of 62.5. Three of four patients with a history of COVID-19 had diabetes mellitus, and another patient had hypothyroidism. All four patients were treated with extensive surgical debridement and intravenous liposomal amphotericin B. The mortality rate of these patients was 50% (2/4).

Conclusion: Mixed fungal infections i.e. CAMA during COVID and post-COVID periods may be an emerging disease. These coinfections are rare but severe. Therefore, mucormycosis and Aspergillosis should be considered one of the critical side effects of post-COVID-19.

Keywords: Aspergillosis; Coronavirus disease; Invasive fungal disease; Mucormycosis

Prevalence of Mucormycosis and Aspergillosis among COVID-19 patients, Northeastern Iran

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Background and Aim: Mucormycosis and invasive aspergillosis (IA) are severe and fatal fungal infections. The concurrence of mucormycosis and aspergillosis with other infections such as COVID-19 can disrupt the treatment and control of the infection. In this study, mucormycosis and *Aspergillus rhinosinusitis* were detected using sinus biopsy specimens among COVID-19 patients, northeastern Iran.

Methods: During an eight-month period, in two tertiary referral hospitals, 71 patients with rhinosinusitis (88 specimens) affected to COVID-19 with suspicion of fungal infections were evaluated. The sinus biopsy specimens were examined using mycological procedures and histopathology. The sinus specimens were examined by direct wet mounts with 20% potassium hydroxide (KOH) and cultured on Sabouraud dextrose agar. The cultures were then examined and evaluated to identify.

Results: The specimens showed broad mycelium without septate (non-septate hyphae) indirect experiments of 48 (54.5%) cases and mycelium with septate (septate hyphae) in 12 (13.6%) cases. Positive culture as *Rhizopus* spp. (66.7%) and *Mucor* spp. (33.3%) in 24 (27%) cases. Positive culture for Flavi section (77%) and Nigri section (23%) in 13 (14.8%) cases. Positive histopathology results of mucormycosis in 41 (46.6%) cases and positive histopathology results of aspergillosis in 2 (2.27%) cases. (10 specimens without histopathology examination). Of the 71 patients, 38 (53.5%) showed mucormycosis, 15 (21%) died, and 14 (19.7%) showed *Aspergillus rhinosinusitis* that 2 (10.5%) them died.

Conclusion: Mycological examination results highly correlated with histopathology results in mucormycosis. However, in aspergillosis, it showed a weak correlation. The prevalence of mucormycosis and aspergillosis were noticeably high in Northeastern, Iran. Among the species that cause mucormycosis and aspergillosis, *Rhizopus* spp and Flavi section were considerable respectively.

Keywords: Mucormycosis; Aspergillosis; COVID-19; Infection; Mortality

Study on Invasive Aspergillosis Using Galactomannan Enzyme Immunoassay and Determining Antifungal Drug Susceptibility among Hospitalized Patients with Hematologic Malignancies or Candidates for Organ Transplantation

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Background and Aim: The incidence of invasive aspergillosis (IA) has dramatically increased during the last decade. This infection is associated with high morbidity and mortality, ranging from 30% to 70%, especially in immunocompromised patients. Delay in diagnosis and treatment is usually associated with high mortality rates. This study was aimed to assess the diagnostic value of Galactomannan EIA for early diagnosis of aspergillosis in hospitalized patients with underlying conditions. Also, the antifungal drug susceptibility profiles of causative agents were investigated.

Methods: In this descriptive cross-sectional study, during the period of 18 months, bronchoalveolar lavage (BAL) and 13 biopsies from infected sinuses were obtained from a total of 150 patients suffering from different types of hematologic malignancies. All the samples were subjected to microscopic examination and fungal culture. Also, serum specimens were obtained from all patients (n = 135). 22 serum and 17 BAL specimens were tested for the GM level. Fungal identified were confirmed through the PCR-sequencing of the β -tubulin gene. The susceptibility to amphotericin B, itraconazole, voriconazole, posaconazole, ravuconazole, and caspofungin was evaluated according to the Clinical and Laboratory Standards Institute document M38-A2 (CLSI M38-A2) broth microdilution protocol.

Results: The results showed that the incident rate of IA was 23.33% and 35 patients with IA (12 proven cases and 23 probable cases) were diagnosed according to the European Organization for Research and Treatment of Cancer and Mycoses Study Group criteria. The 35 patients with IA in the current study comprised 19 men (54.29%) and 16 women (45.71%) with the median age of 42 years. AML (31.5%) was documented as the most prevalent risk factor among our subjects with IA and *Aspergillus flavus* (65.7%) was the most prevailing causal agent in this study. Among patients with IA, ague (71%) and cough (60%) were the most common symptoms. In the present study, a sensitivity of 94% and a specificity of 98% was reported for GM ELISA in BAL specimens. Also, a sensitivity of 58% and a specificity of 98% was reported for GM ELISA in serum samples. Among 6 tested antifungal drugs, the lowest minimum inhibitory concentration (MIC) values were observed for

posaconazole and ravuconazole which showed the range of 0.008–0.0062 and 0.031–0.125, respectively.

Conclusion: The current study has demonstrated that determining the value of GM investigation in BAL and serum specimens can be promising in early diagnosis of IA, also molecular identification of the agents causing IA and their antifungal susceptibility patterns are essential issues for the targeted antifungal therapy and outcome improvement of patients with this life-threatening disease.

Keywords: Invasive aspergillosis; Galactomannan; Bronchoalveolar lavage; Serum; Hematological malignancy; Organ transplantation



The Relationship between Biofilm Formation and Mortality in Patients with *Candida tropicalis* Candidemia

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Background and Aim: Biofilm formation by *Candida* species is an influential virulence factor in candidemia pathogenesis. We investigated the relationship between biofilm formation of *Candida tropicalis* (*C. tropicalis*) isolates with the clinical characteristics and mortality outcomes in patients with candidemia.

Methods: Thirty-nine *C. tropicalis* isolates were recovered from patients with candidemia admitted to two university hospitals in Tehran, Iran. Biofilm mass and metabolic activity of *C. tropicalis* biofilms were assessed *in vitro* with two colorimetric methods. The sessile minimum inhibitory concentrations (SMICs) were evaluated *in vitro* by treating preformed biofilms with diluted concentrations of azoles according to CLSI-M27 A3/S4 protocol and metabolic activity quantification. The expression of ERG11, UPC2, MDR1, and CDR1 genes were also evaluated.

Results: All *C. tropicalis* isolates produced biofilm. Respectively, higher (lower than 7days and higher than 7days) mortality rates were found among cases with high metabolic activity (46.7% vs. 13%, $P = 0.03$) and high biofilm mass (31.8% vs. 0, $P = 0.029$). Sessile cells had high resistance to fluconazole, voriconazole, and itraconazole. The azole minimum inhibitory concentrations (MICs) of *C. tropicalis* sessile were significantly greater than the planktonic minimum inhibitory concentrations (PMICs). In fluconazole-treated biofilms, the expression of ERG11 and UPC2 genes was increased.

Conclusion: Our findings highlight the importance of *C. tropicalis* biofilm formation as an important factor in candidemia pathogenesis and clinical management of patients with candidemia.

Keywords: *Candida tropicalis*; Biofilm; Candidemia; Mortality; Azole resistance



Medical parasitology



Prophylactic Effect of Cutaneous Leishmaniasis against COVID-19: A Case-control Field Assessment

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Background and Aim: This study has been carried out in two series. In the first part, we explored the possible relationship between COVID-19 and laboratory-confirmed CL-registered cases with a history of scar compared with healthy volunteer participants.

Methods: This case-control retrospective study was conducted in southeastern Iran with a high anthroponotic cutaneous leishmaniasis (ACL) burden. The Bayesian logistic regression with noninformative Jeffrey's Prior using statistical software R 4.0.2. was performed to analyze data.

Results: Overall, in this model, $n = 1,010$ cases of CL consisting of $n = 479$ male and $n = 531$ female were analyzed for probable infection with SARS-CoV-2. Altogether, in the case group, two men and one woman (0.3%) were afflicted with a mild form of COVID-19 disease, and none of them were hospitalized or died. In contrast, out of $n=2,020$ healthy participants, $n = 57$ (2.9%) contracted laboratory-confirmed COVID-19, including mild (66.7%), hospitalized (26.3%) and critical and death (3.5%, each). There was a strong negative association between CL infection and COVID-19. The burden of COVID-19 in CL-cured participants significantly reduced the morbidity (OR = 0.12, CI: 0.03 - 0.30).

Conclusion: Participants with a history of previous CL scar significantly prevented the incidence of COVID-19 morbidity and mortality. The cross-protection mediated by the CL-cured cases would presumably retard COVID-19 in endemic countries. However, further longitudinal studies are needed to assess the potential profile and duration of this protection offered by CL against COVID-19. The second part of the study has already been designed and ongoing in the endemic areas of Kerman province, with a larger sample size. Hopefully, the preliminary data confirm the previous findings. We anticipate collecting and analyzing the final results in the 13th Congress of Medical Laboratory and Clinic.

Keywords: COVID-19; Cutaneous leishmaniasis; Coinfection; Prophylactic effect

The Leishmaniacidal Effects of *Lucilia sericata* Larval Excretions/Secretions on *Leishmania major*

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Background and Aim: Leishmaniasis is a complex of vector-borne diseases caused by a kinetoplastid protozoa of the genus *Leishmania*. The disease commonly reveals as characteristic skin lesions. Due to the absence of conclusive treatment and drugs without side effects, many studies have focused on natural compounds as promising drugs for cutaneous leishmaniasis. The aim of this study was to investigate the effects of larval excretion/secretion products (ES) of *Lucilia sericata* in crude and fractionated form on *Leishmania major* *in vitro* and *in vivo*.

Methods: In this study, the effects of crude ES and ES fractions of larval *in vitro* and *in vivo* were investigated. *In vitro*, we evaluated the effect of ES on promastigotes by direct counting and amastigotes by random counting of 100 infected macrophages and the number of parasites inside these infected macrophages to measure cell parasitic load as well as cytotoxicity. Also *in vivo*, we evaluated three groups of mice treated with crude ES and ES fractions (ES above 10 kDa and ES below 10 kDa), one control group with standard glucantime drug, one control group with Eucerin (as a drug base), and finally one control group without treatment.

Results: The 50% inhibitory concentration was (IC₅₀) 38.70 µg/ml, 47.61 µg/ml 63.34 µg/ml, and 29.05 µg/ml for crude ES, ES above 10 kDa, ES below 10 kDa, and glucantime (as positive control), respectively. Significant differences were observed between viability percentages of promastigotes treated with crude ES and its fractions compared to negative control (p less than 0.0001). Evaluation of amastigote forms showed that treatment with 300 µg/ml crude ES was more effective than ES above 10 kDa and ES below 10 kDa. Macroscopic assessment of the lesion sizes of Balb/c mice revealed that the reduction of lesion size in the crude ES treatment group had a better effect than the glucantime group as the standard drug compared to the ES above 10 kDa and ES below 10 kDa groups.

Conclusion: The current study showed that ES in both crude and fractionated forms are effective on both intracellular and extracellular forms of *Leishmania major*.

Keywords: *Leishmania major*, Antileishmanial agents, Larval Excretion/Secretion, *Lucilia sericata*.

The role of *Acanthamoeba castellanii* (T4 genotype) Antioxidant Enzymes in Parasite Survival under H₂O₂-induced Oxidative Stress

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Background and Aim: *Acanthamoeba castellanii* (*A. castellanii*) is an important opportunistic parasite. Induction of oxidative stress by the host immune system is one of the most important defense strategies against parasites. Hence, parasites partly deal with oxidative stress by different mechanisms. Identifying resistance mechanisms of *A. castellanii* parasites against oxidative stress is important to achieve a new therapeutic approach. Thus, this study aimed to understand the resistance mechanisms of *A. castellanii*, against oxidative stress.

Methods: Trophozoites of *A. castellanii* were treated with different concentrations of H₂O₂. The half maximal inhibitory concentration (IC₅₀) of H₂O₂ was determined using the MTT assay. The induction of oxidative stress was confirmed by flow cytometer. The activities of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), and glutathione reductase (GR) were determined. The gene expression levels of CAT and SOD were measured by qRT-PCR. Furthermore, 3-amino-1:2:4-triazole (3-AT) and potassium cyanide (KCN) were used as specific inhibitors of CAT and SOD, respectively. Cell cycle assay and the apoptosis were evaluated by flow cytometer.

Results: The activities of SOD, CAT, GR, and GPx, showed an increase in oxidative stress. The cell cycle analysis revealed that most of the cellular population was in G₀ and G₁ phases. The apoptosis increased in oxidative stress conditions. Moreover, the apoptosis significantly increased after the specific inhibition of CAT and SOD under oxidative stress. The gene expression levels of CAT and SOD significantly increased under oxidative stress.

Conclusion: *A. castellanii* can resist the host immune system through various mechanisms, including evoking its antioxidant enzymes. Therefore, by reducing or inhibiting the activity of the parasites antioxidant enzymes such as SOD and CAT, it is possible to cope with *A. castellanii*.

Keywords: *Acanthamoeba*; T4 Genotype; Oxidative stress; Antioxidant; Enzyme activity; Immune system

Toxocara Co-infection and COVID-19: Serosurvey among Hospitalized Patients During the Fifth Wave of Coronavirus Pandemic in Iran

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Background and Aim: Toxocariasis is a worldwide human helminthiasis and is mostly asymptomatic and caused by *Toxocara* species, intestinal nematodes in dogs and cats. The parasite can cause visceral larva migrans syndrome in humans who ingest eggs from contaminated soil or consume of meat of paratenic hosts. However, due to the pandemic of coronavirus 2019, the status of the disease and co-infection with toxocariasis is unknown, with unintended consequences for people with co-infection. Therefore, we decided to evaluate the prevalence of antibodies against this parasite in patients admitted to the hospital due to relatively severe symptoms of COVID-19.

Methods: This cross-sectional study was conducted during the fifth wave of coronavirus on the serum samples of 92 patients with COVID-19 disease who were diagnosed by PCR test and 56 healthy subjects in Alborz province, Iran between March and August 2021. Anti-*Toxocara* antibodies status was determined in all serum samples, using ELISA technique. A self-developed questionnaire was used to measure demographic information.

Results: The frequency of *Toxocara* infection was found to be significantly higher in the COVID-19 patients as compared to healthy control ($n = 14$, 14.9%, $n = 2$, 3.6%; respectively) ($P < 0.05$). There was no significant difference between COVID-19 patients and control group in the variables of age, education, and gender in the studied subjects ($P > 0.05$). This study indicates that a significant association between *Toxocara* seropositivity and the studied people (COVID-19 patients and healthy individual).

Conclusion: Helminthic infection is a common public health in developing country and there have observed a high incidence of parasitic disease. The clinical relationship between parasitic disease and COVID-19 is little mentioned. We presented the first study of *Toxocara* infection and COVID-19 coinfection. This coinfection is likely underreported or overlooked in endemic areas.

Keywords: Toxocariasis; COVID-19; Co-infection; Serodiagnosis; Anti-*Toxocara* antibodies



Medical bacteriology



Anti-biofilm Effect of *Lactobacillus delbrueckii* and *Lactococcus lactis* on Multiple Drug Resistant Isolates from Chronic Wounds in Human Plasma Biofilm Model

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Background and Aim: Chronic wound infections are difficult to treat due to predisposing factors and microbial biofilm formation. One of the new treatment strategies is bacteriotherapy using probiotics. The aim of this study was to investigate the antibiofilm effect of two probiotic bacteria, *Lactobacillus delbrueckii* and *Lactococcus lactis* on isolates from chronic wounds in the human plasma biofilm model (hpBIOM).

Methods: In this cross-sectional experimental study, 82 samples of chronic wound biofilms were examined and bacterial isolates were identified by phenotypic and molecular methods. The antibiotic resistance pattern of isolates was determined by disk diffusion and micro dilution methods. The antibiofilm activity of two probiotics on isolates with multiple drug resistance (MDR) in hpBIOM was investigated

Results: Among 82 chronic wound samples, 47 wounds (57.3%) were monomicrobial and 35 wounds (42.7%) were multi-microbial. The most common isolates were *Staphylococcus aureus* (18.5%), *Escherichia coli* (17.0%) and *Pseudomonas aeruginosa* (14%), respectively. Approximately 60% of the isolates were MDR. *Lactobacillus delbrueckii* had more anti-biofilm activity than *Lactobacillus lactis*. These probiotics did not show a significant synergistic effect against the biofilm of the isolates in hpBIOM.

Conclusion: The remarkable ability of *Lactobacillus delbrueckii* to destroy the biofilm of isolates with multiple resistance suggests it as a suitable candidate for bacteriotherapy.

Keywords: Biofilm; Probiotic; Wound Infection

Evaluation of the Xpert MTB/RIF Test Accuracy for Rapid Diagnosis of Pulmonary Tuberculosis in a Tuberculosis Endemic Area

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Background and Aim: Objective/background: *Mycobacterium tuberculosis* (MTB) is a causative agent of tuberculosis (TB) which remains as an endemic disease in Iran. One of the most significant steps for control of TB is the fast and accurate diagnosis of patients. Examination of sputum smears is the primary method for TB diagnosis, and the culture method remains as the gold standard. Recently, a molecular assay that named Xpert MTB/RIF assay (Xpert), was introduced for rapid diagnosis of TB. The aim of this study was to evaluate the performance of the Xpert assay for diagnosis of TB in Golestan province.

Methods: The TB suspected cases which referred to Tuberculosis Reference laboratory, Gorgan, Iran, from March 2018 to February 2019 were included to this study. The sputum specimens were examined using smear microscopy after Ziehl–Neelsen staining and graded according to the International Union Against Tuberculosis and Lung Disease scale (scanty, 1+, 2+, 3+ and 4+). The Xpert analysis was performed according to the manufactures instructions and the provided semi-quantitative results for positive specimens (very low, low, medium and high) were documented. The specimens were cultured into Lowenstein-Jensen (LJ) culture media after decontamination by Petroff's method. Sensitivity and specificity of Xpert and smear microscopy were calculated using a culture as reference standard.

Results: Of 102 presumptive TB cases, 17 (16.7%) had culture proven TB. The set of 16 (94.1%) culture-confirmed TB cases were equivalently detected using smear microscopy and Xpert. Compared to the culture method, specificity of Xpert and smear microscopy was 100% (85/85) and 97.6% (83/85), respectively. Both tests had a sensitivity of 94.1% (16/17). When the semi-quantitative results of Xpert were compared with smear microscopy, there was 92.3% agreement for specimens with medium/high results, with those that had 1+ to 3+ scales.

Conclusion: The rapid and accurate laboratory diagnosis of TB is one of the most important steps for TB control programs. In this study, the Xpert was effectively used to early and accurate diagnosis of TB in the study area. In the other hand, correlation between the Xpert semi-quantitative results and smear microscopy showed that Xpert could help in evaluation of patient's transmission potential. Although Xpert showed high overall sensitivity and specificity for diagnosis of MTB in sputum samples, its accuracy must be evaluate for other clinical specimens in Iran.

Keywords: *Mycobacterium tuberculosis*; Xpert MTB/RIF; Lowenstein-Jensen; Smear microscopy; Tuberculosis.

Investigation of Alteration in Expression Levels of lncRNAs Regulating NF- κ B Inflammatory Pathway in the Gastric Tissue of the *Helicobacter pylori* Infected and Non-infected Patients with Chronic Gastritis

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Background and Aim: *Helicobacter pylori* (*H. pylori*) is colonized in gastric mucosa and in the process of infection of this bacterium due to prolonged establishment in the stomach can cause inflammation in the gastric mucosa or gastritis, gastric ulcer and gastric cancer. Numerous virulence factors have been identified in *H. pylori* that play an important role in inflammation of the gastric mucosa and stimulate the release of inflammatory cytokines. lncRNAs, which make the bulk of the genome, play an important role in regulating the expression of these cytokines. In order to understand the role of *H. pylori* and its virulence factors in inducing lncRNAs in gastric tissue and altering NF- κ B expression levels following this interaction, the present study attempted to compare chronic gastritis *H. pylori* infected patients and non-infected patients. The genotype of *H. pylori* isolates was determined by PCR method and investigated the presence of virulence factors associated with the inflammatory response in different strains. In this study, for the first time, the effect of *H. pylori* on NF- κ B pathway regulatory lncRNAs was investigated.

Methods: Sixteen patients with chronic gastritis infected with *H. pylori* and 16 non-infected patients participated. After culturing and extracting *Helicobacter pylori* DNA, genotyping of *H. pylori* strains was performed by PCR. In addition, the expression of NF- κ B and negatively regulatory lncRNAs of this pathway lincRNA-p21, MALAT1 and NKILA were examined and compared by real time-PCR in two groups.

Results: The expression levels of MALAT1, lincRNA-p21 and NKILA were decreased in the group of infected *H. pylori* compared to the group of non-infected patients with chronic gastritis. The difference in expression level in MALAT1 and NKILA was significant with the calculation of $P \leq 0.05$. NF- κ B gene expression was 2.4 times higher in infected patients than in the control group. Negative regulatory effect of regulatory lncRNAs on NF- κ B is also observed, so that decreased expression of regulatory lncRNAs is associated with increased expression of NF- κ B in infected patients. The expression of NF- κ B in infected *H. pylori* patients with oip, iceA1 and iceA2 virulence factors does not show a significant difference compared to infected patients without these factors. However, in patients with oipA +, iceA1 / A +, cagA +, vacAs1m1 / m2 genotypes, a significant increase in NF- κ B expression was observed compared to patients with other genotypes.

Conclusion: According to the information and findings of this study, decreased lincRNA-p21, MALAT1 and NKILA expression in the group of *H. pylori* infected gastritis patients compared to the non-infected patient group and also the negative regulatory role of the studied lncRNAs on NF-KB expression. These findings indicate an important role of *H. pylori* in inducing inflammatory responses through the regulatory pathway of lncRNAs. Also, the relationship between virulence factors oipA, iceA on NF-KB expression and lncRNAs expression was studied in this study and no significant relationship was observed. However, in patients with oipA +, iceA1 / A +, cagA +, vacAs1m1 / m2 genotypes, a significant increase in NF-κB expression was seen compared to patients with other genotypes and could indicate the effect of these factors in this process. Considering study on the small population and the influence of other virulence factors in this process, the selection of strains that do not have other virulence factors except the desired factors in this study to measure these relationships in a larger community can bring more meaningful results.

Keywords: Helicobacter pylori; lncRNA; NF-κB; chronic gastritis



Isolation and Characterization of Bacteriophage against *Klebsiella pneumoniae* from Cystic Fibrosis Patient

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Background and Aim: *Klebsiella pneumoniae* (*K. pneumoniae*) is a species of well-known opportunistic human pathogens that are associated with Cystic Fibrosis disease and chronic pulmonary obstruction. Today, it has become one of the problems in their treatment; however, this pathogen is often resistant to multiple drugs. The discovery of novel agents to control *K. pneumoniae* infections is urgent due to the broad multidrug-resistance. A phage with the ability to efficiently and specifically lyse bacteria is considered an alternative to control these pathogens. This study aimed to isolate a lytic bacteriophage with the potential to lyse clinical isolates of *K. pneumoniae*.

Methods: Antimicrobial Susceptibility of clinical *K. pneumoniae* Isolates was determined using disk diffusion assays then Water samples were collected from a hospital waste-water in Tehran. The samples were filtered and mixed with an overnight grown culture of *Klebsiella pneumoniae* in nutrient broth. Phage titration, latent period, and burst size measurements were carried out by the double-layer agar method using the *K. pneumoniae*. Formation of clear zones indicated susceptibility. The isolated phage was characterized by Transmission Electron Microscopy (TEM), Spot test was employed to determine the bacterial host range for the isolated phage using eleven multidrug resistant *K. pneumoniae* isolates from patients with Cystic Fibrosis and Effectiveness of a phage against *K. pneumoniae* biofilms was determined by Minimum Biofilm Eradication Concentration (MBEC) Assay System.

Results: This phage formed clear, circular plaques with constant diameter surrounded by translucent halos that increased in diameter over the course of time. The isolated phage belongs to the Myoviridae family and infected seven of eleven tested multidrug-resistant *Klebsiella pneumoniae* strains. This phage had large burst sizes, efficient rates of adsorption and were stable under different adverse conditions.

Conclusion: The search for potential therapeutic phages against *Klebsiella pneumoniae* has gained momentum because of the increasing prevalence of ESBL-producing and/or carbapenemase-producing *K. pneumoniae* strains associated with human infections. many *Klebsiella* infections are caused by multidrug-resistant strains that generally cannot be controlled using common antibiotics. Therefore, the use of phage therapy to inhibit the growth of *K. pneumoniae* strains has been suggested. As the phage was virulent and specific for *K. pneumoniae*, and was stable and active at different condition that exhibit a number of properties indicative of potential utility in phage therapy cocktails.

Keywords: Bacteriophage; Cystic fibrosis; *Klebsiella pneumoniae*; Phage therapy

Cell Killing Effects of Cytoplasmic Extract and Cell Wall of *Lactobacillus* and *Bifidobacterium* Bacteria on Brain Neuronal Cancer Cell Line

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Background and Aim: Nowadays, the probiotic bacteria such as lactobacilli are known as preventing factors the development of many diseases including cancer. In this study, the cytotoxicity effects of cytoplasmic extract and cell wall of *Lactobacillus* and *Bifidobacterium* bacteria on the neural cell line of the brain was studied.

Methods: In this cross-sectional study, after collecting samples of traditional and pasteurized dairy products in the region of Fars, MRS medium was used for isolation of lactobacilli and bifidobacteria. The isolates were identified by biochemical and molecular approaches after primary confirmation. The cytoplasmic extract and cell wall were produced in MRS broth, and their supernatants were separated. The inhibitory effect of the extracts of the isolates on PC12 and HEK293 cell lines were studied and their effects were evaluated by microscopy and MTT assay. In this study, 15 isolates of *Lactobacillus* and 2 isolates of *Bifidobacterium* were obtained.

Results: The results of anticancer activity showed that the cell wall of the isolates had a significant anticancer effect on PC12 cancer cells ($p < 0.05$). The results showed that *L. acidophilus* cell wall and cytoplasmic extract of *L. ruteri*, *L. acidophilus* and *B. bifidum* in 1 µg/ml concentration had the highest lethal effect (90%) on normal kidney cells (HEK 293). Also, *Lactobacillus ruteri* and *Lactobacillus casei* showed the highest cytotoxic effect in 0.001 µg/ml concentration on PC12 cancer cell (60%) and *L. acidophilus* cell wall had the highest lethal effect (60%) in 1 µg/ml concentration.

Conclusion: The findings of this study showed that the *L. acidophilus* has a lethal effect on brain nerve cancer cells and also the cell wall of *L. acidophilus* was the best structure for killing brain nerve cancer cells.

Keywords: *Lactobacillus*, *Bifidobacterium*; Cell line; cancer; PC12; HEK293

Determining the Frequency of blaNDM-1 and blaIMP Genes in Carbapenemase Producing *Acinetobacter baumannii* Isolates from Burn Wounds in Motahari Hospital in Tehran, Iran

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Background and Aim: *Acinetobacter baumannii* causes a great range of serious nosocomial infections including burn infections. In recent years, *A. baumannii* has shown a rapid progressive resistance to last resort antibiotics such as carbapenems and has been announced as the first priority of the WHO critical pathogens list for research and development of new antibiotics. Carbapenemase enzymes are known as the most frequent mechanism of resistance in *A. baumannii*. blaNDM-1 and blaIMP genes are among these Beta-lactamases which increasingly contribute to carbapenemase production. So, the aim of this study was determining the frequency of blaNDM-1 and blaIMP genes in carbapenemase producing *A. baumannii* isolates from burn wounds in Motahari Hospital in Tehran, Iran.

Methods: In the present study, 42 isolates of *A. baumannii* were obtained from wounds of burn patients in Motahari Hospital in Tehran during 2018, identified by conventional microbiological phenotypic methods and confirmed by PCR for blaOXA-51 gene. Antibiotic susceptibility test (AST) was performed using the Kirby-Bauer method and the CLSI guidelines. A combined disk diffusion test (CDDT) was used to identify metallo- β -lactamases (MBL). The frequency of blaNDM-1 and blaIMP genes were also determined by PCR. Statistical analysis of data was performed using SPSS software.

Results: *A. baumannii* showed resistance to: Ceftazidime (100%), Ciprofloxacin, Amikacin, Imipenem (98.33%), and Gentamicin and Piperacillin-Tazobactam (93.33%). No resistance to Colistin was evident. All 42 isolates of *A. baumannii* produced MBL by CDDT. It was also observed that blaNDM-1 and blaIMP genes are present in (14.29%) and (26.19%) isolates of MBL producing *A. baumannii*.

Conclusion: In this study, we observed that multi drug resistance rate among *A. baumannii* isolates has reached to 98.33%. It was confirmed that carbapenemase play a great role in resistance of *A. baumannii* to antibiotics and among them, enzymes related to blaNDM-1 and blaIMP genes show important and continuously increasing contribution.

Keywords: *Acinetobacter baumannii*; carbapenemase; blaNDM-1; blaIMP; burn



Medical Virology



Designing of DNA Vaccine Based on A Secretory Form of Major Capsid Protein of Human PapillomaVirus Type 18

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Background and Aim: More than 99% of cervical cancers have been associated with human papillomaviruses (HPVs) worldwide. Although commercial HPV vaccines are safe and have been effective in HPV protection against specific HPV types, but they are expensive and not affordable for under developing countries. Therefore, the design of new generation of vaccines like DNA vaccines seems essential. The aim of this study was to construct a recombinant eukaryotic expression plasmid containing L1 of HPV-18, tPA and PADRE genes in pVAX1 vector.

Methods: The L1, tPA and PADRE genes were amplified using Polymerase chain reaction (PCR) method. The PCR products were cloned and the insertion of genes were confirmed by colony PCR, restriction enzymes analysis and sequencing methods. The expression of target gene in HEK-293 cells was confirmed by Indirect immunofluorescence (IF), RT-PCR and western blot assays. We measured the serum total IgG and IgG isotypes in mice immunized by ELISA technique.

Results: Western blot analysis showed a protein band of about 67.5 kDa in supernatant and cell lysate of transfected cells. Four group mice were immunized with two different constructs i.e pVAX-L1, pVAX-tPA-PADRE-L1 and pVAX1 and PBS as controls. The results of immunization in mice showed the levels of antigen-specific antibodies significantly increased by the tPA-PADRE signal sequence

Conclusion: Findings supported that pVAX1-tPA-PADRE-L1 recombinant plasmid have highly immunogenic and is a promising candidate vaccine development against HPV type 18.

Keywords: human papillomaviruses; cervical cancers; DNA vaccines; Polymerase chain reaction; Indirect immunofluorescence

Evaluation of Covid-19 Hospitalized Children in Terms of Clinical Symptoms and Laboratory Tests in Iran

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Background and Aim: The rapid spread of coronavirus disease 2019 (COVID-19) infection has challenged the world to a new crisis. On March 2020, the World Health Organization declared this outbreak a global pandemic and Iran reported its first confirmed COVID-19 cases on February 19, 2020. The range of this infection in children is wide; asymptomatic & symptomatic COVID-19, which are usually characterized by fever, respiratory symptoms, and sometimes gastrointestinal indications, headache, and anosmia. This study describes epidemiological & laboratory findings and aims to help provide a comprehensive understanding of the characteristics of COVID-19.

Methods: This cross-sectional study was conducted on Medical records about epidemiologic data, underlying diseases, symptoms and laboratory tests from March 2020-2021, on 278 hospitalized confirmed COVID-19 pediatrics (up to 18) in several children's hospitals of Tehran, Ahwaz, Isfahan and Bandar Abbas, Khorramabad.

Results: The average age of 278 confirmed COVID-19 children was 5.3 years that 59.4% were boys. Under 40% of cases had an underlying disease, of which the most common conditions were malignancy and diabetes with 10% and 5% respectively. Approximately 40% of our study population were exposed to indicator cases. Fever and cough were the most common symptoms. In our research some laboratory factors were abnormal; the mean initial CRP in the under 1-year-olds was lower than others, high LDH was seen in over 55% of patients. Abnormal AST was found in 37.17%. And Leukocytosis was more common than leukopenia. 3.6% of these hospitalized pediatrics needed oxygen therapy with ventilators, 83.09% had been treated by antibiotics. In this study, the mortality rate was 4.3%.

Conclusion: Most children admitted to our study received antibiotics therapies. Antivirals and steroids were prescribed for a minor number. Oxygen therapy was performed less frequently in our patients. Among the children who died in this study, the majority had underlying diseases.

Keywords: COVID-19; Children; Clinical; laboratory tests; Treatments

Evaluation of Oncolytic Reovirus-Loaded AD-MSCs Efficacy in Therapy of Tumoral C57BL/6 Mice Model

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Background and Aim: Previously, the application of oncolytic reovirus in the treatment of various cancers has been confirmed. To overcome the delivery problems, using cell-based carriers have been suggested. This study was designed to evaluate the efficacy of cancer treatment based on using adipose-derived mesenchymal stem cells (AD-MSC) loaded with oncolytic reovirus in the treatment of tumoral C57BL/ 6 mice model.

Methods: Tumors were established in C57BL/6 mice that inoculated S.C. with TC-1 cell lines in the right flank. The mice were randomly allocated into five groups based on injected elements. Each group was treated systemically with defined elements included reovirus, reovirus-loaded MSCs, MSCs, and PBS through the tail vein. After treatment, tumor size, apoptosis rate in tumoral tissue, CTL-assay as well as cytokine release pattern in isolated splenocytes were measured. Additionally, cell tracing was performed by both ex vivo and in vivo imaging the injected-stained AD-MSC through the mice's bodies. Detection of the reovirus genome in the tumor was performed via Real-Time PCR.

Results: Our study indicated that the reovirus infection effect on AD-MSCs does not have a devastating outcome on the migration potency mainly in MOI 1 and 5. On the other hand, AD-MSCs played an efficient role as a cell carrier to deliver oncolytic reovirus into the tumor site in comparison with systemic administration of oncolytic reovirus alone. Results showed the apoptosis rate directly relies on viral titration and passing time after virus inoculation. Followed by systematic administration, both treatments with reovirus-loaded AD-MSCs and MSCs alone had indicated significant inhibition in tumor progress. Also, treatment with reovirus causes an increase of inflammatory cytokines like IL-6 and IFN- γ secretion in all reovirus-related groups.

Conclusion: The results showed that via using the AD-MSC as a cell carrier for oncolytic reovirus, enhancement in systemic delivery, antitumor activity and tumor regression could be achieved.

Keywords: Cell carrier; Target delivery; Anti-tumor effect; Cytokines; Oncolytic

IL-22 Polymorphism May Have A Protective Role in HBV Infection Outcome

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Background and Aim: Background: Genetic variations can play an important role in the modulation of immune system responses. IL-22 is a recently discovered cytokine of the IL-10 family that can affect the immune response to cancer, inflammation and infectious diseases. Therefore its polymorphisms might influence the HBV infection outcome. Aim: This study aimed to evaluate rs1179251 single nucleotide polymorphism in the IL-22 gene as a host genetic factor and its effect on hepatitis B infection outcome.

Methods: Methods: DNA was extracted from 100 HBV infected patients with cirrhotic / hepatocellular carcinoma (C/HCC), 100 patients with chronic active hepatitis (CAH), 100 asymptomatic healthy carriers (HC) and 58 individuals that their infection was spontaneously cleared (SC). IL-22 rs1179251 single nucleotide polymorphism (SNP) was then genotyped in all cases using PCR-RFLP method.

Results: Results: The results of rs1179251 SNP genotyping demonstrated that G allele in HC (30%) was significantly more frequent than CAH group (17%) ($P = 0.003$). HC group also showed more CG genotype (46%) than CAH group (28%) ($P = 0.006$) but less GG genotype (7%) than SC group (13.8%) ($P = 0.01$).

Conclusion: Conclusion: Our findings collectively suggest that CG and GG genotype or generally G allele of rs1179251 might have a protective role against HBV infection. However how it can be involved in helping the immune system to deactivate the virus remained to be elucidated.

Keywords: IL-22; HBV infection; hepatocellular carcinoma; single nucleotide polymorphism

Evaluation of Blood lncRNA NEAT1 Levels in COVID-19 Patients

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Background and Aim: The coronavirus disease 2019 (COVID-19) caused by severe acute respiratory coronavirus 2 (SARS-CoV-2), was first time detected in Wuhan, China in December 2019, posing a threat to public health with high mortality rates. Long non-coding RNAs play crucial roles as regulators in various biological processes such as transcription, translation, and gene expression regulation. With the virus invasion into the host, innate immune responses are activated as the first line of protection, which contain a set of pattern recognition receptors (PRRs), including RIG-I-like receptors (RLRs) and Toll-like receptors (TLRs) that recognize the RNA viral genome. TLR2 and TLR4 have been shown to be able to induce different lncRNA expressions. Due to the role of lncRNA in innate immune response regulation against viral infections, identification them can be useful in diagnosis and treatment of viral diseases. The aim of this study was to analyze the expression of lncRNA NEAT1 in the blood, as a circulating biomarker for diagnosis of COVID-19 patients' complications.

Methods: The total RNA was extracted from the blood of the 20 COVID-19 patients and 20 healthy control subjects. Participants in this study were 12 males and 8 females in the patient group and the healthy control group were 12 males and 8 females. qPCR was performed to quantify the mRNA level of lncRNA NEAT1 in the blood of patients and healthy groups using the SYBR Green approach.

Results: The relative expression of lncRNA NEAT1 was significantly higher in COVID-19 patients compared healthy control, (fold change= 3.634 ± 1.143 , $p= 0.0029$). The AUC of blood lncRNA NEAT1 for diagnosing COVID-19 compared to controls was 0.7212 (95% CI: 0.6067 to 0.8356, $p= 0.0006$) and the optimal cut-off value was calculated to be >0.1994 (Sensitivity, 71.43%, Specificity 56.67%).

Conclusion: Our findings demonstrated that the mean expression level of lncRNA NEAT1 are significantly increased in the SARS-CoV-2 infected patients compared with healthy group and NEAT1 has good specificity for diagnosis COVID-19 patients.

Keywords: COVID-19; SARS-CoV-2; long non-coding NEAT1

Robust Antimicrobial Photodynamic Therapy with Curcumin-Poly (Lactic-co-Glycolic Acid) Nanoparticles Against COVID-19: A Preliminary in Vitro Study in Vero Cell Line as A Model

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Background and Aim: In this study, the ability of antimicrobial photodynamic therapy (aPDT) as a treatment approach and adjuvant therapy using Curcumin-Poly (Lactic-co-Glycolic Acid) nanoparticles (Cur@PLGA-NPs) to inactivate Coronavirus disease 2019 (COVID-19) in plasma was investigated. Furthermore, to verify whether the quality requirement of aPDT-treated plasma is acceptable, the differences of the levels of clotting factors, total plasma proteins, and anti-A and/or anti-B antibodies titrations in plasma of patient before and after aPDT treatment were investigated.

Methods: Cur@PLGA-NPs was synthesized using Electrospinning process and characterized by different analysis including Scanning Electron Microscope (SEM), Transmission Electron Microscope (TEM), and Fourier Transform Infrared (FTIR) spectroscopy assays. The presence of the SARS-CoV-2 in the plasma samples of patients suspected of having COVID-19 was confirmed by real-time reverse transcription-polymerase chain reaction (RT-PCR) assay. Then, the treated plasma samples with Cur@PLGA-NPs plus blue laser were exposed to Vero cells. Eventually, cell cytotoxicity and apoptotic effects of treated Vero cells were evaluated. Levels of clotting factors including prothrombin time (PT) and activated partial thromboplastin time (APTT), total plasma proteins, and anti-A and/or anti-B antibodies measurements were performed using the coagulometer, method of Bradford, and titration procedure, respectively.

Results: The presence of SARS-CoV-2 was positive in 84.3% of samples. Different concentrations of Cur@PLGA-NPs (3, 5, 7, and 10% wt.), the irradiation times of blue laser (1, 3, and 5 min), and aPDT with the maximum dosed of blue laser light (522.8 J/cm²) plus 10% wt. Cur@PLGA-NPs had no cytotoxicity. Although there were significant cell degradation and apoptotic effects in treated Vero cells with treated plasma using 10% wt. Cur@PLGA-NPs, and a blue laser at an energy density of 522.8 J/cm², no visible changes in cells and apoptosis were observed following aPDT. Total plasma protein content, PT, APTT, and anti-A and/or anti-B antibodies titers showed no significant changes ($P > 0.05$ for all comparisons) in treated plasma as compared to untreated plasma.

Conclusion: aPDT exhibited in vitro anti-COVID-19 activities in the treated plasma containing SARS-CoV-2 without Vero cell apoptosis and any adverse effects on plasma quality in aPDT-exposed plasma.



Keywords: Antimicrobial photodynamic therapy; Coronavirus; COVID-19; Curcumin; PLGA; SARS-CoV-2

Biochemistry



Anti-Inflammatory Effects of *Securigera securidaca* L. Extract on Bile Duct Ligation in Rats

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Background and Aim: Cholestasis and its associated oxidative stress can lead to liver injury and fibrosis. Some medicinal plants and antioxidants have potential protective effects against liver fibrosis. *Securigera securidaca* (SS), from the Fabaceae family, is rich in flavonoids and hence, seems to have high antioxidant effects. This study aimed at evaluating the effects of SS seed extract on cholestatic liver injury induced by bile duct ligation (BDL) in rats.

Methods: Forty eight adult male Wistar rats (200–250 grams) were allocated to six groups. Rats in four groups underwent BDL at the common bile duct and were treated for 21 days with either oral distilled water as vehicle, vitamin C, 100 mg/kg SS seed extract, or 200 mg/kg SS seed extract. Rats in the fifth group underwent abdominal incision without BDL and were treated with distilled water and rats in the sixth group were healthy and received nothing. Finally, rats were sacrificed, blood samples were analyzed through biochemical methods, liver tissues were histologically assessed, and the expression of the TGF β -1, iNOS, caspase-3, and α -SMA genes in the liver were assessed through quantitative real-time polymerase chain reaction.

Results: BDL significantly increased and SS seed extract significantly decreased the serum levels of bilirubin and liver function enzymes. Moreover, SS seed extract suppressed the expression of the TGF β -1, iNOS, caspase-3, and α -SMA genes, reduced the levels of nitric oxide, malondialdehyde, and protein carbonyl, and increased the levels of glutathione, total antioxidant capacity, as well as SOD and catalase enzyme activity in serum and liver. It also had significant positive effects on liver morphology and parenchyma structure, particularly at 100 mg/kg dose.

Conclusion: SS seed extract has protective antioxidant effects against BDL-induced cholestatic liver injury as determined by reduction in histopathological changes, fibrosis, inflammation, oxidative stress, and some biochemical indices in serum.

Keywords: Antioxidants; Liver fibrosis; Herbal medicine; Oxidative stress; *Securigera securidaca*

Chlorogenic Acid Improves Anti-Lipogenic Activity of Metformin by Positive Regulating of AMPK Signaling in HepG2 Cells

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Background and Aim: Metformin improves lipid profile, however, combination therapy is developing to increase the effectiveness and reduce the deleterious effects of metformin. Chlorogenic acid (CGA) has exhibited lipid-lowering effects. This study aimed to investigate the combined effect of metformin and CGA on lipid accumulation, as well as to elucidate the engaged mechanism in HepG2 cells.

Methods: To find the non-lethal doses of metformin and CGA, MTT assay was performed. High Glucose (HG) at 33 mM was used to induce lipogenesis in HepG2 cells. Following treatment with different concentrations of metformin and CGA, total lipid content (Oil Red O-staining), triglyceride level, the genes expression of SREBP-1c and FAS, and phosphorylation of AMPK and ACC were measured.

Results: Both Metformin and CGA decreased HG-induced lipid accumulation individually, by decreasing total lipid content and triglyceride level. The lowest effective doses of metformin and CGA were 0.25 mM and 5μM, respectively, which significantly reduced SREBP-1c and FAS genes expression. The combination of these concentrations reinforced these effects. The phosphorylation of AMPK and ACC were more increased by metformin in combination with CGA than both individually.

Conclusion: Our findings suggest that CGA synergistically enhances metformin lipid reducing action via the regulating of involved factors in fatty acid synthesis. Therefore, co-administration of metformin with CGA may have further medical value in treating lipid metabolism disorders.

Keywords: AMP-Activated Protein Kinases; Combined Therapy; Chlorogenic Acid; Lipid metabolism; Metformin

Evaluation of the Relationship Between Biochemical Parameters and COVID 19 Disease in Patients Admitted to Hospitals

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Background and Aim: Coronavirus family is a group of familiar diseases that human beings have encountered; The coronavirus epidemic has become a global emergency in the 21st century. Which is evolving rapidly and continues to put pressure on healthcare systems. The virus is a phenomenon that has caused a crisis in the world due to its rapid spread and unstoppable nature. In response to the disease, several national governments have imposed quarantine restrictions to reduce the rate of infection. Clinical studies have shown that the most common manifestations of Covid-19 are fever, fatigue, and dry cough. Although laboratory changes have been widely reported in patients with mild to severe Covid-19, little information appears to be available on laboratory findings. The incidence of abnormal liver biochemical tests, including (AST), (ALT), (ALP), (GGT), and albumin (ALB), has been calculated to investigate the relationship between biochemical tests and the severity and prognosis of COVID 19 patients. Case and laboratory monitoring is based on the ongoing availability of diagnostic tests for all segments of society. Some laboratory markers may predict the prognosis of COVID 19. Therefore, the aim of this study was to investigate the relationship between biochemical parameters and COVID 19 disease in hospitalized patients.

Methods: The present study is an applied-comparative study that was performed to investigate the relationship between biochemical parameters and covid 19 disease in hospitalized patients from the beginning of March 2017 to the end of May 1400. Data collection tools in this study were patient records and the use of HIS hospitals in which all patient information is recorded. Finally, the data were used by SPSS software version 24 and descriptive and analytical methods were used to analyze the data. To compare the means, t-test or Anova test was used.

Results: Diagnostic tests are typically characterized by simple performance criteria such as sensitivity and specificity. Measurement of biochemical parameters may help to assess the prognosis of Covid 19. These measures are widely known. In addition, according to the findings of studies, some laboratory tests have a very high accuracy in predicting the diagnostic cases of Covid 19. However, estimating the sensitivity and characteristics of these tests are always subject to various biases. With the study, the use of biochemical tests and parameters can be used on the one hand to diagnose and to evaluate the treatment process. A number of different possible problems are also described with reference to diagnostic texts. These problems can be effective in evaluating and designing diagnostic tests and selecting appropriate measures for the effectiveness of the tests. Also, in the present study, unlike some studies, there was no significant difference between biochemical

parameters and gender relationship of patients. Regarding the relationship between biochemical parameters and different age groups of patients, in the age group of young and adolescents, there are statistically significant results between biochemical parameters and age of patients.

Conclusion: In this study, patients with Covid-19 disease were diagnosed by RT-PCR or CT scan of the lungs. On the first day of hospitalization, they were evaluated by medical diagnostic tests. The levels of AST, CRP, LDH, ALT, Urea, CK, CK-MB and LDH in their serum were examined. These parameters are used in particular in one of the most widely used experiments in laboratories. Based on the findings, some medical diagnostic tests, including ALT, CRP, LDH, and urea, are highly accurate in diagnosing Covid-19 for prognosis associated with RT-PCR-positive patients, respectively. Some laboratory markers may have a prognosis of COVID-19. To predict. Findings are usually associated with a worse prognosis, including elevated levels of D-dimer, protein C (CRP), and high-sensitivity LDH. Affect the distribution of the age category of positive results of Covid 19 and confirmed cases. Due to the significant differences in laboratory parameters in the results of coronavirus testing, it can be expected to be modeled or predicted based on routine laboratory tests.

Keywords: Covid 19; biochemical parameters; Hospital; provinces

AMLS
Archives of Medical Laboratory Sciences

Recombinant Subunits of SARS-CoV-2 Spike Protein as Vaccine Candidates to Elicit Neutralizing Antibodies

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Background and Aim: The spike protein has been reported as one of the most critical targets for vaccine design strategies against the SARS-CoV-2 infection. Hence, we have designed, produced and evaluated the potential use of recombinant proteins derived from spike protein as vaccine candidates capable of neutralizing SARS-CoV-2 virus.

Methods: In silico tools were used to design spike-based subunit recombinant proteins (RBD (P1), fusion peptide (P2), and S1/S2 cleavage site (P3). These proteins were checked for their ability to be identified by the anti-SARS-CoV-2 antibodies by exposing them to COVID-19 serum samples. The proteins were also injected into mice and rabbits. the antibody titers were measured for 390 days to assess their neutralization efficiency.

Results: The antibodies that existed in the serum of COVID-19 patients were identified by designed proteins. The anti-spike antibody titer were increased in the animals injected with recombinant proteins. The VNT results revealed that the produced antibodies could neutralize the cultured live virus.

Conclusion: Subunit vaccines could also be considered as robust tools for effective vaccination against COVID-19. Using a combination of in silico, in vitro, and in vivo experiments, it was shown that the injection of spike-based recombinant proteins could stimulate long-lasting and neutralizing antibody responses.

Keywords: SARS-CoV-2; spike Protein; vaccine; neutralizing antibody; in silico



Medical Genetics



A Comparison of Thioredoxin 1 Gene Expression in Schizophrenia Patients before and after Treatment with Risperidone

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Background and Aim: Schizophrenia is a mental disorder characterized by distortions in thinking, perception, emotions, language, self-sense, and behaviors. Recent research suggests that Reactive Oxygen Species (ROS) are involved in the pathophysiology of schizophrenia. Studies have also shown the increased plasma and serum levels of the Trx1 molecule in schizophrenia patients. In the present study, the researchers compared the expression levels of Trx1 mRNA in peripheral blood leukocytes of Iranian schizophrenia patients compared to healthy controls.

Methods: First-episode patients (n=35) who met DSM-IV criteria for schizophrenia were recruited from patients referred to psychiatrists in the city of Ilam and Farabi Hospital in Kermanshah. Healthy people were also selected by recruiting people who, according to a psychiatrist, did not have any mental illness. Diagnoses were made for each patient by two independent experienced psychiatrists and confirmed by the Structured Clinical Interview for DSM-IV (SCID). Patients were treated with risperidone for three months and then compared with thirty- five healthy volunteers. Patients were sampled before and after treatment and then by RNA Extraction and DNA synthesis, Trx1 gene expression was performed by real-time PCR method.

Results: Comparison of Trx1 gene expression in PBMCs of schizophrenic patients before and after treatment with the control group showed that the expression of the Trx1 gene of the “before” treatment group was significantly increased compared with that of the control group ($P= 0.0007$). Also, Trx1 gene expression in PBMCs of “before” and “after” groups showed that Trx1 gene expression of the “after” group was significantly decreased compared to the “before” group ($P= 0.014$). These results showed that the mean of positive, negative, and general psychopathology was reduced significantly in schizophrenic patients before and after treatment in all three cases (P

Conclusion: the expression of TRX in PBMCs of schizophrenic patients decreased after risperidone treatment. This reduction of expression was statistically significant and indicates the possible effect of risperidone on the expression of the TRX gene in PBMCs of these patients and decreased gene expression is associated with reduced symptoms. Confirmation of the achievement of this study requires further research.

Keywords: Schizophrenia; thioredoxin; risperidone

Design of a Diagnostic Test to Combine Direct and Indirect Prenatal Diagnosis of Common Types of Hb Bart's Hydrops Fetalis

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Background and Aim: Alpha thalassemia is one of the most common hemoglobin disorders worldwide. Deletion or inactivation of four alpha-globin genes leads to a severe form of the disease called Hb Bart's hydrops fetalis. Depending on the outcome of the disease for both mother and fetus, prenatal diagnosis is offered to couples at risk of having a fetus affected by this disease. Gap-PCR is currently used to detect common deletions, and linkage analysis using STR markers helps to confirm the results. Here a diagnostic kit is suggested to combine direct and indirect testing.

Methods: A literature review was performed to find breakpoints of the most common cis deletions in Iran, as well as STR markers linked to alpha-globin genes cluster. Sanger sequencing was performed to confirm breakpoint sequences. Primers were designed using Primer3, Gene Runner, and Primer Blast for a fluorescent multiplex PCR

Results: Primers were designed to specifically amplify undeleted and deleted gene sequences with a PCR product length range of 150-450 bp. A multiplex PCR was designed to combine mutation detection of MED and 20.5 deletions as well as STR markers analysis.

Conclusion: Here, for the first time, a method for combining direct and indirect prenatal diagnosis of alpha thalassemia is presented. In addition to prenatal diagnosis, this method can be used in preimplantation genetic diagnosis.

Keywords: Hb Bart's hydrops fetalis; Gap-PCR; diagnostic kit; STR marker

The Study of Relationship Between Epstein - Barr virus and Breast Cancer in

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Background and Aim: Breast cancer is one of the most common cancer in women. While the etiology of breast tumor is not completely understood, exposure to Epstein-Barr virus (EBV) is proposed as a risk factor for breast malignancy. Educations have stated since 1995 that EBV is complicated in the expansion of breast carcinoma. The purpose of this education was to assess the presence of EBV in patients with breast cancer in East Azerbaijan.

Methods: This study was achieved using 40 paraffin-embedded tumor tissues and 40 tumor- free breast tissues from women with breast cancer in East Azerbaijan province. After extraction of the DNA using salting-out method and the amplification of housekeeping gene beta- actin, all samples were examined for EBV DNA using PCR (polymerase chain reaction) method. Data were analyzed with chi-square test using SPSS 16software.

Results: EBV was discovered by PCR in 20 out of 40 (50%) cases of breast cancer samples and 5 out of 40 (12.5%) control samples. The chi-square statistics for the analysis of EBV infection in tumor and normal samples was 0.000 indicating a significant relationship between breast cancer and EBV infection in East Azerbaijan.

Conclusion: The attendance of EBV gene in a significant subset of women with breast malignancy in East Azerbaijan shows EBV can be one of the reasons for breast malignancy, but more studies are needed to determine the association among the virus and breast cancer.

Keywords: Breast Malignancy: Epstein- Barr virus- PCR



Physiology and Pharmacology



COVID-19 Pandemic and Kidney Manifestations: Mechanisms, Pharmacological and Nutritional Strategies

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Background and Aim: Since 2019, the novel coronavirus disease 2019 (COVID-19), has been paid much attention of scientists and researchers where it has a high global mortality and morbidity rate among older people suffering from chronic diseases, such as chronic kidney diseases (CKD). It is pivotal to clarify molecular mechanisms by which COVID-19 affects the kidneys and accordingly develop appropriate nutritional regimen and pharmacological strategies.

Methods: "Blank"

Results: Theoretically, kidney infections by SARS-CoV-2 can be caused in a variety of ways. When trapped in glomeruli, SARS-CoV-2 accumulate and multiply, and may cause direct damage to the host tissue. In this study, we evaluated the most important evidence regarding causality, potential dietary supplements, therapeutic options, and management of COVID-19 infection in vulnerable individuals and CKD patients. To date, there is no effective and effective drug to treat COVID-19-induced kidney dysfunction, and current treatments are yet limited to anti-inflammatory (e.g. ibuprofen) and anti-viral medications (e.g. Remdesivir, and Chloroquine/Hydroxychloroquine) that may increase the chance of treatment. Supportive treatments such as nutritional supplements and vitamins could show protective effects by down-regulating inflammatory cytokines such as TNF, IL-6, NF- κ B, IL-2, IL-7, IL-10, GSCF, IP10, MCP1, and MIP1A. Taken together, current evidence suggests the therapeutic potential of supportive treatments such as nutritional supplements and vitamins against COVID-19 in patients with kidney diseases. Supplementation of Vitamin A, D, E, and several trace elements including zinc, selenium, copper, magnesium, flavonoid quercetin, and probiotics may be critical for both prevention and treatment of viral infections including COVID-19.

Conclusion: In conclusion, our knowledge about renal failure in COVID-19 patients is very limited, and this review improves our ability to introduce novel approaches for future clinical trials for this contiguous disease. However, supplementation with selective micronutrients may be beneficial especially for vulnerable people such as patients with kidney diseases.

Keywords: COVID-19; Renal failure; Nutritional supplements; Trace elements

Efficacy and Safety of Novel Herbal Tablets in COVID-19 Patients in Hospital Stay Days, ICU Admission and Mortality Rate There of: An Open-Label, Single-Blind Randomized Clinical Trial

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Background and Aim: Severe Acute Respiratory Syndrome Coronavirus2 (SARS-CoV-2), the super-spreading virus, has claimed hundreds of thousands of lives worldwide. This study aimed to evaluate the effectiveness of the novel suggested herbal compound, formulated as compressed tablets, in reducing the length of hospital stay (LoS), intensive care unit (ICU) admission, and mortality in confirmed COVID-19 cases.

Methods: Following an open-label, single-blind randomized clinical trial design, a total of 200 patients aged 18-65 admitted to Imam Reza hospital in Tabriz, northwest of Iran, were randomized to intervention and control groups in a 1:1 ratio, i.e., 100 subjects in each group. The former received standard treatment along with the compressed herbal tablets, and the latter only received the standard treatment. Adverse reactions incidence within 180 days after the beginning of the intervention was set as the primary safety endpoint. The most important and active ingredients of the tablets were Terminalia chebula, Glycyrrhiza glabra, Anacyclus pyrethrum, Senna alexandrina, Ferrula asafoetida, Pistacia lentiscus, Zizyphus jujuba, Crocus sativus, Echinacea angustifolia, and Hyssopus officinalis. This trial is registered at the Iranian Registry of Clinical Trials (code: IRCT20200522047545N1).

Results: Those in the intervention arm had significantly lower rates of LoS (7.38 vs. 9.45, $P = 0.030$), ICU admission (6 out of 100 vs. 32 out of 100, $P=0.000$), and mortality (1 vs. 19 out of 100, $P=0.000$).

Conclusion: Our observations suggest that adequate improvement is provided by the prepared herbal compound along with substantial savings in hospitalization hoteling costs. While further multi-center studies with a larger sample size are needed to extend our knowledge regarding the effect of this new option, these novel clinical data may well provide a new alternative for the management of COVID-19 disease.

Keywords: COVID-19 Disease; SARS-CoV2; COVID-19 Treatment; COVID-19 Pneumonia; Randomized Clinical Trial; Pharmacognosy

The Evaluation of Physio Pathological and Immunological Exercise Effects During Pandemic and Pathogenicity on Coronavirus COVID-19 (with Considerations and Instructions of Doing Related Physical Activity)

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Background and Aim: Corona is a family of viruses that causes some diseases such as cold, Severe Acute Respiratory Syndrome and Middle East respiratory syndrome. The virus is known as acute coronary syndrome 2. Some Corona challenges are included increased sedentary lifestyle, obesity, and increased inflammatory factors and related diseases. In conducting this research, the latest articles of prestigious domestic and foreign journals that indexed in the databases of PubMed, Google Scholar and science direct, etc. have been used. There are various hypotheses about the intensity of exercise and the amount of infection. Although moderate-intensity exercise increases immune function and reduces mortality, while high exercise intensity and stressful exercise has the opposite effect. High exercise intensity and prolonged exercise when you are exposed to viral infections can suppress immune parameters, while moderate-intensity exercise reduces inflammation and improves the immune response to respiratory viral infections. Therefore, due to the fact that exercise has an effect on the strength and anti-inflammatory mechanism in the immune system, it is an important factor to cause fewer complications for patients, so it can be concluded that exercise can strengthen the immune system and reduce inflammation and make favorable physiological and immunological conditions to help athletes overcome the virus.

Methods: In conducting this research, the latest articles of prestigious domestic and foreign journals that indexed in the databases of PubMed, Google Scholar and science direct, etc. have been used

Results: Therefore, due to the fact that exercise has an effect on the strength and anti-inflammatory mechanism in the immune system, it is an important factor to cause fewer complications for patients, so it can be concluded that exercise can strengthen the immune system and reduce inflammation and make favorable physiological and immunological conditions to help athletes overcome the virus.

Conclusion: There are various hypotheses about the intensity of exercise and the amount of infection. Although moderate-intensity exercise increases immune function and reduces mortality, while high exercise intensity and stressful exercise has the opposite effect. High exercise intensity and prolonged exercise when you are exposed to viral infections can suppress immune parameters, while moderate-intensity exercise reduces inflammation and improves the immune response to respiratory viral infections.

Keywords: COVID-19; physiopathology; immunology; exercise; pandemic



Toxicology



COVID 19 and Acute Poisoning Patients in a Referral Poisoning Center

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Background and Aim: The Covid 19 pandemic, spread throughout the world, affecting all hospital wards, including the toxicological emergencies ward. In this study, we evaluated the demographic and clinical manifestations of patients with acute poisoning concomitant Covid 19 disease

Methods: This cross-sectional study was performed in two referral poisoning hospitals, Khorshid and Al-Zahra hospitals affiliated to Isfahan University of Medical Sciences, Isfahan, Iran. The records of patients over 20 years of age who were admitted to the toxicological ward from February 2020 until August 2021, with diagnosis of poisoning and Covid 19 disease were evaluated. Demographic and clinical information was recorded in the data collection form and analysed by SPSS 20 software. P values less than 0.05 were considered statistically significant

Results: During this period, the number of patients admitted to the poisoning emergency ward was 2793, of which 48 patients were diagnosed with both poisoning and Covid 19 disease. 77% of patients were male, self-employed (58%) with no history of underlying disease (68%). In other patients, the most common underlying diseases were cardiovascular disease (16%) and kidney disease (8%). The opioids poisoning was the cause of hospitalization in 54% of patients, of which methadone poisoning (41%) was more common than other opioids. 64% of patients received naloxone. The type of poisoning in most patients was abuse (22%) and suicide (30%). 75% of patients had a decrease in the level of consciousness upon arrival. Arterial blood oxygen saturation was abnormal in 45% of patients. Patients were treated for poisoning as well as Covid 19. The majority of patients survived without complications (81%). Mortality was 12% and was observed in poisoning with paraquat, aluminium phosphide and opioids. Complications and mortality were more common in patients with a history of liver and kidney disease

Conclusion: Decreased level of consciousness and decreased arterial O₂ saturation, are seen in poisoned patients, especially opioid intoxication, but Covid 19 should be considered also particularly when the patient does not respond to naloxone. In other poisonings during the Covid epidemic, this disease should be considered along with poisoning. Referral of patients with a history of pulmonary disease to the infectious and pulmonary wards and non-transfer of this group of patients to the toxicological emergency ward may be the reason for the lower number of patients with a history of lung disease among this research population



Keywords: COVID 19; Poisoning; Opioid-poisoned



The Effect of Ferulic Acid Loaded on Cyclodextrin Nanosponges on Decreasing Parquat Induced Necrosis in MRC-5 Cell Line Lung

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Background and Aim: Ferulic acid (FA) is a naturally-occurring potent antioxidant and free radical scavenger. The aim of this study was to examine the effects of FA encapsulated with cyclodextrin nano-sponges (CD-NSs) in MRC-5 cell lines when exposed to paraquat (PQ).

Methods: We obtained IC₅₀ for PQ toxicity. FA was encapsulated into the CD-NSs in the proportion of 1:4 (FA: NS). Diphenyl carbonate was used as a cross-linker in different proportions with CD. Characterization of obtained CD-NSs was performed using scanning electron microscopy, X-ray diffraction (XRD), differential scanning calorimetry (DSC), and Fourier transform infrared (FTIR) analysis.

Results: Our results revealed that the solubility of encapsulated FA was increased up to fifteen fold compared with pure FA in the proportion of 1:4. The results of FTIR, XRD, and DSC confirmed the interaction of FA with NSs. The cytotoxicity of encapsulated FA against MRC-5 cell lines was investigated using different concentrations of FA in 24, 48, and 72 hrs when exposure to PQ. The cytotoxicity assay indicated that FA treatment increases viability and prevention of apoptosis of lung cells against PQ toxicity. The IC₅₀ value of encapsulated FA (250 ppm) was decreased threefold when compared with pure FA (750 ppm) against PQ toxicity in vitro.

Conclusion: In general, CD-NSs were found to be a suitable delivery system for poorly soluble bioactive such as FA and could be effective in PQ induced lung toxicity.

Keywords: Nano encapsulation; Nano sponges; cyclodextrin; ferulic acid; Paraquat

The Role of NAC and Some Other Clinical Antidotes in The Treatment of Patients with COVID-19

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Background and Aim: The novel coronavirus disease (COVID-19) is known to be caused by the SARS-Cov-2 and cause multiple systems of organ dysfunction associated with significant morbidity and mortality. Therefore, the present study aimed to review the effect of NAC and some other clinical antidotes in the treatment of patients with COVID-19.

Methods: We systematically searched and retrieved the relevant articles using keywords on the online databases, including Embase, Scopus, PubMed, Web of Science, and Cochrane databases up to December 1st, 2021. The retrieved records underwent a title/abstract and full-text screening process, and the eligible papers were identified.

Results: Antiviral and anti-cytokine drugs have been shown to be inadequate or ineffective in stopping the progression of the disease. The relatively low efficacy of anti-cytokine drugs is explained by the fact that they act on one or a several of the dozens of cytokines involved, and because other mediators of inflammation such as reactive oxygen and nitrogen species are not targeted. When overproduced, the reactive species cause extensive cell and tissue damage. The only drug known to inhibit the overproduction of reactive species and cytokines is methylene blue. In this regard, the effectiveness of methylene blue along with other antioxidants has been shown to prevent excessive inflammatory responses in COVID-19 patients. On the other hand, N-acetylcysteine (NAC) has antioxidant, anti-inflammatory, and immune-modulating effects and has been shown to be beneficial in the treatment and prevention of this virus. Also, given that the majority of these patients suffer from hypoxic respiratory failure and require oxygen supplementation in the hospital, hyperbaric oxygen therapy (HBOT) appears to can be an alternative treatment being considered. Finally, there appears to be a significant association between vitamin K and thiamine deficiency with the severity of COVID-19. These vitamins play an important role in the coagulation system.

Conclusion: High-dose oral NAC (1200 mg, twice daily) can improve adaptive immunity by increasing lymphocyte glutathione levels and regulating neutrophil function during COVID-19 development. Also HBOT can increase the circulation and delivery of oxygen under high pressure, making the tissue uptake more efficient and improving hypoxia in patients with COVID-19. Overall, a potential mechanism for associating COVID-19 with coagulopathy has been established, and vitamin K and thiamine can play a regulatory role in coagulation in relation to the etiology of the disease. Therefore, it can be used as a supplement or treatment to improve the outcomes of COVID19.

Keywords: N-acetylcysteine; Hyperbaric oxygen therapy; Vitamin K; Thiamine; methylene Blue



Posters Presentation



Hematology



A New Approach on Isolation and Short Time Culture of Cord Blood-Derived Mesenchymal Stem Cells and Differentiation to Neural Stem Cells

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Background and Aim: Mesenchymal stem cells (MSCs) are adult stem cells initially isolated from bone marrow. However, subsequent research has shown that other adult tissues also contain MSCs. These cells actively proliferate, giving rise to new cells in some tissues but quiescent in others. MSCs can differentiate into multiple cell types, including adipocytes, chondrocytes, osteocytes, cardiomyocytes, and neural lineages. Isolation of MSCs from umbilical cord blood (UCB) from full-term deliveries is a laborious, time-consuming process that results in a low yield of cells.

Methods: We evaluated isolated MSCs from UCB with higher efficacy and relatively short period culture and differentiated them into neural cells. UCB samples were collected from 12 full-term natural delivery cases. Immunophenotyping analysis performed on UCB-MSC of passage 3. After reaching 70% confluence and using trypsin-EDTA, MSCs, of course, three were used to differentiate neural cells. The cells were evaluated by flow cytometry.

Results: The average volume of UCB in 12 samples was 67.2 ± 14.2 ml. The mean MNCs isolate count was $58.2 \pm 10.7 \times 10^6$, and the average viability was $90 \pm 3\%$. As expected, the cells showed MSCs positive markers: CD73, CD44, CD29, CD106, CD105, and CD90. The cells were also negative for hematopoietic stem cells surface markers: HLA-DR, CD34, CD45, and CD14. More than 70% of MSCs differentiated to neural cells. The control cell did not show any features of neural cells and remained un-stained in immunohistochemistry staining by neural cells specific dyes.

Conclusion: The results were confirmed by observation of altering morphology to neural cells and expression of NSE and β -tubulin as neural cells markers by ICC method. Approximately 70% of cells showed these markers.

Keywords: mesenchymal stem cells; umbilical cord blood; neural cells; flow cytometry

A Report of Fanconi Anemia Findings from Northeast of Iran

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Background and Aim: Fanconi anemia (FA) is an autosomal recessive and rarely X-linked genetic disorder. FA manifestations include pancytopenia, hyperpigmentation, and skeletal complications. Mutation in the DNA repair regulatory genes is associated with the development of this disease. Although different methods are used to diagnose this disease, the examination of chromosomal fractures when chromosomes are exposed to cross-linking agents is the common method.

Methods: In this study, we evaluated 312 suspected FA patients, who were referred to the laboratory of Ghaem Hospital for diagnosis. Mitomycin C (MMC) method was used for evaluated positive FA subjects

Results: Among positive FA patients, 48 patients (57.1%) were male and 36 patients (42.9%) were female. Thumb abnormality was reported in 43.2% of FA and was the most common congenital abnormality.

Conclusion: Since the different frequency of clinical symptoms in eastern Iran compared to the north of the country, more studies need to evaluate various factor can affect clinical manifestation incidence.

Keywords: Fanconi anemia; Mitomycin C; chromosomal breakages; congenital abnormality

Acute Hemolytic Transfusion Reaction Due to Anti-c Rhesus Antibody in a Patient with Subdural Hematoma: A Case Report Emphasizing the Shortcoming of Spin Cross-Match

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Background and Aim: The Rh blood group system is a complex blood group that includes different antigen specificities such as c antigen. The anti-c antibody is associated with both acute and delayed hemolytic transfusion reactions as well as hemolytic disease of the newborn (HDN). Rh mediated hemolytic transfusion reactions (HTR) are mostly immunoglobulin G (IgG) mediated and results in extravascular hemolysis and delayed HTR (DHTR). However, we are presenting a case of acute intravascular hemolytic transfusion reaction due to anti-c in a patient with acute subdural hematoma.

Methods: A 77-year-old woman was referred to our hospital with a loss of conscious and left-sided hemiparesis. After an emergency MRI, she was diagnosed with Acute Subdural hematoma, and an emergency craniotomy was performed. Since Acute Subdural hematoma is a neurosurgery emergency, laboratory technician performed an Immediate-spincrossmatched for blood bag to preserve time. During the transfusion of the first packed cell, the patient developed severe hypotension and tachycardia. Thus, the transfusion was stopped.

Results: Laboratory results raised the suspicion of an Acute Intravascular Hemolysis. Antibody identification revealed that the patient had an irregular blood phenotype (C2+/c-/E-/e3+/K-), and the presence of alloantigen-c Rh antibody confirmed the suspicion of HTR.

Conclusion: In patients with multi-transfusion history and pregnant women, pre-transfusion screening of irregular antibodies must be performed. The immediate spincrossmatch must be prevented in patients with a history of multi-transfusions, even in emergency situations.

Keywords: Antibody; Hemolytic; Rhesus; Transfusion; Transfusion reaction

Altered Peripheral Blood Mononuclear Cells Cytokines in Children with Autism Spectrum Disorder after Hepatitis B Virus Vaccine Booster Administrations

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Background and Aim: Deviation in immune response and dysregulation in cytokine profiles have been reported in subjects with autism spectrum disorder (ASD). The relative quantity of cytokines was determined in the peripheral blood mononuclear cells (PBMC) of ASD children after primary vaccination against Hepatitis B virus (HBV) and after vaccine booster challenge.

Methods: PBMC samples were prepared before and after booster injections from 16 autistic children in comparison with 4 healthy subjects. Total RNA were extracted and the cDNA were synthesized. The relative quantities of transcripts for cytokines were assessed.

Results: The mean age of participants was 7.6 ± 2.5 . Both groups responded similarly to the booster vaccine. Before booster injection, levels of IFN- γ were somehow similar between both groups. The relative mean values for IL-4, IL-10 and IL-12 in ASD were higher; however, TGF- β relative mean levels were significantly lower than that of controls. One month after injection, in ASD, the relative mean of IL-4 up regulated slightly. IL-10 was decreased to control group values. TGF- β value was increased in both groups with no major difference and the levels of expression for IL-12 and IFN- γ were significantly increased in ASD patients. IL-17 mRNA was not detected in PBMCs from ASD and control groups. Before booster, Th-2 type cytokines were significantly higher in ASD patients whereas, after booster injection the Th-1type significantly increased in comparison with control subjects.

Conclusion: These results indicated a potential role of immune dysfunction in ASD which would assist discovering of novel possible therapeutics for this condition.

Keywords: Autism spectrum disorders; Cytokines; Hepatitis B virus; Vaccine; Immune response

Assessment of Renal Function in Children with Wilms Tumor: A Study Based on Urine Analysis and Ultrasound

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Background and Aim: Renal dysfunction is one of the inevitable complications in patients with Wilms tumor (WT). The purpose of this study was to assess the renal function in children with WT at baseline and every three months to 2 years.

Methods: In a descriptive-analytical study from 2018 to 2020, 48 children with WT were selected. Urine Creatinine (UCr), serum calcium (SCr), blood pressure (BP), estimated glomerular filtration rate (eGFR), and urinary protein (UPro) were evaluated at baseline and every three months during the study. Spot UCa/UCr and spot UPro/UCr ratio were calculated. Kidney ultrasonography was used in all patients to determine kidney size

Results: The mean age of patients at follow-up was 7.3 years. There was no significant difference in mean UCr, SCr, eGFR, 24-h UPro, UCa/UCr ratio, and spot UPro/UCr ratio at baseline and end of study ($P_{\text{baseline}} > 0.05$, $P_{\text{end of study}} > 0.05$). Analysis of kidney size showed a statistical association with tumor stage (P less than 0.05).

Conclusion: This study showed that as WT progressed, the size of the kidneys increases without any renal dysfunction. Therefore, it seems that urinalysis of patients with WT along with sonography is necessary to determine renal dysfunction and the use of ultrasound alone to determine kidney dysfunction is not recommended

Keywords: Renal dysfunction; Nephroblastoma; Wilms tumor

Blood Thrombosis after ChAdOx1 nCoV-19 (AZD1222) Vaccine: A Case Report

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Background and Aim: ChAdOx1 nCoV-19 (AZD1222) is a replication-deficient simian adenoviral vector that expresses the full-length SARS-CoV-2 spike protein. This vaccine causes T cell sensitivity against covid-19 virus spike protein and is a product of AstraZeneca Company and has been imported from South Korea to Iran through the World Health Organization's COVAX program. Iran Food and Drug Administration (IFDA) licensed general use of this vaccine. The fluidity of the blood in the circulatory system depends on various factors such as the speed and pressure of the flow provided by the heart and arteries, the coagulation system, etc., and an imbalance in any of these factors can lead to thrombosis. Blood coagulation due to most vaccines is a rare phenomenon, but it has clinical and pathological importance. In this case report, blood coagulation in a 63-year-old man from AZD1222 vaccine will be discussed. Following the injection of the vaccine, the man revealed symptoms of coagulation, including purpura and thrombocytopenia. Some secondary symptoms associated with thrombosis, such as hemolysis, anemia, and schistocytosis (an indicator of microangiopathic hemolysis) were also observed.

Methods: The man is 63 years old and has a history of thrombosis. He is also positive for HBV and HIV and has a history of kidney disease and high blood pressure. He is being treated for ART because of AIDS, and he is taking oral warfarin because of thrombosis. At the clinical examination, he complains of lethargy and dyspnea. He was vaccinated 6 days ago. The result of the oxygen saturation test is normal and has been measured at about 96%. The CBC test results show anemia (HGB:9.3 g/dL, HCT:29.5%) and thrombocytopenia (22 K/uL). In the results of biochemical analysis: increase in bilirubin (1.8 mg/dL) (indirect bilirubin 1.4 mg/dL), a decrease in haptoglobin (10 mg/dL) and an increase in lactate dehydrogenase (1229 U/L) are evident. This person has 354 CD4 T Cell lymphocytes without detectable viral load, which indicates that HIV is controlled. Reticulocyte levels have increased slightly (2.8%) due to a history of anemia. Peripheral blood smears include schistocytosis and spherocytosis, which confirm hemolysis. Von Willebrand factor activity is normal (97 IU/dL). Finally, CT-scan results rule out pulmonary embolism. By ruling out other causes of thrombosis and based on the available evidence, thrombosis caused by vaccination was diagnosed.

Conclusion: According to previous reports, thrombosis can be created by acquired causes such as autoimmune diseases, malnutrition and cancers (due to bone marrow dysfunction) or hereditary causes. Mutation in the ADAMTS13 gene is one of the most common inherited causes of thrombosis. According to recent studies, covid-19 also rarely leads to thrombosis, which has been shown to be due to the overproduction of coagulation factors in the infected people. Because of covid-19 outbreak,

which poses a serious threat to human society, scientists have made great efforts to develop vaccines with various techniques, including killing or attenuating viruses or using vectors for viral immunogenic subunits. Vaccines by stimulate immune system sometimes lead to autoimmune diseases. Previous studies demonstrated progression of vaccine activated immunity to autoimmune diseases such as Immune Thrombocytopenic Purpura (ITP) or Thrombotic Thrombocytopenic Purpura (TTP). Also vaccine-induced thrombosis has been shown to occur due to the production of anti-platelet factor (PF4) or anti-ADAMTS13 antibodies. Plus, one study in consistence with the results of our study reported thrombosis in a 55-year-old woman vaccinated with AZD1222, However, there is generally little information about this vaccine effects, and there is need for more extensive studies.

Keywords: Covid-19; AZD1222; Thrombosis



CDK Blockade Using AT7519 Suppresses Acute Myeloid Leukemia Cell Survival Through the Inhibition of Autophagy and Intensifies the Anti-Leukemic Effect of Arsenic Trioxide

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Background and Aim: The strong storyline behind the critical role of cyclin-dependent kinase (CDK) inhibitor proteins in natural defense against malignant transformation not only represents a heroic perspective for these proteins, but also provides a bright future for the application of small molecule inhibitors of CDK in the novel cancer treatment strategies. In the present study, we examined the restrictive potential of pan-CDK inhibitor AT7519 on the cellular and molecular features of acute myeloid leukemia-derived KG1 cells.

Methods: AML derived-KG1 cells were cultured with increasing concentrations of AT7519 either as a single agent or in combination with other anti-cancer agents. At different time intervals, to investigate the impact of CDK inhibition on cells survival and proliferation, its effects were analyzed by AnnexinV/PI, Trypan blue, MTT, Bromodeoxyuridine, Cell cycle, Acridine orange staining, and Real-time PCR tests.

Results: The results of the present study revealed that the inhibition of CDKs using AT7519, as revealed by the induction of G1 cell cycle arrest as well as the reduction of cyclins expression, resulted in decreased survival in AML-derived KG1 cells, either in the context of single-agent or in combination with ATO. Apart from alterations in the expression of the proliferation and apoptotic genes, the anti-survival property of AT7519 was coupled with the inhibition of autophagy-related genes. our study suggested for the first time that the activation of both PI3K and c-Myc signaling pathways could overshadow, at least partly, the efficacy of AT7519in KG1 cells.

Conclusion: Overall, this study suggests that AT7519 is a promising anticancer agent, either as a single agent or in a combined-modality strategy; however, further investigation should be accomplished to determine the usefulness of the inhibitor in cancer therapeutics, in particular for the treatment of acute myeloid leukemia.

Keywords: Acute myeloid leukemia; KG1 cells; Cyclin-dependent kinase; AT7519; autophagy; arsenic trioxide

Clinical Characteristics and Laboratory Outcomes of Multiple Myeloma Patient Infected with Covid-19 in Seyed Al-Shohada Hospital in 2020-2021

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Background and Aim: Multiple myeloma (MM) patients are immunocompromised, and infection is a big issue for them; therefore, the impact of covid-19 in these patients should be determined.

Methods: In a retrospective cohort study from March 2020 to October 2021, we analyzed 48 MM patients with positive SARS-CoV-2 test by polymerase chain reaction (PCR) or lung involvement confirmed by chest CT-scan hospitalized in Seyed-al-Shohada hospital, Isfahan, Iran. Patients' symptoms, laboratory findings, treatments initiated, and living status at the end of hospitalization were collected. The association between the symptoms and living status was assessed using Fisher exact test, and the risk was reported by risk ratio (RR) and its 95% confidence interval (CI). Moreover, the differences in laboratory parameters after receiving treatment for COVID-19 between alive and deceased patients were analyzed via ANOVA/ANCOVA; the standardized mean difference (SMD) was reported to show the measure of the relationship between parameters in two groups. P less than 0.05 was considered statistically significant.

Results: Results: Of 48 MM patients with a median age of 67, 13 died during treatment. Hypertension (13%) was the most common morbidity. In laboratory findings, high amounts of CRP (SMD=0.71; 95%CI: 0.01, 1.40; p=0.05), D-dimer (SMD=1.85; 95%CI: 0.91, 2.77; p less than 0.001), LDH (SMD=1.35; 95%CI: 0.52 , 2.16; p=0.001), and low platelet levels (SMD=-0.97; 95%CI: -1.61 , -0.31; p=0.004) were shown in deceased patients. Furthermore, dyspnea increased death risk by about 2.5 times (RR=2.57; 95%CI: 1.04, 6.34), but this finding was not statistically significant (p=0.08).

Conclusion: This study provided initial data to develop recommendations for MM patients with COVID-19 infection.

Keywords: COVID-19: Multiple Myeloma: Mortality

Coagulopathy Associated with COVID-19

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Background and Aim: Coagulopathy is a prominent feature of COVID-19; about one-third of deaths are directly attributed to pulmonary embolism (PE), more than 50% of patients experienced DVT, and increased D-dimer is a common finding and strong predictor for the more severe form of the disease. All of these findings highlight the occurrence of a hypercoagulable state in patients infected by the causative virus, SARS-CoV-2. The aim of this study was to assess the COVID-19 associated coagulopathy.

Methods: A systematic literature review was performed using appropriate keywords including, COVID-19, coagulopathy, bleeding, thrombosis, pulmonary embolism, hemorrhage, platelet in three different databases: PubMed, Scopus, and Google Scholar.

Results: Coagulopathy, most notably venous thromboembolism (VTE), is a leading cause of death in COVID-19. Increased FVIII and VWF levels are relatively common in COVID-19, directly related to the occurrence of thrombosis. A significant increase of FV also has been observed, directly associated with thrombotic events. Increased coagulation factor levels, shortened PT and APTT, and occurrence of VTE demonstrate the crucial role of coagulation factors in COVID-19-associated thrombosis. Damage to endothelial cells and release of procoagulants can trigger the coagulation cascade and activate platelets, resulting in thrombosis, a phenomenon that is less probable in inherited hypercoagulability. Although in normal circumstances, the endothelium protects against thrombosis, damaged endothelial cells provoke thrombosis by releasing VWF, FV, tissue factor, and plasminogen activator inhibitor (PAI)-1. Increased levels of all of these factors have been reported in COVID-19. Thrombosis may occur in COVID-19 as a result of vessel wall injury; this leads to the expression of TF, which proceeds to the extrinsic pathway of the coagulation cascade and has an important role in thrombosis.

Conclusion: Coagulopathy is the leading cause of death in COVID-19, thus identification of pathophysiology of COVID-19 associated coagulopathy can help to better management of COVID-19 and subsequently can decrease related morbidity and mortality.

Keywords: COVID-19; Coagulopathy; Thrombosis; Bleeding; Pulmonary embolism; venous thromboembolism

Comparison of Hematologic Parameters and C-Reactive Protein of Survivor and non-Survivor Patients with COVID-19

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Background and Aim: The complexity of the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) makes clinical course of the disease to develop rapidly, causing severe and deadly complications. Due to the variety in the clinical severity of COVID-19, several laboratory characteristics of patients such as erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), and hematologic parameters indicators can help predict patients based on their risk. This study was aimed to look for those tests in hospitalized patients with COVID-19.

Methods: Studied population included 114 patients with positive PCR COVID-19 test admitted to Valian hospital in Aligudarz (Iran) from October to December 2020. The disease outcome was followed along with the hospital course of every patient at the time of analysis. Among laboratory findings, complete blood count (CBC), erythrocyte sedimentation rate (ESR), and C-reactive protein (CRP) of all patients were compared between two admitted groups survivors (n=73) and non-survivors (n=41). Statistical analysis was conducted using IBM SPSS.

Results: The non-survivors revealed a significantly lower mean Hemoglobin (Hb), Hematocrit (HCT), and PLT levels also the results of qualitative assessments on CRP of survivor and non-survivor groups showed a significant association (P less than 0.05).

Conclusion: The identification of effective laboratory parameters can help predict clinical course, severity, and need for hospital resources and patient's outcomes in patients with COVID-19.

Keywords: COVID-19; Laboratory Test; Mortality; prognosis

Comparison of Secreted Frizzled-Related Protein -4 & -5 Promoter Methylation in Patients with Acute Myeloblastic Leukemia and Healthy Individuals

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Background and Aim: DNA methylation patterns are often changed in cancer cells. Many of the tumor inhibitor genes are silenced by methylation, such as CDKN2B, p73, and the suppressor of cytokine signaling in patients with acute myeloblastic leukemia (AML). Secreted frizzled-related proteins -4 and -5 (SFRP4, 5) are negative regulators of the Wnt signaling pathway. We aimed to evaluate the methylation status of SFRP4 and SFRP5 genes in patients with AML.

Methods: Blood samples were isolated from 60 patients with AML and 30 healthy controls. DNA was exploited, treated with sodium bisulfite, and tested utilizing methylation-specific polymerase chain reaction with specific primers for methylated and unmethylated sequences of the SFRP4 and SFRP5 genes.

Results: The frequency of unfit hypermethylation of SFRP4 and SFRP5 genes in patients with AML was characterized to be 50% (30/60) and 40% (24/60), respectively. Moreover, for all the subjects in the control group, methylation of SFRP4 and SFRP5 genes was negative. The spread of SFRP4 and SFRP5 promoter methylation in patients with AML were higher than in the control population

Conclusion: The present study showed that hypermethylation in SFRP-4 and -5 genes occurs in AML similar to solid tumors. Assessment of other antagonists of Wnt signaling pathways is recommended to further explore the status of DNA methylation, cell differentiation, and proliferation in different kinds of malignancies.

Keywords: Secreted frizzled-related proteins; DNA methylation; Acute myeloblastic leukemia; Hypermethylation

Consequence of Neutrophil Extracellular Traps in Covid-19 Patients

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Background and Aim: Neutrophils due to encountering pathogenic molecules produce NETs, the structure indicated with decondensed chromatin along with histones and intracellular components that can trigger immunothrombosis. Interaction of NETs with platelet and plasma coagulation factors may cause thrombotic events in COVID-19 patients.

Methods:

Results: Previous studies have shown that acute and chronic inflammatory disease such as sepsis and acute respiratory distress syndrome (occurs in 20% of COVID-19 patients) associates with dysregulated NETosis, accounts for hypercoagulability and thrombosis. the methodology used for assessment of NETs formation in these studies were common myeloperoxidase-DNA enzyme linked immune sorbent assay beside pioneer methods like NET imaging, revealed the undoubted correlation between COVID-19 severity and elevated levels of MPO-DNA, citrullinated histone H3, and cell free DNA (factors related to NETs) also pulmonary autopsies gathered from COVID-19 patients has confirmed neutrophil-platelet infiltration with NETs containing microthrombi. Other findings showed SARS-CoV-2 virus has the ability to active NETosis in consequence of increased level of intracellular Reactive Oxygen Species (ROS) while it can promote venous thrombus formations through its roll on fibrinolysis system of coagulation and complement system, moreover ROS indirectly stimulate pathogen elimination via NETs. Furthermore, therapeutic value of dexamethasone and Recombinant DNase1 has been demonstrated recently.

Conclusion: In this Review, we illuminate the latest updates of dysregulated neutrophil responses among severe COVID-19 patients with ARDS in charge with uncontrolled disease manifestations. Identifying high risk COVID-19 patients by screening for NETosis would be beneficial by applying an effective therapeutical strategy. It may be helpful in preventing destructive complications like lung injury, kidney damage, and immunothrombosis in severe COVID-19 cases.

Keywords: COVID 19; Neutrophil Activations; Thrombus

Convalescent Plasma Successfully Treats a Severe COVID-19 Patient with Multi-Organ Failure

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Background and Aim: Due to the high mortality rate of coronavirus disease 2019 (COVID-19) and the lack of specific treatment for the disease, it's essential to find new therapies. This study aimed to describe the first clinical experience for convalescent plasma (CP) transfusion administered to a patient with SARS-CoV-2 in Kermanshah, Iran.

Methods: We reported a severe case of COVID-19 with multi-organ failure, who had reduced oxygen saturation (92%) after several courses of antiviral therapy. Moreover, computed tomography (CT) scan results showed patchy lesions in the base of lungs and the result of the pharyngeal swab for SARS-CoV-2 using the Quest RT-PCR assay was positive 72 hours after admission. Therapeutic measures, including endotracheal intubation and plasmapheresis with convalescent plasma were performed. Two donors of convalescent plasma who recovered from SARS-CoV-2 infection were invited to donate their convalescent plasma. After testing negative for hepatitis B virus, hepatitis C virus, HIV, and syphilis before blood donation, the donors were invited to donate their plasma. the patient's blood group was determined for adaptability with the CP donors, and apheresis was performed with a cell separator.

Results: Following this, 1500 mL of convalescent plasma was obtained from the donors over three sessions. At each session, the patient received 500 mL of convalescent plasma. Therefore, the patient received CP from compatible donors during the three plasmapheresis sessions (days 3, 5, and 7 after admission), as an adjunctive treatment during septic shock. As clearly shown, this treatment reduced the count of activated leukocytes and increased lymphocytes. Moreover, renal function tests showed a relative improvement in renal failure and these results are consistent with the absorption of pulmonary lesions and increased arterial oxygen saturation (94.2%) after plasma transfusion.

Conclusion: Hence, our findings demonstrate that convalescent plasma can be a potentially effective treatment in SARS-CoV-2-infected patients.

Keywords: COVID-19; severe acute respiratory syndrome coronavirus 2; convalescent plasma; plasmapheresis

Correlation of Hematological Factors in Patients with COVID-19 Compared with Healthy Individuals

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Background and Aim: Coronavirus 2019 (COVID-19) is caused by a new virus, the acute respiratory syndrome of Coronavirus 2 (SARSCoV-2), which first occurred in December 2019. The virus penetrates the surface of the host cell through interaction with the angiotensin 2 (ACE2) receptor. In these patients, lymphopenia and leukopenia have been reported significantly in the Articles. Also, a blood test performed to measure variation in the size of red blood cells, called an RDW test, can accurately determine the likelihood of death in any of Corona patients. Of course, to increase the accuracy of this prediction, other indicators such as age, underlying diseases, the results of some other tests, etc. are also considered. In general, the higher the RDW obtained in an experiment, the higher the probability of death of a person with coronary artery disease. Higher numbers are more dangerous in young people with coronary heart disease than in the elderly. Also, people who see an increase in their RDW while in hospital are at greater risk of suffering long-term coronary heart disease.

Methods: We examined complete blood counts and peripheral blood smears of 20 patients with Covid-19 and 20 healthy individuals by reverse transcriptase-polymerase chain reaction (RT PCR). Peripheral blood samples were taken from every 20 healthy and sick individuals and the morphological characteristics of white blood cells, red blood cells, and platelets were examined.

Results: Of the 20 patients studied with a mean age of 45 ± 3.13 , 13 were male and seven were female, and of the 20 healthy subjects with a mean age of 43 ± 4.1 , 13 were male and 7 were female. The most common blood abnormalities seen in complete blood count (CBC) were low white blood cells, or lymphopenia and leukopenia. Also, our findings show that RDW was 14.8 ± 2.38 , and Lym was 19.2 ± 4.6 , and WBC changed to 3.88 ± 4.99 .

Conclusion: In conclusion, our study identifies and describes the morphological changes in the peripheral blood cells of COVID-19 patients. Although this is a pilot study with a small sample, it is a step towards understanding the hematologic manifestations of COVID-19. We observed that the levels of lymphocytes and white blood cells and RDW in patients are associated with significant changes and compared to healthy individuals, these indices are significantly reduced. And people at higher risk can be distinguished from people at lower risk with these tests.

Keywords: COVID-19; RDW; Lym; WBC

COVID-19 Infection in Patients with Hematological Cancer

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Background and Aim: COVID-19 pandemic has been started by the SARS-CoV-2 Infection. There is limited information about the risk of COVID-19 infection in hematological cancer and their clinical signs and outcomes. An inflammatory process called “cytokine storm” which is a deadly complication of COVID-19 infection is mediated by hyperactivation of effector T cells and excessive production of inflammatory cytokines like IL-6, IL-1, interferon-gamma, and TNF.

Results: BCR-ABL tyrosine kinase inhibitors (TKI) therapy in CML patients shows a slight increase in infection risk, probably because of off-target kinases inhibition which have a role in immune cell function. Clinically significant immune suppression is not caused by chronic phase CML or BCR-ABL tyrosine kinase inhibitors. Furthermore, no evidence suggests that chronic phase CML patients are at a higher infection risk by SARS-CoV-2 in comparison to population. Therefore, stopping TKI treatment is not suggested when symptoms of SARS-CoV-2 exist since it may cause loss of response and CML relapse/progression. Studies showed that Abl1 tyrosine kinase adjusts budding or release of poxviruses and the Ebola virus. These results show that c-Abl1 kinase signaling pathways have a vital role in viral departure and may be important in coronavirus replication. Imatinib may have anti-coronavirus activity in two points of life cycle of virus. In the early infection phase, it may block virion fusion with endosome and release into cytoplasm which inhibits viral entry and replication by Abl-mediated cytoskeletal rearrangement. In the latter phase, the expression of Abl2 protein which activates SARS-CoV and MERS-CoV replication is inhibited by imatinib and dasatinib.

Conclusion: Ultimately, these studies imply a potential protective role of the TKIs in SARS-CoV-2 infection. Moreover, it should be mentioned that TKIs have previously been shown to be effective against other coronaviruses. Finally, Abl2 could be a novel host cell protein required for viral growth.

Keywords: SARS-CoV-2; COVID-19, CML; Hematological Cancer; BCR-ABL tyrosine kinase

COVID-19 Vaccine: Adverse Effects after the First Dose of the Oxford-AstraZeneca within 12 Hours after vVaccination

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Background and Aim: Introduction The Oxford–AstraZeneca is one of the COVID-19 vaccines which plays an important role in controlling the pandemic. Oxford–AstraZeneca COVID-19 vaccine has given rise to some concerns due to happen mild to severe side effects in people who receive a vaccine.

Methods: Methods Case Presentation: A 26-year-old Iranian female with a lack of medical history presented with mild thrombocytopenia ($131 \times 10^3/\mu\text{l}$) 12 hours after getting the first dose of the Oxford-AstraZeneca vaccine. The patient presented with symptoms of purpura, nausea, muscle ache, vomiting, lethargy, and fainting. Also, laboratory testing revealed mild thrombocytopenia and lymphopenia. Her Troponin level, CPK, CK MB, LDH serum, ESR, and CRP were all normal.

Results: Result The patient felt better after serum therapy. Symptoms lasted for about 72 hours after vaccination. After 5 days, a temporary drop in a platelet count went back to the normal range ($196 \times 10^3/\mu\text{l}$) and her prothrombin time, partial thromboplastin time, and D-dimer were all normal.

Conclusion: Discussion & Conclusion Similar to other vaccines, the Oxford–AstraZeneca COVID-19 vaccine has some side effects which are identified within the first 24 hours following vaccination and usually lasted for about 24–72 hours. Vaccine-induced thrombotic thrombocytopenia(VITT) has already been described after vaccination with Oxford–AstraZeneca COVID-19 vaccine. In cases with mild to moderate symptoms, close monitoring via regular complete blood counts(CBC) and coagulation testing are highly recommended to diagnose VITT.

Keywords: COVID-19; Oxford–AstraZeneca; Side-Effect

D-Dimer Test Related to Coagulation Abnormalities in COVID-19 Patients

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Background and Aim: COVID-19 pandemic has become an urgent issue in the world. SARS-CoV-2 is a new and mutated type of coronavirus that may cause acute respiratory illness. COVID-19 can cause increased coagulation and inflammation, which is associated with increased levels of procoagulation factors. Disturbances in the normal state of homeostasis such as microangiopathic effects and localized clot formation, as well as systemic coagulation defects, leading to clot formation in large vessels and major complications of thromboembolism, including pulmonary thromboembolism, which are seriously observed in hospitalized patients.

Methods: In this study, based on hospital information, 100 ICU admitted COVID-19 patients in Ganjavian hospital, Iran (IR. DUMS.REC.1399.072) in the age range of 31 to 84 years that have a definitive positive diagnostic test for new COVID-19 (RT-PCR) without heart disease and anticoagulant therapy are enrolled. The results of laboratory tests including D-dimer, PT, PTT and platelet count was extracted from their medical files.

Results: Of 100 patients, the median age was 58.4-year (31-84), and 24 (48%) patients were men. Ten patients [20%] died and had comorbidities such as diabetes mellitus and hypertension. 9 of 50 [18%] and 7 of 50 [14%] patients had diabetes and hypertension, respectively. Laboratory data analysis of PT (mean 12.13, SD 2.8), PTT (mean 34.7, SD 8.5, and platelet count (mean 230.3, SD 123.0) not show significant changes compare to discharge group (P less than 0.05).

Conclusion: Coagulopathy is one of the most important poor prognosis factors in patients with COVID-19, which is associated with hospitalization in the intensive care unit and increase their mortality. Elevation of D-dimer above 1000 ng/ml was reported as a risk factor of death in COVID-19 patients (4). The present study has agreement with previous researches and showed increased D-dimer level is related to death compare to discharge patients.

Keywords: COVID-19; SARS-COV-2 infection; D-dimer test; Coagulation abnormalities

Detection and Quantification of HBV Genomes in PBMC of Chronic HBV Infection, Cirrhosis and Hepatocellular Carcinoma Patients

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Background and Aim: Increasing study revealed that the hepatitis B virus (HBV) exist in peripheral blood mononuclear cells (PBMCs). It remains poorly understood whether HBV DNA and covalently closed circular DNA (cccDNA) can be emerged in the PBMC of patients with different stage of HBV infection. The aim of this study was to compare the detection of HBV DNA and quantification and presence of cccDNA within PBMC from chronic HBV infection (CHB), cirrhosis and hepatocellular carcinoma (HCC) individuals.

Methods: Present study has been conducted on 120 participants (30 CHB patients, 30 cirrhosis patients, 30 HCC patients and 30 healthy control) from Tehran, Iran. HBV serological markers were tested by ELISA assay. PBMC of all individuals were assayed for HBV DNA detection and quantification and presence of cccDNA.

Results: Of all 90 HBV patients, 58 (64.4%) were positive for HBV DNA in PBMC. HBV DNA was detected in PBMC isolated from 13/30 CHB patients, 20/30 cirrhosis patients and 25/30 HCC patients. Also, 6 (20%) of CHB, 13 (43.3%) of cirrhosis, and 16 (15.3%) of HCC patients were cccDNA positive. The serum HBV viral loads were statistically higher than the HBV viral loads of PBMCs (P -value < 0.001. Positive correlation was found between HBV DNA loads in serum and PBMC of patients. Moreover, HBV DNA quantity of serum and PBMC in terms of HBeAg status showed considerable association.

Conclusion: HBV quantity in PBMC correlated with serum HBV viral load. HBV genomes in PBMC may be a risk factor for HBV disease progression.

Keywords: Hepatitis B virus; Chronic hepatitis B; Cirrhosis; Hepatocellular carcinoma; peripheral blood mononuclear cells; Covalently closed circular DNA

Determinants of Out of Pocket Payments, Productivity Lost and Influential Factors on them Among Children Cancer (Infant)

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Background and Aim: Cancer is the second cause of morbidity and mortality in developed countries and the third leading cause of death in developing countries. Cancer not only threatens life and welfare (•) but also threatens financial security. treatment of cancer patients causes financial problems for patients and their families and patients may face high expenses for their treatment with high expenses. Although it is primarily important to treat physicians and patients, financial concerns can affect their decisions. Therefore, the high costs of paying out of pocket may delay the treatment or failure of patients, especially those who have experienced financial problems. Therefore, this study aimed to determine the amount of expenses incurred from the pocket of patients with cancer and the cost of loss of parents, and the factors affecting it in children with cancer in pediatric cancer hospital. (Mohamad Kermanshahi)

Methods: This study was a cross-sectional study that was carried out as a cross-sectional study from October 2009 till end of September 2009 (in hospital Mohamad Kermanshahi). A questionnaire was used to collect data. All of the children who were diagnosed with cancer who had an active file to undergo chemotherapy or hospitalization were invited to participate in the study. The data were collected at the time of discharge through interviews with the patient or with the patient. Finally, the data were analyzed by (SPSS) software and statistical tests according to variables

Results: The results show the mean of direct treatment and non-treatment costs of patients in patients ' pockets during initial treatment were 22,779,129 and 23,908,083 rials, respectively. The total cost of 44,193,314 was 44,193,314 rial. Examples of chemotherapy, chemotherapy, surgery and diagnostic services such as radiology, laboratories, were the most responsible causes of cost. As part of the total cost, chemotherapy drugs had the highest share of the total cost. The results showed that 11 % of patients rated the cost of financial pressure as heavy and intolerable, and 23 % heavy but tolerable. It is also an important part of costs for cases such as travel and settlement, which are not covered by insurance , especially for patients who live far from the center of cancer.

Conclusion: It is necessary to adopt new policies regarding the heavy costs of cancer patients, especially from insurance organizations and financial support from the government and other organizations to reduce financial burdens on cancer patients and they can better manage their health. Also, considering the fact that the costs of cancer children are related to non-therapeutic costs. To do the costs that are not in the commitment of premiums, such as accommodation of patients and their companions especially cancer children should be considered. Considering that many of the costs cannot be calculated for these patients, loans or without interest should be considered.



Keywords: Cancer children: paying out of pocket: loss of parents; hospital



Determining the Treatment Strategy of Thrombocytopenia in Leukemic Patients with Coronavirus Disease(SARS-CoV-2)

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Background and Aim: Thrombocytopenia is a clinical disorder that occurs due to a number of reasons: decreased platelets production, increased peripheral destruction, increased spleen sequestration, and dilution. The normal range of platelets is 150-450x10⁹/L, and thrombocytopenia refers to when the platelet count falls below 150x10⁹/L. thrombocytopenia is common in patients with hematological malignancies. Thrombocytopenia in patients with leukemia is due to the invasion of cancer cells into the BM or to the suppression of chemotherapy drugs or megakaryocyte(MK) disorders. On December 2019, a severe lung disease of unknown origin was reported in China. It was later discovered that the cause was a new strain of coronavirus, now known as severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2). The virus can cause mild to severe respiratory infections in humans. Common symptoms of coronavirus disease include fever, dry cough, difficulty breathing. The risk of the venous thromboembolism and chemotherapy-induced thrombocytopenia is significantly higher in cancer patients than in non-cancer patients. In addition, inflammation caused by coronavirus disease can lead to endothelial dysfunction, resulting in pro-thrombotic and hyper-coagulable states and before starting treatment with anticoagulants, chemotherapy-induced thrombocytopenia, and coronavirus disease-induced thrombocytopenia should be considered. The aim of this study was to develop a treatment policy for leukemic patients who also have coronavirus disease.

Methods: We searched PubMed; Scopus and Google Scholar for case reports and series of COVID-19 patients with concomitant hematological malignancy; published until November 2021.

Conclusion: According to the treatment plans defined in coagulation and hemorrhagic disorders of leukemia patients, it seems that the addition of infections such as Covid-19 requires that pre-determined treatment guidelines be prepared and implemented for this group of patients.

Keywords: COVID-19; Thrombocytopenia; Leukemia

Dwindled Serum IgG Levels of Rubella, Diphtheria Toxin, Hepatitis B Virus (HBV) and Tetanus Toxoid after Chemotherapy; a Report from Iranian Children with Malignancy

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Background and Aim: Chemotherapy suppresses immunoglobulin production as a result of cell toxicity. Decreased immunoglobulin levels can result in the onset of opportunistic infections. The aim of the current study is to compare the immunoglobulin G (IgG) levels of the selected vaccine-preventable disease (VPD) before and six months after chemotherapy in a group of Iranian children with malignancies.

Methods: In this interventional study, serum levels of Rubella, Diphtheria toxin, Hepatitis B virus (HBV), Tetanus Toxoid, Mumps, and Measles IgG were measured in 30 children with malignancy and previously vaccinated for these diseases. Six months after chemotherapy, serum IgG levels were reassessed and compared with their corresponding pre-chemotherapy levels.

Results: In this study, 17 (56.7%) male and 13 (43.3%) female were included. The mean age was 7.69 ± 3.09 years. After chemotherapy, Rubella IgG levels dropped from 73.88 ± 85.11 to 56.59 ± 72.84 IU/mL (P less than 0.05; $r = 0.956$; 33.4% become serum negative (SN)). Diphtheria toxin IgG was diminished from 0.683 ± 0.454 to 0.174 ± 0.248 IU/mL (P less than 0.05; $r = 0.601$; 26.7% SN). Anti - HBV IgG showed a reduction from 46.26 ± 101.56 to 25.56 ± 80.49 IU/mL (p less than 0.05; $r = 0.524$; 60% SN) and Anti -Tetanus Toxoid IgG fell down from 1.031 ± 0.582 to 0.321 ± 0.408 IU/mL (p less than 0.05; $r = 0.365$; 33.4% SN). Anti -Measles and Anti -Mumps IgGs showed no significant change (p greater than 0.05).

Conclusion: Pediatric chemotherapy was associated with dropped serum IgG levels of most VPDs. A good correlation was also observed between serum levels of IgG before and six months after chemotherapy. Revaccination of children with malignancies may be necessary upon declined serum IgG titers.

Keywords: Chemotherapy; Immunoglobulin G; Neoplasm; Vaccine-preventable diseases

Effect of Transgenic Leishmania Major Expressing mLLO-Bax-Smac Fusion Gene in the Apoptosis of the Infected Macrophages

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Background and Aim: Leishmaniasis is a complex infection against which no confirmed vaccine has been reported so far. Transgenic expression of proteins involved in macrophage apoptosis-like BAX through the parasite itself accelerates infected macrophage apoptosis and prevents Leishmania differentiation. So, in the present research, the impact of the transgenic Leishmania major including mLLO-BAX-SMAC proapoptotic proteins was assayed in macrophage apoptosis acceleration.

Methods: The coding sequence mLLO-Bax-Smac was designed and integrated into the pLexyNeo2 plasmid. The designed sequence was inserted under the 18srRNA locus into the L. major genome using homologous recombination. Then, mLLO-BAX-SMAC expression was studied using the Western blot, and the transgenic parasite pathogenesis was investigated compared with wild-type L. major in vitro and also in vivo.

Results: Western blot and PCR results approved mLLO-BAX-SMAC expression and proper integration of the mLLO-Bax-Smac fragment under the 18srRNA locus of L. major, respectively. The flow cytometry results revealed faster apoptosis of transgenic Leishmania-infected macrophages compared with wildtype parasite-infected macrophages. Also, the mild lesion with the less parasitic burden of the spleen was observed only in transgenic Leishmania-infected mice. The delayed progression of leishmaniasis was obtained in transgenic strain-injected mice after challenging with wild-type Leishmania.

Conclusion: This study recommended transgenic L. major including mLLO-BAX-SMAC construct as a pilot model for providing a protective vaccine against leishmaniasis.

Keywords: Transgenic Leishmania major; Apoptosis; mLLO-Bax-Smac

Evaluation of Blood Coagulation Changes in Patients with COVID-19

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Background and Aim: The discovery of new biomarkers in Covid-19 disease can play a very important role in the diagnosis, prognosis, or even treatment of this disease. The aim of this study was to evaluate the coagulation parameters in patients with confirmed Covid-19 infection compared with healthy control.

Methods: In this case-control study, 92 patients with Covid-19 and 46 healthy individuals were included in the study. Primary and secondary hemostasis in patients and healthy individuals were assessed by prothrombin time (PT), international normalization ratio (INR), partial thromboplastin time (PTT), thrombin time (TT), and fibrinogen tests. To evaluate the fibrinolytic system, the levels of anti-thrombin (AT), d-dimer, and fibrin degradable products (FDP) were measured according to current guidelines. Also, complete blood count (CBC) was assessed using an automated hematology analyzer.

Results: In the present study, the mean age of patients and healthy individuals were 49 ± 15 , and 38 ± 11 years, respectively. Based on the CBC parameters, there was no statistically significant difference in the erythrocyte, leukocyte, and platelet indices between patients and healthy individuals (p greater than 0.05). Coagulation tests showed that there were no statistically significant differences in PT, INR, PTT, and TT between patients and healthy individuals (p greater than 0.05). However, it was shown that the level of fibrinogen in the patients was significantly higher than healthy group (p less than 0.001).

Conclusion: Coagulation tests and CBC indices were not significantly different in patients with Covid-19 compared with healthy individuals. However, measuring fibrinolytic system variables such as d-dimer, FDP, and AT can be helpful in the initial evaluation of patients with Covid-19, and determining prognosis.

Keywords: Coagulation; Fibrinolytic; Covid-19

Evaluation of Changes in Erythron, Erythropoietin and Proinflammatory Cytokines in Mice Exposed to Bisphenol A and Evaluation of the Protective Role of Zinc Oxide Nanoparticles

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Background and Aim: Bisphenol A is the raw material for the production of polycarbonates and materials such as Epoxy resin, helmets, danger signs, etc. that humans are always exposed to this substance (BPA). In the present study, the toxic effects of BPA, as well as the possible protective effects of Zinc oxide nanoparticles on erythron, erythropoietin and proinflammatory cytokines in mice, Were investigated. For this purpose, 40 mice with an approximate age of 7-8 weeks and an Approximate weight of 30 g were selected and divided into two groups of 20 (BPA +), (BPA -). each group includes two subgroups (ZNO +) and (ZNO -). the results of the present study showed that administration of BPA significantly increased the production of proinflammatory cytokines such as IL-6, TNF- α and increased serum urea and creatinine, which indicate the induction of inflammatory and kidney damage. while the level of EPO and The number of red blood cells and hemoglobin in mice with chronic poisoning with BPA Decreased significantly, which indicates that these mice have anemia.

Methods: Clinical Trial

Results: Administration of ZNO NPS had no side effects on control mice. it was also observed that the administration of this zinc supplement prevented the increase of urea, creatinine, IL-6, and TNF- α by preventing the induction of kidney damage. it also prevented the reduction of EPO, RBC and Hb. These findings indicate the reduction of kidney damage and the protective effects of this NPS against the induction of anemia due to BPA poisoning.

Conclusion: This finding indicates the reduction of kidney damage and the protective effects of this NPS against the induction of anemia due to BPA poisoning

Keywords: Bisphenol A; Zinc oxide nanoparticles; erythropoietin; proinflammatory cytokines

Evaluation of Cinnamon Inhibitory Effect on Prevention of Foam Cell Formation in Platelet and Monocytes Coculture (U937 cell line)

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Background and Aim: Atherosclerosis is one of the leading causes of cardiovascular disease. Following endothelial damage and platelet aggregation in that area and the recruitment of monocytes and their conversion to macrophages, LDL gradually accumulates under the endothelial artery wall and gradually oxidized and convert to oxi-LDL. By swallowing it, the macrophages turn into foam cells and then atheroma formation. Medicinal plant extracts are used to treat diseases due to their low side effects. Cinnamon water extract modulates macrophage activation and lowers cellular cholesterol and is useful in the treatment of atherosclerosis. Therefore, in this study, the inhibitory effect of cinnamon in preventing the foam cells formation in platelets and monocytes co-culture was evaluated.

Methods: The cellular toxicity of cinnamon cassia water extract was assessed by the MTT test on the U937 (monocyte) cell line and the best dose was selected. The formation of foam cells was measured by Oil Red O staining and the expression of cholesterol receptor genes in macrophages using Real time PCR.

Results: The data showed that the formation of foam cell when treating macrophages and platelets with cinnamon water extract compared to control, had a significant reduction. The expression of LXR α , CD36, ABCA1, and PPAR γ genes, as the output receptor, input, and nucleus of cholesterol in macrophages, increased significantly compared to control in samples treated with cinnamon. (P value less than 0.05)

Conclusion: Cinnamon water extract reduces the formation of foam cells and can be recommended in the treatment of atherosclerosis.

Keywords: Cinnamon; platelet; foam cell; atherosclerosis

Evaluation of Comparative Effect Microvesicles Derived from Human Umbilical Cord and Placental Mesenchymal Stem Cells on the Apoptosis and Prolifration of Acute myeloid Leukemia Cell Line (KG1)

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Background and Aim: AML is a malignant hematological disease. New biologicals reagents for the treatment of AML are urgently needed now, MVs have been identified as candidate biomarkers for disease treatment and prognosis. We investigated and compared the effects of MVs derived from MSCs of umbilical cord and placental on cell proliferation through apoptosis on the KG1 cell line.

Methods: MSCs of confirmed umbilical cord and placenta were cultured and in the sixth passage, supernatant was collected and MVs derived were isolated by ultracentrifugation. The concentration of MVs was determined by Bradford, their properties by TEC and DLS method. The KG-1 cell line was treated with MVs at concentrations of 25, 50 and 100 µg/ml for 24h. Annexin V-FITC/PI kit was used to evaluation apoptosis. RT-PCR was used to study the expression of BCL-2 and BAX genes involved in the apoptotic pathway. Finally, statistical calculations were performed using ANOVA using t-test and GraphPad Prism software.

Results: The rate of apoptosis by flowcytometry in KG1 cells treated with PMSC-MVs at concentrations of 25, 50 and 100 µg/ml, 54.95%, 49.77% and 52.5% and with UCMSC-MVs is 47.85%, 47.15% and 51.35%. Therefore, a significant increasing was observed in all concentrations in both groups compared to the control sample. BCL-2 proapoptotic gene expression showed a significant increase and anti-apoptotic BAX gene decreased significantly compared to the control sample. BCL-2/BAX ratio showed a significant decrease. Also, the comparison of apoptosis and expression of these two genes in the 2 groups of compared to each other did not show a statistically significant difference.

Conclusion: PMSC-MVs and UCMSC-MVs have anti-proliferative effects by inducing apoptotic activity in the KG-1 cells, which may be due to increased BAX gene expression and decreased BCL-2 gene.

Keywords: Apoptosis; AML; PMSC-MVs and UCMSC-MVs

Evaluation of Inflammatory Indices in Gestational Diabetes Mellitus of Pregnant Women

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Background and Aim: Gestational diabetes mellitus (GDM) is one of the main threats during pregnancy whose consequences are dangerous for pregnant women and their infants. Given the undeniable role of chronic subclinical inflammation in developing GDM, inflammatory indices such as hemoglobin-to-platelet ratio (HPR), platelet-to-lymphocyte ratio (PLR), and blood biomarkers can help predict the consequences of GDM in pregnant women.

Methods: This retrospective study analyzed the profiles of 300 pregnant women with a definitive diagnosis of GDM hospitalized in Imam Reza Hospital (Khorasan Razavi Province, Iran) and Mohebbe Yas Hospital (Tehran Province, Iran) between the years 2018 and 2020. GDM diagnosis was confirmed based on WHO (2013) and NICE (2015) protocols by measuring OGTT, FBS, and 2-hour PG. This study evaluated absolute cell counts, hematological and biochemical parameters, and cellular indices in two groups (Group I: gestational age ≤ 28 ; Group II: gestational age > 28). The ROC curve analysis evaluated the sensitivity and specificity of statistically significant variables in predicting GDM and inflammatory consequences. The data were given as mean and standard deviation (mean $\pm$ SD), and the statistical significance was defined at a *P*-value of less than 0.05.

Results: According to the results, PLR and HPR in Group I were 33.46 ± 62.47 SD and 0.0432 ± 0.0287 SD, respectively, and both were statistically significant (*P*-value for PLR: 0.004 and *P*-value for HPR: 0.001). Platelet count was also higher in Group I (599.140 ± 111.849 SD $\times 10^9/L$) than in Group II and was statistically significant (*P*-value: 0.001). The ROC curve analysis revealed that the sensitivity and specificity of PLR were 81.39 and 27.36 %, respectively (95% CI: 0.532 – 0.662). The sensitivity and specificity of the HPR index were calculated as 30.81 and 78.23%, respectively (95% CI: 0.579 – 0.705).

Conclusion: The results of this study reveal that PLR and HPR indices are significantly associated with GDM in gestational age ≤ 28 , showing that they can be used as predictors of GDM in the future.

Keywords: Gestational diabetes mellitus; Pregnancy; Inflammatory markers; Risk prediction.

Evaluation of Intestinal Parasitic Infections in Patients Referring to the Clinical Laboratory in Urmia City, Iran from March 2019 to March 2021

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Background and Aim: Parasitic contamination is one of the most important worldwide health problems, especially in developing countries. Various types of intestinal parasites can cause contamination in humans, and create a wide range of symptoms, depending on different immunological, physiological, and social factors. The risk of contamination is high in immunocompromised patients, in patients receiving immunosuppressive drugs and in patients undergoing chemotherapy this study aimed to determine the prevalence of parasitic infections in patients referred to the Medical Laboratory in Urmia city.

Methods: All specimens were examined for the presence of parasites by direct test. Then, to accurately detect the type of parasite, positive specimens were determined by genus and strain using ether formalin concentration and trichrome staining.

Results: Of 10000 stool samples 1032 samples (10.32%) had a parasitic infection, The prevalence of parasitic infections was as follows: *Chilomastix mesnili* with 42.05% (270 men and 164 women with 26% of the total positive samples were related to the male samples and 15.89% were related to the female samples); *Blastocystis hominis* with 13.75% (87 men and 55 women with 8.43% of the total positive samples were related to the male samples and 5.32% were related to the female samples), *Entamoeba coli* with 9.10% (53 men and 41 women 5.13% of the total positive samples were related to the male samples and 3.97% were related to the female samples); *Giardia lamblia* with 3.97% (29 men and 12 women with 2.81% of the total positive samples were related to the male samples and 1.16% were related to the female samples); *Trichomonas hominis* with 1.55% (6 men and 10 women with 0.58% of the total positive samples were related to the male samples and 0.96% were related to the female samples). The lowest infection rate was related to *Ascaris lumbricoides* (3 men), *histolytica* / *dispar amoeba* (3 men) and *Hymenolepis nana* (3 women) each Which with 0.04 % contamination was.

Conclusion: By promoting the level of health, it can be prevented from getting this Various types of intestinal parasites can cause contamination in humans and create a wide range of symptoms, depending on different immunological, physiological.

Keywords: intestinal parasitic; infections in patients

Evaluation of Membrane Progesterone Receptor Expression on K562 Cells and the Effect of Progesterone on Growth of the Cells

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Background and Aim: Chronic myeloid leukemia (CML) is an uncommon type of cancer of the white blood cells that starts in the bone marrow and invades the blood. Progesterone inhibited growth of tumor cells that expressed progesterone receptors. Here, we first investigated membrane progesterone receptor expression (mPR-beta) on K562 cells and then evaluated the progesterone effect on the growth of the cells.

Methods: After culturing K562 cells in a medium containing serum, expression of mPR-beta using flow cytometry and progesterone effect using MTT cell viability assay were investigated.

Results: K562 cells expressed mPR-beta. Progesterone inhibited growth of K562 cells in a dose- and time-dependent manner.

Conclusion: Progesterone due to killing K562 cells may be considered as a promising therapeutic potential in CML patients in the future.

Keywords: CML; Progesterone; mPR-beta

Evaluation of Prognostic/Diagnostic Value of Hematological Markers in the Detection of Inflammation in Coronavirus Disease: A Review Study

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Background and Aim: The Coronavirus infectious disease 2019 (COVID-19) has recently emerged as a pandemic and has endangered the lives of thousands of people worldwide. No specific treatment has been identified so far. However, the identification of diagnostic and prognostic factors in patients can be valuable for the application of preventive strategies. In this study, we evaluated the diagnostic value of inflammatory markers in COVID-19 infected patients and identified associated organ disorders.

Methods: This review article provides an overview of the published papers on COVID-19. The relevant literature was searched in the PubMed database (from December 2019 to April 2020); the search was limited to the English language and the terms “COVID-19”, “Inflammation”, “Platelet”, “Lymphocyte”, “Neutrophil” and “Diagnosis” were used. This study was also written using 30 articles, including 25 cohorts and 5 case-control articles. We included studies that complied the following criteria: prospective or retrospective observational studies (cross-sectional, case-control and cohort studies), adult patients (aged > 18 years old) who were diagnosed with COVID-19, NLR and PLR values reported at hospital admission and the association between NLR and PLR values and disease severity or other clinical outcomes in COVID-19 patients was reported. We did not expect to find randomized controlled trials of NLR and PLR, as NLR and PLR cannot be randomized as interventions. Moreover, we excluded studies that were conducted in animals, duplicated, conference abstracts, case reports, systematic reviews, scoping reviews and editorials or commentaries. Our primary outcome was disease severity, which was defined as meeting at least one of the following criteria: ICU admission, shortness of breath, respiration rate ≥ 30 times per minute, blood oxygen

Results: Increased platelet-to-lymphocyte ratio (PLR), neutrophil-to-lymphocyte ratio (NLR), prothrombin time (PT), and D-dimer indices have been found in patients with increased inflammation and thrombosis and can lead to heart disease. Also, an increase in these indices is accompanied by worsening of the disease and impairment of the respiratory tract, which necessitates ventilation for the patients eventually. The evaluation of NLR, PLR, and coagulation parameters can be useful for identifying high-risk individuals who need to be intubated. Patient survival will improve by the timely identification and the use of appropriate treatment strategies.

Conclusion: Recently, the use of hematological markers has become a significant research topic because of the diagnostic and, occasionally, a prognostic role for identifying high-risk individuals. It seems that these markers' evaluation is inexpensive and of high value in the diagnosis and response



to treatment. Identifying and monitoring COVID-19 patients, which requires aggressive methods to track patients' clinical status, is a significant problem, and the timely detection and identification of high-risk individuals can help reduce the use of aggressive therapies such as intubation. Evaluating said parameters at the time of diagnosis can increase patient survival by identifying high-risk individuals who need to be intubated.

Keywords: COVID-19; Diagnosis; Inflammation; Lymphocyte; Neutrophil; Platelet



Frequency and Types of Cytogenetic Abnormalities in Pediatric Acute Lymphoblastic Leukemia: Report of 11 Years

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Background and Aim: Acute lymphoid leukemia (ALL) is the largest subset of hematologic malignancies, accounting for approximately 70-80% of childhood leukemia, and is most common at age 4 years. The aim of this study was to define the frequency of all cytogenetic abnormalities impact of cytogenetics on outcomes in pediatric ALL during the 11-year period.

Methods: In this 11-year retrospective study, we investigated 99 patients which referred to our department due to ALL from 2010 to 2020. The age group of the patients ranged from 6 months to 14 years with a mean of 6.71 ± 4.09 years. Clinical and diagnostic findings were extracted from patients' medical records.

Results: We showed cytogenetic abnormalities of 99 pediatric ALL patients, including 78 (78.8%) pre B-ALL, 9 (9%) common B-ALL, and 12 (12.2%) T-ALL cases. The 5-year overall survival rate (OSR) and event-free survival (EFS) of all cytogenetic abnormalities ($n = 99$) were 48% and 43%, respectively. The 5-year OSR for B-ALL and T-ALL abnormalities was 63% and 46% ($P = 0.269$), whereas EFS was 69% and 51% ($P = 0.236$), respectively. There was a significant relationship between the two cytogenetic abnormalities, hypodiploidy and $t(9;22)$, with death ($P < 0.05$). On comparing the subjects with normal cytogenetics to the other cytogenetic abnormalities, EFS was significantly low for hypodiploidy ($P = 0.0163$, hazard ratio = 0.5308) and $t(9;22)$ ($P = 0.0131$, hazard ratio = 0.4908), while other cytogenetic abnormalities did not have a statistically significant difference in EFS.

Conclusion: Our results emphasized the importance of the cytogenetic findings in evaluating the survival outcomes, which allows identifying a variety of OSR and EFS, because some of the cytogenetic abnormalities may interfere with the death and prognosis.

Keywords: Acute lymphoblastic leukemia, Cytogenetic abnormalities, Karyotype, survival

Golden Vision for Quick Access of COVID-19 Patient's Management in Based on Clinical Results by Cytoscape

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Background and Aim: In the age of research, data in unprocessed form cannot be used even by professional scientists. To solve this issue, bioinformatics has emerged. Bioinformatics includes the application for the development of computational tools to bring together visualize, archive store, and acquire data in the right form to help interpret them. Bioinformatics manages complexity and reduces wasting time, human error in any process that human involves. Due to bioinformatics, the researcher can access information rapidly for their experiment and have a place to share and publish their result as a basis for another research. Furthermore, evolving in precision medicine, both for patients and doctors include more accurate diagnosis and treatment, safer drug prescription. Bioinformatics plays a vital role in providing every element required for processing, visualization, and interpretation of a patient's based on particular attributes that are unique to a person, thereby personalized medicine is improved. Network interaction a branch of bioinformatics is a structure of linkages and interactions. Cytoscape a popular bioinformatic package is for creating network visualization. In this app, based on what is connected to what, nodes represent clinical data of COVID-19 patients, and edges are used as the biological linkage between nodes. Besides, a graphical abstract has been prepared.

Methods: Data gathered from google scholar and PubMed sites.

Results: Creating a network visualization with Cytoscape can provide quick and correct supervision for handling the COVID-19 pandemic.

Conclusion: In conclusion, perhaps it could be possible to combine precision bioinformatics and network visualization as a guideline to create a network model for covid-19 patients based on clinical results to improve the accuracy of treatment decisions and prognosis prediction to achieve a better outcome and change human life.

Keywords: Cytoscape; Bioinformatic; COVID-19

Hematological Abnormalities in Patients with COVID-19: An Emerging Approach to Differentiate between Severe COVID-19; Compared with Non-Severe forms of the Disease

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Background and Aim: The 2019 novel coronavirus (2019-nCoV) or the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has emerged as a pandemic threat from December 2019. Coronavirus can cause varying degrees of illness that range from mild to severe or fatal disease. The exact mechanism of hematopoiesis induced by this coronavirus is not yet well understood, but scientific evidence indicates that COVID-19 can cause hematological changes in infected patients.

Methods: The present study summarized pieces of literature regarding hematologic findings of COVID-19 and their correlation with disease severity.

Results: Among hematological parameters, decreased hemoglobin rather than anemia, leukocytosis, lymphopenia, neutrophilia, and thrombocytopenia have been observed in conducted studies in some patients with COVID-19. Furthermore, as the disease progresses to severe COVID-19, hemoglobin decline, leukocytosis, lymphopenia, neutrophilia, and thrombocytopenia continue to exacerbate. In addition, the neutrophil-to-lymphocyte ratio is also considered as an independent risk factor for severe infection in COVID-19 patients.

Conclusion: We offered some laboratory abnormalities which help to differentiate severe COVID-19 from non-severe forms of the disease.

Keywords: Coronavirus disease (COVID-19); Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2); Hemoglobin; Lymphopenia; Neutrophilia; Thrombocytopenia

Homocysteine Biomarker and Coagulation Abnormalities Related to COVID-19 Infection

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Background and Aim: Coronavirus 19 is a new type of coronavirus that causes acute respiratory pneumonia (1, 2). Due to the possibility of vascular thrombosis and myocardial infarction in patients with new coronavirus, evaluation of these patients in terms of effective biomarkers in causing thrombosis seems necessary (3). Homocysteine is known as a biomarker of thrombosis (4, 5). In this research, we aimed to assess the serum homocysteine level and PT, PTT, D-dimer as the parameters related to coagulation abnormalities.

Methods: Material and Methods: In this case-control study, 50 patients of COVID-19 hospitalized in ICU ward in Ganjavian (IR.DUMS.REC.13399.057) who have a definitive positive diagnostic test of COVID-19 (RT-PCR) and 50 healthy persons as control group that have negative RT-PCR test and also have no history of cardiovascular, kidney, liver and stroke diseases and also have no history of smoking and anticoagulants therapy that are in the same age range are enclosed. The serum level of homocysteine, d-dimer, platelet count was determined by conventional analysis.

Results: The patients' levels of homocysteine, vitamin B12, and folic acid in the patient group did not show a significant difference compared to the control group ($p < 0.05$). Additionally, patients D-dimer (mean 1420, SD 1230) show a significant and parallel elevation with ferritin (mean 337, SD 123) level as a marker of inflammation and the disease severity in patients group compare to control group ($p < 0.05$).

Conclusion: Homocysteine is a metabolite derived from the demethylation of the amino acid methionine, essential amino acid and is a biomarker of thrombosis (4, 5). Despite the elevation of D-dimer as determined coagulation activation test no significant elevation was obtained in the patients group compared to the control group ($p < 0.05$). This showed homocysteine may not be a good biomarker in coagulation abnormalities and severity in COVID-19 patients.

Keywords: Homocysteine; Biomarker; Coagulation abnormalities; COVID-19

Human Platelet Antigens Polymorphisms in Hepatitis C Virus Infection

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Background and Aim: Polymorphism in human platelet antigen (HPA)-1 and HPA-3 (GPIIb/IIIa), HPA-2 (GPIb/IX), HPA-4 (GPIIIa), HPA-5 (GPIa/IIa), & HPA-15 (CD109) was investigated in 71 patients with minimal/mild stages of fibrosis (Group 1; G1) and patients with moderate/severe degrees of fibrosis (Group 2; G2).

Methods: HPA genotyping was done by PCR-SSP method.

Results: Lower HPA-3a and higher HPA-3b ($P=0.028$) allele frequencies were seen in G2 patients than G1 patients, and homozygosity for HPA 3b ($P=0.038$) alleles was more prevalent in G2 patients than in G1 patients. The allele and genotype distributions of the other HPA polymorphic variants were similar between G1 and G2 patients. Logistic regression analysis showed that genotype HPA3 bb ($p=0.015$) and severe fibrosis degree (p less than 0.011) were independent predictors of low platelet count.

Conclusion: This is the first evidence demonstrating the differential association of the common six HPA gene variants and specific HPA genotype combinations with fibrosis progression in hepatitis C virus infection.

Keywords: Human platelet antigen; Hepatitis C virus; Liver fibrosis

Inhibitory Effect of Interferon- Beta and Glatiramer Acetate on the Release of Endothelial Cell Derived Microparticles in Patients with Multiple Sclerosis

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Background and Aim: Increasing concentrations of Microparticles (MPs) have been found in many autoimmune diseases including Multiple sclerosis (MS). In response to the activation of lymphocytes, and the production of inflammatory cytokines, brain endothelial cells release MPs from their membrane. We aim to study the inhibitory effect of interferon-beta (INF β) and glatiramer acetate on the release of endothelial cell-derived microparticles (EnMPs) in patients with MS.

Methods: In this clinical trial study, 40 patients who were new cases of MS, were divided into two groups (N=20). The first group (G1, n=20) received INF β and the second group (G2, n=20) received glatiramer acetate. In two stages, before the beginning of treatment and three months after the start of treatment, blood samples were taken. Flow cytometry was used to count and determine the phenotype of microparticles. Phycoerythrin (PE) anti-human CD51 (BD) and FITC anti-human CD31 (BD) were used to label EMPs.

Results: In the present research, 40 MS patients, including 4 men and 16 women in the G1 and 6 men and 14 women in G2 were studied. The mean age of patients in G1 was 31.3 $\pm$ 8.3 years (48-20), and 32.1 $\pm$ 8.6 years (18-46) in G2. Following treatment, the level of CD31+ MPs in both G1, and G2 groups decreased by 40% and 31%, respectively. Also, the level of CD51+ MPs decreased by 32% and 23%, respectively, which were statistically significant (p less than 0.001).

Conclusion: The results of this study show that INF β and glatiramer acetate have inhibitory effects on the release of EnMPs, so they can be considered as sensitive biomarkers for disease progression and treatment effectiveness.

Keywords: Multiple sclerosis; Interferon-beta; Glatiramer acetate; Endothelial cell-derived microparticles.

Investigation of Expression of MiR-214 and MiR-30c-2* and also Their Role in Resistance to Proteasome Inhibitor (PI) Drug in Resistance Multiple Myeloma Cells to This Drug

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Background and Aim: Multiple myeloma is the second most common blood cancer. The endoplasmic reticulum plays an important role in the folding of proteins. With an overgrowth of unfolded and malformed proteins, the cell goes to death (apoptosis) via the UPR (Unfolded protein response) pathway. Various genes and microRNAs are involved in this pathway. The most important UPR pathway protein is the XBP1 protein, which is directly affected by two microRNAs called miR-214 and miR-30c-2*. Because the UPR pathway is activated in resistant cancer cells by increasing cellular stress, these microRNAs reduce the expression of the XBP1 gene by inactivating it, inactivating the UPR pathway, and then resulting in the survival of resistant cancer cells. The present study was performed to compare the expression of XBP1, miR-214, and miR-30c-2 * genes in multiple myeloma cells that are sensitive and resistant to bortezomib.

Methods: In order to produce bortezomib-resistant cell line, drug-sensitive multiple myeloma cells were exposed to concentrations of 2.5, 5, 10, 20, and 40 nM of bortezomib (BTZ). To confirm the resistance of the cells, MTT assay test was performed between two sensitive cells and resistant to bortezomib. In this way, both cell groups were treated with concentrations of 2.5, 5, 10, 20, 40, and 80 nM of bortezomib. Then, by the MTT Assay method, IC₅₀ of both cell groups was obtained. The IC₅₀ of resistant cells was higher than that of sensitive cells, indicating that the cells were resistant. Then, the relative expression of XBP1, miR-214, and miR-30c-2 * genes between resistant and sensitive cells was measured by the Real Time-PCR method.

Results: Our study showed that the relative expression of the XBP1 gene in cells resistant to susceptible cells is significantly reduced. However, the expression of miR-214 and miR-30c-2 * genes were not significant in the group of resistant cells relative to the group of sensitive cells to bortezomib.

Conclusion: From the above information, it can be concluded that other microRNAs may affect the expression of the XBP1 gene and, by lowering the XBP1 gene and affecting the UPR pathway, that leads to multiple myeloma cells becoming resistant to the drug bortezomib.

Keywords: Multiple myeloma; Bortezomib; Drug resistance; miR-214; miR-30c-2; UPR

Lymphopenia as a Prediction Marker for Prognosis of COVID-19 Patients

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Background and Aim: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) rapidly spreads out around the world since December 2019 and became a pandemic. SARS-CoV-2 causes coronavirus disease-19 (COVID-19). As of 6 December 2021, 266,264,966 cases and 5,273,756 deaths have been reported. COVID-19 was recently considered a communicable systemic disease that affects various parts of the body such as hematopoietic system. The aim of this study is to review lymphopenia, as a COVID-19 complication, and its predictive potential for disease prognosis.

Results: Lymphopenia, decreased number of lymphocytes in the blood, is one of COVID-19 complications. In acute conditions, the immune system intensely confronts virus infection by immune cells and the production of cytokines. In the chronic state, natural killer and lymphocyte T cells get exhausted that resulting in a reduction in their number i.e., lymphopenia. In this condition, the infection cannot be eliminated by exhausted immune cells. Immune system compensates for this inability through excessive production of pro-inflammatory cytokines named cytokine storm syndrome. B and T cells apoptosis rate increases significantly in COVID-19 patients, and T cells apoptosis rate rises with the disease severity. Studies have suggested that the induction of early apoptosis in lymphocytes is the main cause of lymphopenia. Lymphopenia has been reported in 85% of patients with severe condition. Lymphopenia on admission and in death cases illustrates its clinical importance as a prediction marker for the prognosis of COVID-19 patients.

Conclusion: The measurement of lymphocyte count in COVID-19 patients can be used as an important factor for the prediction of disease outcomes.

Keywords: COVID-19; SARS-CoV-2; Lymphopenia; Prognosis

Magnesium Supplementation May not be Protective Against Carboplatin Induced Nephrotoxicity but May be Beneficial for Children Suffering Malignancies: A Randomized Clinical Trial

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Background and Aim: Magnesium oxide is proposed to be effective in the prevention of the nephrotoxic impact of carboplatin therapy. The aim was to evaluate the magnesium oxide supplementation effect on serum creatinine, blood urea nitrogen (BUN), and glomerular filtration rate (GFR) in cancerous children

Methods: 18 children were treated with 250 mg/day magnesium oxide and the other 18 children with placebo for 14 days before the onset of carboplatin chemotherapy based on the routing protocols in oncology. Serum creatinine (Cr), BUN, and GFR were measured and compared before, on days 3 and 7 of magnesium carboplatin therapy

Results: There were 7 (38.89%) and 12 (66.67%) male patients in the treatment and control groups, respectively. Serum Cr and BUN were increased significantly 3 and 7 days after intervention in both groups. There was an insignificant difference for serum Cr and BUN between treatment and control groups before the intervention, 3 days or 7 days after carboplatin administration onset ($p > 0.05$). The GFR was reduced significantly from 101.38 ± 14.67 to 90.11 ± 10.52 mL/min/1.73m² in treatment and from 97.5 ± 9.71 to 92.33 ± 10.61 mL/min/1.73m² in the control group. After 7 days the GFR was reduced to 84.11 ± 12.47 mL/min/1.73m² in treatment and 85.38 ± 10.66 mL/min/1.73m² in control group ($p = 0.371$).

Conclusion: Current study shows that magnesium supplementation did not prevent carboplatin-induced nephrotoxicity in cancerous children. Anyway, we propose magnesium oxide supplementation for such patients because magnesium is an essential element for cell and tissue growth, maintenance, and metabolism.

Keywords: carboplatin; magnesium; nephrotoxicity; pediatrics

PEAR1 Genetic Variants in Essential Thrombocythemia: The Prevalence and Association with Hematological Parameters and ET Mutations

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Background and Aim: Essential thrombocythemia (ET) is a type of myeloproliferative neoplasm characterized by the expansion of the megakaryocytic/platelet line. Given the undeniable role of genetic variations in the pathogenesis of ET, as well as the proven effects of PEAR1 SNPs on platelet function, the innovative purpose of this study is to investigate the prevalence of PEAR1 variants (rs12041331 and rs12566888) and their relationship to hematological parameters and ET-related mutations.

Methods: We studied 105 ET patients and analyzed ET patients' mutational profiles, including JAK2 V617F mutation (detected by Allele-specific PCR), CALR, and MPL mutations (both through PCR amplification). Two SNPs of the PEAR1 gene were assessed through ARMS-PCR, and the Sanger method was used for the validation of ARMS-PCR amplification.

Results: The prevalence of rs12041331 and rs12566888 in ET patients were 43.9% and 38.5%, respectively, and rs12041331 was significantly associated with increased platelet counts (P-Value: 0.02). As expected, the incidence of thrombotic events in JAK2 + patients was high and significantly associated with JAK2 mutation (P-Value: 0.02). The prevalence of thrombotic events was also high in patients with the rs12041331 variant. Besides, a significant relationship was also found between the rs12041331 and CALR mutation (P-Value: 0.03).

Conclusion: In recent years, the footprint of the PEAR1 variant's effect on platelet aggregation led to evaluating these variants in ET patients. Finally, the significant relationship between the rs12041331 variant and increased platelet count and CALR mutation announced that the idea of this study could be pursued and challenged in the future.

Keywords: Essential Thrombocythemia; platelet endothelial aggregation receptor 1; polymorphisms; platelet; Janus kinase 2; ARMS-PCR; Prevalence.

Platelet and Coagulation Factors Change in SARS-CoV-2 Patients

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Background and Aim: There are few guidelines for interpreting and treating hematologic problems in COVID-19 patients. We intend to publish the results of coagulation tests and platelet counts of COVID-19 patients, in the hope that this knowledge will be useful to all physicians, laboratory scientists, and researchers.

Methods: We have studied 90 articles on hematologic changes in Covid-19 patients and information on coagulation and platelets is included in this article. This study was compiled using reports retrieved from PubMed, Google Scholar, Islamic Citation and Magiran databases from 2019 to 2021. "Hematological parameters", "coagulation" and "platelets

Results: COVID-19 patients usually show mild and platelet intake appears to be increased. Viruses can potentially inhibit platelet production by interacting with megakaryocytes. Patients with thrombocytopenia had higher rates of morbidity and mortality during the epidemic. Severe thrombocytopenia has only rarely been described in combination with a disease similar to immunologic thrombocytopenic purpura. Platelets are involved in inflammatory signals as well as the immune response induced by Toll-like receptors. COVID-19 infection initially causes a significant increase in fibrinogen degradation products and D dimer/fibrin. This is associated with systemic hypercoagulation and high rates of venous and arterial thromboembolism. In patients with Covid-19, increased D-dimer rates are associated with mortality, DIC, and serious bleeding events are rare. Coagulation is most likely the product of a significant inflammatory response to COVID-19 and endothelial activation/damage. Active or damaged endothelial cells, tissue factor expression, Weibel-Palade body release, which contain very large multimers with vWF molecular weight, and coagulation cascade activation are all associated with COVID-19 pneumonia. In COVID-19 individuals, the activity of ADAMTS13 is somewhat reduced, and therefore very large vWF multimers are increased, as well as the FVIII clotting activity is increased and the platelets bind spontaneously, causing microthrombosis.

Conclusion: responses exacerbate oxygen delivery problems, and local hypoxia produces a positive inflammatory thrombus feedback loop. Covid-19 has prolonged aPTT and/or PT prolongation, hypofibrinogenemia is uncommon, and laboratory data support hyperfibrinolysis. Higher platelet counts during treatment were associated with longer hospital stays, or with increased platelet-to-lymphocyte ratio.

Keywords: COVID-19; Laboratory Hematology; Platelet; Coagulation Factor

Platelet-Related Indices as Useful Prognostic Markers in Severe Patients with Covid-19

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Background and Aim: Coronavirus disease 2019 (Covid-19) is quickly spreading all over the world. This infectious disease results from severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in lung tissues. In most cases, manifestations are relatively mild to moderate; however, some patients develop to severe stages of the disease, presenting acute respiratory distress syndrome and septic shock. The elderly or comorbid patients experience severe conditions versus the general population. Interestingly, thrombosis is a common finding in Covid-19 patients which is contributed with some hematologic indices. We aimed to review the association of Platelet-related indices such as mean platelet volume (MPV), platelet distribution width (PDW), and platelet-large cell ratio (P-LCR), with the mortality in Covid-19 patients.

Results: Laboratory results indicated some thrombotic events during Covid-19 hospitalization. Platelets play critical roles in the stimulation of thrombosis by modulation of inflammatory and pro-coagulatory cytokines. Platelet-related parameters such as MPV, PDW, and P-LCR are widely used for diagnosis and prognosis in Covid-19 patients. MPV is a marker for the determination of platelet function and activation. Larger platelets with high MPV value are functionally more active, and produce a high amount of thromboxane A₂ (TXA₂), as a prothrombotic factor. The increased concentrations of TXA₂ enhance thrombotic complications. Furthermore, MPV is also contributed to pro-inflammatory cytokines. PDW shows the distribution in platelets size. During infection, the degradation of platelets alongside the production of immature platelets increases PDW since new platelets are larger with various sizes. Additionally, studies reported a substantial increase in the P-LCR index, the mean percentage of platelets with high volume, in hospitalized patients with poor outcomes.

Conclusion: MPV, PDW, and P-LCR indices at hospital admission can distinguish Covid-19 patients with high risk for thrombotic complications, allowing preventive strategies.

Keywords: COVID-19; Prognosis; mean platelet volume; PDW; P-LCR; thrombosis

Portal Vein Thrombosis Might Develop by COVID-19 Infection or Vaccination: a Systematic Review of Case-Report Studies

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Background and Aim: Infection by the novel coronavirus (COVID-19) has been associated with different types of thrombotic complications same as portal vein thrombosis (PVT). However, by emerging COVID vaccines, the thrombosis did not seem to be concerning anymore. Until new findings showed that, the COVID vaccine itself can cause PVT.

Methods: We performed an electronic search in PubMed, Scopus, and Web of Sciences to evaluate the possibility of occurring PVT due to COVID-19 infection and vaccination. The results were reported in a narrative method and categorized into tables.

Results: Overall, 40 cases of PVT from 34 studies were reviewed in this article. The prevalence of PVT following COVID-19 was more remarkable in males. However, it was more common in females after COVID-19 vaccinations in the reviewed cases. regardless of etiology, 20 of PVT cases reviewed in this article, had at least one comorbidity. The most common clinical presentation was abdominal pain. After anticoagulant therapies, most of the patients improved or discharged.

Conclusion: As long as the laboratory findings are not appropriate enough to predict PVT, the diagnosis of this complication with whatever underlying reason are challengeable while rapid diagnosis and treatment of that is vital. Therefore, by providing available data in an organized way, we aimed to prepare the information of infected patients for better and easier future diagnosis of PVT in new cases.

Keywords: COVID-19; Vaccines; Liver Diseases; Portal Vein; Venous Thrombosis

Potential Use of Umbilical Cord Mesenchymal Stem Cells for the Improving of COVID-19 Patients

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Background and Aim: The SARS-CoV-2 virus is a member of the coronavirus family that caused the COVID-19 respiratory disease epidemic in China before the global pandemic of the disease in late 2019. The virus's genome is of 79% similarity to that of the SARS-CoV virus, using the ACE2 receptor to enter its target cells. The most common symptoms of this disease include fever, cough, pulmonary involvement, and sometimes gastrointestinal symptoms. The aim of this review study, use the therapeutic potential of umbilical cord mesenchymal stem cells in COVID-19.

Methods: Our study was based on PUBMED and GOOGLE SCHOLAR databases from December 2019 to 2021 by related keywords such as: "COVID-19", "SARS-CoV-2", mesenchymal stem cells", "umbilical cord" and "Immunomodulatory effects". Also was searched with the keywords: "mesenchymal stem cells" and "umbilical cord" without a time limit.

Results: A decline in both the number and function of lymphocytes and a severe increase in leukocyte inflammatory activity are among the most obvious immunological complications of this disease. If the immune system response to the virus is inadequate, the disease can become acute. Immune cells activity leads to a sharp increase in the number of blood cytokines, causing cytokine storm, which in turn can cause systematic damages to the heart, lungs, and kidneys, and ultimately may lead to death. Mesenchymal stem cell therapy offers a promising approach to reducing the destructive impacts of infection in COVID-19 patients. Mesenchymal stem cells can secrete immune-modulating factors that suppress cytokine storms.

Conclusion: Furthermore, the role of mesenchymal stem cells in preventing cell death and inhibiting tissue fibrosis has been well demonstrated. This review shows available clinical trials that have tapped into the therapeutic potential of the umbilical cord mesenchymal stem cells in COVID-19 patients.

Keywords: mbilical cord; Mesenchymal stem cell; COVID-19; SARS-CoV-2; Immunomodulatory effects; Clinical trials

Relation of Vitamin D Status to Hematologic, Biochemical and Serological Parameters in COVID-19 Patients

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Background and Aim: From December 2019 severe acute respiratory syndrome coronavirus (SARS-COV-2) infection has been the most challenging pandemic in the world (1, 2). Almost all countries are affected by the disease and there is no definitive treatment for COVID-19 infection (3). Recent studies show the disease severity in the geographic region that have more vitamin D deficiency and suggested the acceptability of vitamin D prophylaxis therapy, especially in where the deficiency is more prevalent (4, 5). The aim of this research was to find the relation between serum vitamin D level and laboratory tests including ALP, AST, AST, ferritin, CBC, PT, PTT, D-dimer and death in the patients with positive RT-PCR for SARS-CoV-2 infection in a teaching hospital, Dezful, Khuzestan, Iran (IR.DUMS.REC.1400.035).

Methods: In this cross-sectional study additionally, the demographic and comorbidity information were taken, the parameters were measured by conventional methods. The statistical analysis compared the level of deficiency and insufficiency among different groups of COVID-19 patients.

Results: Of 50 patients, the median age was 56.8 years (33-70 years), and 28 of 50 (56%) patients were men. The total prevalence of vitamin D deficiency was 25% and insufficiency was 40% respectively. A significant relation between d-dimer level and the level of patient vitamin D and death was determined compare to discharge group (P less than 0.05).

Conclusion: Vitamin D deficiency is related to the fatality rate of SARS-CoV-2 infection in various regions of words, implying that adjuvant. therapy with vitamin D may reduce the mortality rate of COVID-19 patient

Keywords: Vitamin D; COVID-19; SARS-CoV-2; Clinical Outcome

Role of Interleukin-21 in Aplastic Anemia

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Background and Aim: Interleukin (IL)-21 is a pro-inflammatory autocrine cytokine that is mainly produced by T cells (CD4+) and plays an important role in the immune system. This cytokine modulates the balance between T-helper (Th)-17 and T regulatory (Treg) cells. Aplastic Anemia (AA) is a rare disorder characterized by pancytopenia of the peripheral blood and hypocellular bone marrow. Studies show that dysfunction of Th17 and Tregs cells (CD4+CD25hiFoxP3+) is involved in the pathophysiology of AA. Therefore, IL-21 can play a role in the pathogenesis of this disease.

Methods: For this study, the PubMed database in all available years was examined for the association of IL-21 with aplastic anemia. The keywords "IL-21", "aplastic anemia" and "T regulatory" were used for this study. Inclusion criteria were only original articles related to the topic and no review, systematic review and, meta-analysis articles were selected.

Results: Increased IL-17 expression has been found in AA. Due to the fact that IL-21, as an autocrine cytokine for the differentiation of Th17 in AA, is produced more than usual and significantly increases the production of IL-17. Thus IL-21 directs Th17 immune responses in AA. CD4 + T cells that produce IL-21 are inversely related to Tregs in AA. In addition, IL-21 acts against FoxP3 expression in cultured CD4+ T cells of AA patients. Thus, IL-21 reduces Tregs and impairs their suppressive function in AA. Therefore, IL-21 is one of the cytokines that can be responsible for the overactivity of T cells in the BM of AA patients.

Conclusion: In general, IL-21 alters the balance between Th-17 and Treg cells by enhancing T cell proliferation and impairs Tregs suppressive function, and contributes to the pathogenesis of AA.

Keywords: "IL-21; aplastic anemia; T regulatory; Th-17"

Routine Hematologic Laboratory Testing as a Predictive and Prognostic Parameter in Covid-19 Pandemic

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Background and Aim: To date, there is more than 150 million people has infected with the Corona virus disease (covid-19) all around the world. The variety in infected people symptoms has great impact on choosing appropriate possible treatments such as antiviral agents, antibiotic drugs hyper immune immunoglobulin, etc. Thus there is need for accurate clinical and laboratory prognostic parameters to evaluate disease severity. The aim of this study was to determine laboratory and in specific Hematological laboratory parameters as a predictive and diagnostic marker for covid-19. For this goal, routine Hematological data of 42 covid-19 patients obtained from Imam Reza Hospital of Tabriz. Then obtained data amalgamated for harvesting new unseen information. The results of this study showed that activated Partial Thromboplastin Time (aPTT) can be used as criteria for patients' hospitalization in the ICU as well as predicting the outcome of patients. The use of such Hematological parameters also helps to prevent the spread of the disease because aPTT is easily evaluated in the early stages of the disease and is preferable to Ct-scan, that only can be evaluated after lung infection occurrence. Overall, the results of this study showed predictive importance of routine Hematological data for evaluating covid-19 severity.

Methods: This study designed as a retrospective cohort and required data collected between December and January 2020 from Imam Raza Hospital of Tabriz at the third wave of covid-19 in Iran. Ethical permission obtained from Tabriz University of Medical Science Ethical Committee (IR.tbzmed.rec.1399-8213). The samples included 42 patients with confirmed covid-19 by RT-PCR analysis of nasopharyngeal swab specimens, including 20 critically ill patients admitted to the ICU and 22 patients with stable conditions as control group. During the study, out of 22 patients with stable conditions, 8 patients were discharged from the hospital, 3 were still hospitalized, no deaths occurred and the data of 1 patient was not recorded. Out of a total of 20 patients with serious conditions, 13 patients were discharged from the hospital, 5 patients died and 2 were still hospitalized. Information collected from patients includes characteristic as well as hematological data extracted from routine hematological tests. The characteristics and hematology variables studied were included Age (years), gender (n %), D-Dimer (mg/L) (0.0–0.5), Fibrinogen (g/L) (1.5–4.0), aPTT (s) (25.0–36.5), PT (s) (10.4–13.0), P-LCR (%), PCT (%). Data amalgamation was done by SAS Viya (3.5) software.

Results: The results of study show a higher mean age of severe patients (68.74 ± 16.9 ; $p = 0.042$) compared to stable patients (56.7 ± 9.94). In addition, there is no significant difference in the gender proportion between the two groups, so that in both severe and stable patients, women's account for

52%, 57% and men's for 58%, 53% of total patients in both groups respectively. Also, after examining the hematological variables, it was revealed that there is a sharp increase in D-dimer level in severe patients group (5.65 ± 5.45 mg/L) compared to the stable group (0.98 ± 0.67 mg/L). In addition, a slight increase in aPTT was observed in severe patients group (33.08 ± 8.35 s) compared to the stable group (28.33 ± 1.85 s). However, no significant change was observed in Fibrinogen and PT between the two groups. The analyzing variables by SaS Viya indicates the importance of aPTT in predicting severity in covid-19 patients and values of >30.9 determined as a predictive warning limit of severity in covid-19 patients. The results clearly showed that the aPTT parameter can be useful for predicting the outcome of covid-19 patients, which is inexpensive, easily accessible and also does not require experienced staff.

Conclusion: In this cohort study, the potential of using routine hematological data to predict the severity of covid-19 patients was demonstrated. Recent studies have used CT-scan to assess the severity of the disease, while such tests can be measured only after the onset of the patient's lung infection and do not seem very useful to prevent the spread of the infection to vital organs and it is necessary to predict the progression of disease faster. In this regard, hematological parameters that are measured in the early stages of patient's hospitalization can help to detect possible progression more quickly and prevent its risks. Based on our findings, prolonged aPTT can be considered as one of the indicators of patient admission, but our small study dataset (42 patients) should be considered. Also, caution should be exercised in cases where the patient has taken anticoagulants. One of the limitations we encountered in this study, in addition to small sample size, is pre-diagnosis death or pre-diagnosis improvement in patients, which calls into question the comprehensiveness of the results and their generalizability throughout society. Despite these limitations, this study still has valuable information on potential ability of Hematological parameters to predict the severity of covid-19 disease.

Keywords: covid-19; hematology; aPTT; predictive marker

Study of Immunohematologic Adverse Reaction Blood Transfusion in Fasa Valiasr

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Background and Aim: Any adverse reaction to the Transfusion of Blood components should be reported to Blood bank personnels as soon as. Because each blood product transfused carries an immunohematology small risk of an acute or late adverse effect.

Methods: This descriptive-analytical study was carried out of Fasa Valiasr hospital. 1450 units of Blood and Blood components were transfused, a date was retrieved during 12 months from the Blood Bank unit of this center. Alloimmune transfusion reactions, platelet refractoriness, hemolytic of RBCs and Bacterial contamination reaction were studied.

Results: From 1450 units of Blood and Blood components, 9 cases of adverse reaction are reported and the following results we obtained: 1- 3 positive direct antiglobulin test (DAT) 2-3 positive hemoglobinuria 3-2 cases of thrombocytopenia. 4- No bacterial contamination reactions.

Conclusion: The present finding shows that with continue reporting Blood transfusion-associated an adverse event to blood bank. Blood transfusion organizations help to diagnosis, Therapy and preventing of adverse reactions.

Keywords: Blood Transfusion; Blood Banks; Hemoglobinuria

Study on Hypoxia-Inducible Factor and Its Roles in Immune System

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Background and Aim: The Hypoxia-Inducible Factor-1 (HIF-1) is a dimeric protein complex that plays a significant role in responding to low oxygen or hypoxia concentrations. Chronic inflammation is one of the immune system responses and can increase HIF expression in involved tissues through lowering oxygen and hypoxia. The HIF factor has many critical roles in immunity, and thus, we reviewed the crucial roles of this factor in the immune system. The results showed various key roles on the immune system, including physical defenses, innate immune (neutrophils apoptosis, macrophages) and inflammatory responses (pyrexia and local heat, iron access, etc.), upregulation in response to microbial infections, cytokines expression (IL-1, IL-2, IL-6, IL-8, IL-12, IL-18, TNF, etc.), drug targeting, etc. The HIF roles in the acquired immune system include: enhance the adaptation of cells (dendritic cells) to new conditions and triggering the signal pathways. The findings of the present review demonstrated that the HIF has important roles in the immune system, including physical defense, innate immune as well as acquired immunity; therefore, it may be considered as a potent drug targeting several diseases such as cancers, infectious diseases, etc.

Methods: To identify all relevant literature, a review was conducted on studies found on such online databases as Google Scholar, PubMed, and Scopus by Internet-based search for Hypoxia-Inducible Factor, innate immunity, immunodeficiency, inflammation, infections profiling studies published between 2005 and 2018. Our search strategy aimed to identify all studies published in English that reported results of only immunodeficiency studies associated with Hypoxia-Inducible Factor that presented essential results. The literature search identified 116 potentially relevant articles. After exclusion of irrelevant or duplicate articles by reading titles and abstracts, 87 articles were retrieved for further evaluation.

Results: The results showed various key roles on the immune system, including physical defenses, innate immune (neutrophils apoptosis, macrophages) and inflammatory responses (pyrexia and local heat, iron access, etc.), upregulation in response to microbial infections, cytokines expression (IL-1, IL-2, IL-6, IL-8, IL-12, IL-18, TNF, etc.), drug targeting, etc. The HIF roles in the acquired immune system include: enhancing the adaptation of cells (dendritic cells) to new conditions and triggering the signal pathways.

Conclusion: The findings of the present review demonstrated that the HIF has important roles in the immune system, including physical defense, innate immune as well as acquired immunity; therefore, it may be considered as a potent drug targeting several diseases such as cancers, infectious diseases, etc.

Keywords: Hypoxia-inducible factor; innate immunity; immunodeficiency; inflammation; infection

The Apoptotic Effect of Cassiopea Andromeda Crude Venom on the Acute Lymphoblastic Leukemia Cell Line(Jurkat)

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Background and Aim: Acute lymphoblastic leukemia (ALL) is the most common childhood malignancy, accounting for only 77% of all childhood leukemias. Chemotherapy used in leukemia treatment can also have adverse side effects in patients. Study on the Biochemical, toxicological, and pharmacological characteristics of venoms has attracted the attention of researchers to search for new sources of medicine. Hemolytic, anti-microbial, cardiovascular, insecticidal, antioxidant, and cytotoxic effects of jellyfish venoms have been revealed. Cassiopea andromeda (Cnidaria, Rhizostomeae, Scyphozoa), so-called upside-down jellyfish, is a venomous jellyfish. This study aimed to evaluate the apoptotic effect of Cassiopea andromeda crude venom on the acute lymphoblastic leukemia cell line(Jurkat).

Methods: the apoptotic effect of the crude venom on the Jurkat cell line was assessed by Flowcytometry assay using Annexin-V-fluorescein isothiocyanate (FITC) and propidium iodide(PI) stains. in brief, Jurkat cells were seeded in a 12-wells plate at a density of 5×10^5 cells/well and incubated for 24h at 37°C. then the cells were treated with crude venom (500µg/mL; IC₅₀ in 48h). after that, the cells were washed twice with cold PBS, resuspended in binding buffer, and stained with Annexin V and PI, and incubated for 15 min in the dark at room temperature. Ultimately, the samples were analyzed using a flow cytometer. The cells were regarded as viable (annexin V-/PI-), early apoptotic (annexin V+/PI-), late apoptotic (annexin V+/PI+), and necrotic (annexin V-/PI+). Data analysis was done by the FlowJo software.

Results: Based on Flowcytometric analysis, about 90%of control cells were annexin V-/PI- (viable). about 55% of treated cells were annexin V+/PI- and annexin V+/PI+ which represented the cells in early and late apoptosis respectively.

Conclusion: These results showed the apoptotic effect of the crude venom from Cassiopea Andromeda on the Jurkat cell line. So this venom can be used in further Pharmaceutical studies in the field of cancer treatment.

Keywords: Cassiopea andromeda: acute lymphoblastic leukemia; Jurkat: apoptotic

The Effect of Cardiopulmonary Bypass on Platelet-Derived Microparticles Levels and Platelet Counts in Patients Undergoing Heart Valve Surgery

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Background and Aim: Most valvular heart disease (VHD) patients require heart surgery under cardiopulmonary bypass (CPB). Platelet-derived microparticles (PMPs) are small membrane vesicles with a size ranges from 0.1-1 μ m. PMPs originate from platelets through activation and apoptosis. In this study, we investigated the effect of CPB on platelet counts and PMPs levels in patients undergoing heart valve surgery.

Methods: This study was performed on forty adult patients undergoing heart valve surgery with CPB at shaheed rajaie cardiovascular Center. Plavix was discontinued 5 days prior to surgery and warfarin was discontinued 72 hours before surgery. Patients with known bleeding disorders and emergency surgery were excluded. Before skin incision and 30 minutes after administration of protamine sulfate, 6ml venous blood samples were collected. MPs were isolated with centrifugation. Total concentration of MPs was determined using Bradford method. Flow cytometry analysis was performed to determine PMPs levels using CD41-FITC and yellow-green microbeads with 1 μ m in diameter. The Wilcoxon signed-rank test was applied to compare pre- and postoperative PMPs levels and platelet counts. The association between the variables was assessed through employing Spearman's rank correlation analysis. Statistical significance was considered as P less than 0.05.

Results: The post-operative MPs concentration and PMPs levels were increased significantly than the preoperative stage (P less than 0.0001). The post-operative platelet counts were decreased significantly than the preoperative stage (P less than 0.0001). The MPs concentration was significantly and positively correlated to CPB time ($P=0.03$, $\rho=0.4$). But no significant relationship was observed between PMPs levels and CPB time (P more than 0.05). The platelet counts were significantly and negatively correlated to CPB time ($P=0.04$, $\rho=-0.3$).

Conclusion: Platelet count declined by increasing PMPs levels. This decrease seems to indicate the activation of platelets due to blood contact with the CPB circuit and their consumption with PMPs formation.

Keywords: Cardiopulmonary bypass; Platelet; Cell-Derived Microparticles

The Effect of Convalescent Plasma Therapy on COVID-19 Patients: A Retrospective Study

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Background and Aim: SARS-COV2 virus was first detected in December 2019 in Wuhan, China. Different treatments have been used to fight against virus, but one of the most effective and accessible is Convalescent Plasma therapy (CPT). Therefore, in the present study we investigated the effect of CPT on improving the clinical conditions of Corona Virus Disease 2019 (COVID-19) patients.

Methods: In this study 440 individuals participated, including 220 COVID-19 recovered individuals as plasma donor and 220 plasma recipients. Sampling method in this study is random sampling and the above study was conducted in the period from May 2020 to February 2021 in Shiraz. After complete recovery from COVID-19, Patients were recalled for apheresis and plasma isolation for Convalescent Plasma (CP) preparation; they were referred to blood donation centers of Shiraz city, Fars province. According to the hospital request, compatible and related plasma was sent for COVID-19 hospitalized patients. At the same time, hospitals were asked to send the clinical condition document of the patients based on the medical records and the outcomes after CPT; blood transfusion organization evaluated the results.

Results: Median antibody titer was 1/300; titers higher than 1/300 were considered as high, and the titers lower than 1/300 were considered as low. Blood group O had 54 times higher antibody titers above 1/300 compared to other blood groups (OR:54, CI:19.5-205.3, P less than 0.001).

Conclusion: In this study, it was shown that patients with COVID-19 who received plasma with a titer above 1/300 and a minimum volume of 387 ml had minimal clinical symptoms and were able to be discharged from the hospital. Since blood group O plasma has a higher neutralizing antibody titer compared to other blood groups. Therefore, the use of blood group O plasma with a minimum volume of 387 ml can be very effective in improving the clinical symptoms of patients with COVID-19 and increase the chances of survival for these patients.

Keywords: Blood Donors; COVID-19; Plasma; Therapeutics

The Effect of SARS-COV-2 Infection on RDW Red Blood Cells in COVID-19 Patients

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Background and Aim: Since the onset of COVID-19 in December 2019 Comparison of hematological test results with different outcomes showed that red cell distribution width (RDW) may be a predictor of prognosis in patients with severe COVID-19. Epidemiological and clinical data showed that patients with poor outcome were older than patients with good outcome. The higher RDW is, the more significant the heterogeneity in RBC volume.

Results: RDW is a low-cost parameter which increases in response to many acute and chronic proinflammatory conditions. The results also showed that the number of RETs in critical patients was significantly higher than the mild patients. Because the size of an RET is more important than an adult RBC, increasing RETs may increase RDW. This may be due to increased erythropoietin synthesis and active erythroid hyperplasia due to prolonged hypoxia and or due to malnutrition and iron deficiency. The findings show that the increase in RDW is not only due to the increase in the number of RETs in patients with COVID-19 with poor outcome. Also, oxidative stress levels and insufficient nutrient circulation in patients with poor outcome may lead to increased RBC membrane instability and further increase RDW and RETs and decrease MCV in patients with poor outcome. SARS-CoV-2 infection can cause structural changes in lipids and proteins in RBC membranes. Thrombosis is commonly seen in patients with severe Covid-19, which may lead to RBC damage and morphological abnormalities. However, there is no agreement on the optimal cut for RDW (from 12.5% to $\leq 16\%$). It should also be noted that any abnormalities such as thyroid disease, kidney or liver dysfunction.

Conclusion: Finally, RDW, along with other inflammatory agents, can provide accurate information about the inflammatory status and prognostic symptoms of patients.

Keywords: SARS-CoV-2; COVID-19; red cell distribution width (RDW); MCV

The Effects of Anabolic-Androgenic Steroids on DNA Damage in Bodybuilders' Blood Lymphocytes

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Background and Aim: Nowadays bodybuilders use anabolic steroids frequently. Abuse of these substances can cause significant side effects; therefore, we aim to investigate the effect of anabolic steroids on DNA damage in bodybuilders' blood lymphocytes.

Methods: This case-control study was performed on 36 male bodybuilders in Gonabad. The case group included bodybuilders with a history of taking anabolic-androgenic steroids (n=18), and the control group composed of bodybuilders who did not use anabolic-androgenic steroids (n=18). Intravenous blood samples were obtained and then the lymphocytes, cells and electrophoresis of blood were extracted. Afterward, the coloured slides and DNA damage were measured using a fluorescent microscope and Comet Score software. The comet assays analysis was carried out under alkaline conditions, basically as described by Singh et al., with minor modifications (Singh et al. 2010). Electrophoresis was carried out for 20 minutes at 0.65 V/cm and 300mA to allow the damaged DNA to migrate towards the anode. After electrophoresis, the slides were rinsed gently three times with a neutralisation buffer (0.4M Tris-HCl, pH 7.5) to remove excess alkali and detergents and then fixed with methanol. Each slide was stained with ethidium bromide (20 lg/ml) for five minutes and finally dipped in ice cold water to remove the excess ethidium bromide (EtBr) and covered with a coverslip. It is worth mentioning that hydrogen Peroxide exposure was used as a positive control. The DNA damage was compared using t-tests.

Results: Results showed that there was no significant difference between ages, marital status, BMI, systolic and diastolic blood pressure in the case and control group. However, parameters related to the DNA damage including tail length, percent tail DNA, and tail moment were significantly higher in the case group.

Conclusion: The use of anabolic-androgenic steroids increases DNA damage in the bodybuilders' blood lymphocytes.

Keywords: Anabolic-androgenic steroids; DNA damage; Bodybuilders; Comet assay; Blood lymphocytes

The Expression Level of CCDC26 and FOXCUT Genes in Acute Lymphoblastic Leukemia

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Background and Aim: Background and Aim: Acute lymphoblastic leukemia (ALL) is a malignant disease of lymphoid progenitor cells affecting both children and adults. Long non-coding RNAs (lncRNAs) are one kind of non-coding RNAs (ncRNAs), reported modulating the initiation or progression of diverse cancers. However, the role of CCDC26 and FOXCUT long non-coding RNAs in ALL has been unknown. In this study, we explored the expression of FOXCUT and CCDC26 lncRNAs in acute lymphoblastic leukemia cell lines.

Methods: Methods: Acute T lymphoblastic leukemia cell lines, RPMI 8402, Jurkat, B lymphoblastic leukemia, Daudi, and Ramos cell lines were used. After culturing the cells, RNA extraction and cDNA synthesis were performed. The real-time PCR technique was then used to study the expression of CCDC26, FOXCUT, C-kit, and FOXC1 genes.

Results: We found a significant increase of CCDC26 expression in RPMI 8402 (p-value is less than 0.0001) and Ramos (p-value is less than 0.05) cell lines compared to the control, while decreased expression of these genes was observed in Jurkat and Daudi cell lines. Furthermore, FOXCUT gene had a significant increase in expression in all cell lines compared to the control (p-value is less than 0.01 in Daudi and RPMI 8402 cell lines) (p-value is less than 0.001 in Jurkat and Ramos cell lines).

Conclusion: Our results demonstrated that CCDC26 and FOXCUT genes can play a regulatory role in acute lymphoblastic leukemia and may serve as a potential diagnostic biomarker and therapeutic target of acute lymphoblastic leukemia.

Keywords: Acute Lymphoblastic; Leukemia; Long Non-Coding RNA CCDC26 C-Kit FOXCUT

The Potential Similarities of COVID-19 and Autoimmune Disease Pathogenesis and Therapeutic Options: New Insights Approach

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Background and Aim: Cytokine pathways and their signaling disorders can be the cause of onset and pathogenesis of many diseases such as autoimmune diseases and COVID-19 infection. Autoimmune patients may be at higher risk of developing infection due to the impaired immune responses, the use of immunosuppressive drugs, and damage to various organs. Increased secretion of inflammatory cytokines and intolerance of the patient's immune system to COVID-19 infection are the leading causes of hospitalization of these patients.

Methods: The content used in this paper has been taken from English language articles (2005–2020) retrieved from the PubMed database and Google Scholar search engine using “COVID-19,” “Autoimmune disease,” “Therapeutic,” “Pathogenesis,” and “Pathway” keywords.

Results: The emergence of COVID-19 and its association with autoimmune disorders is a major challenge in the management of these diseases. The results showed that the use of corticosteroids in the treatment of autoimmune diseases can make the diagnosis and treatment of COVID-19 more challenging by preventing fever.

Conclusion: Due to the common pathogenesis of COVID-19 and autoimmune diseases, the use of autoimmune drugs as a possible treatment option could help control the virus.

Keywords: Autoimmune disease; COVID-19; Pathogenesis; Pathway; Therapeutic

The Role of Neutrophil Extracellular Traps (NETs) in Hematologic Malignancies

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Background and Aim: A great variety of diseases that are recognized by excess blood cells in peripheral blood and bone marrow are called hematologic malignancies. Because of the growth of these diseases, investigation of newly involved mechanisms is compulsory. However, in recent years, several studies recommended the role of neutrophil extracellular traps (NETs), also called NETosis, in tissue damage in various diseases and the elimination of these lesions by inhibiting NETs formation. NETosis, one of the essential processes to defend against pathogens, is orchestrated by Neutrophil. Numerous recent studies also showed the susceptibility of neutrophils to NETosis formation in malignancies, including leukemia. To clarify this susceptibility, we aimed to evaluate the actual role of NETosis in the pathogenesis of hematologic malignancies that might help with supplementary therapeutic approaches.

Methods: Relevant literature was identified by a PubMed search (1999-2021) of English language papers using the terms “: Neutrophil extracellular trap (NET),” “NETosis”, “Hematologic malignancies” and “leukemia”.

Results: Studies have indicated the dual role of NETosis in different conditions. It can have a positive effect on the elimination of pathogens in numerous diseases, whereas, can play a negative role in the pathogenesis of some others. Recent studies, however, showed that the risk of thrombosis in patients with blood malignancies could be similar or even higher than patients with solid tumors. This might be due to the involvement of NETs as a scaffold for platelets and red blood cells, providing thrombosis promotion. Therefore, NETosis may predispose a person to thrombosis.

Conclusion: Recently, by identifying the role of NETosis in the pathogenesis of thrombosis, research in acute and chronic leukemia, lymphoma, and particularly in chronic myeloproliferative disorders have risen. Extra platelet-neutrophil crosstalk in these malignancies may, however, be the main reason for amplifying the NETosis. Nevertheless, the Studies of acute leukemia did not accurately determine the positive or negative role of NETosis in their pathogenesis. In contrast, the development of thrombosis by NETosis contribution, using relevant inhibitors prevents complications and mortality from thrombosis in MPN patients. Besides, the suppression of NETosis demonstrates a new approach for DLBCL treatment. However, the role of NETs in other types of lymphomas is not determined so far. Taken together, using well-known NETosis biomarkers to diagnose and follow-up might be useful to help patients with hematologic malignancies using supplementary therapeutic approaches accordingly.

Keywords: Neutrophil extracellular trap (NET); NETosis; leukemia; Hematologic malignancies

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The Role of Thrombophilia Factors & Cytogenetic Analysis in Recurrent Pregnancy Loss (RPL) in Northeastern Iran

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Background and Aim: Recurrent pregnancy loss (RPL) is the occurrence of three or more miscarriages before the 20th week of pregnancy. Thrombophilia factors are one of the main causes of RPL.

Methods: This retrospectively study was performed on women with more than two miscarriages. 620 patients' documents with pregnancy loss were investigated. Based on number of pregnancy loss, the women between 22-37 ages divided to a pregnancy loss (PL) with less than three miscarriages (212) and RPL (180) group and 228 cases role out due to lack of information. In addition, we investigate 150 normal pregnant women without any illness. Cytogenetics analysis like chromosomal culture and thrombophilia panel (MTHFR 677 C/T/ FV Leiden G1691A/Prothrombin G20210A (FII), ACE I/D, PAI1) were performed for all patients.

Results: In the analysis between PL and RPL groups, none of the studied polymorphisms (MTHFR 677 C/T /Factor V Leiden /Prothrombin G20210A/ ACE I/D/ PAI-1) showed a significant relationship ($P\text{-value} > 0.05$). Cytogenetic analysis showed 2 numerical and 9 structural abnormalities among both groups. Statistical analysis indicated significant association between number of abortion and age ($P\text{ value} = 0.005$, $r = 0.139$). We even realized that there was significant relationship between polymorphism number and recurrent number of abortion ($P\text{ value} = 0.018$, $r = 0.6$).

Conclusion: We showed that polymorphisms analysis for thrombophilia factors is more precious test than cytogenetics analysis for RPL detection due to frequency in pregnant woman. We even indicated that no association was found between thrombophilia polymorphisms in PL and RPL groups. This means that screening for Factor V Leiden, prothrombin G20210A, MTHFR C677T, ACE I/D and PAI-1 and cytogenetic analysis in patients with a history of RPL is not recommended.

Keywords: Pregnancy loss; Thrombophilia factors; Cytogenetic; Recurrent Pregnancy Loss

The Thrombotic Effect of Neutrophil Extracellular Trap (NETosis) in Novel Coronavirus (nCoV-2019) Disease

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Background and Aim: Novel coronavirus (nCoV-2019) disease, also called COVID-19, is a highly contagious disease, caused by a recently-revealed coronavirus. Infected people may suffer from respiratory infections, which may be more progressive in patients with a defective immune system and underlying medical problems. In this regard, the innate immune system, particularly neutrophils as the primary cells, plays a decisive role. One of the vital roles of neutrophil extracellular traps (NETosis) has been recently revealed. This process triggers the widespread release of fibrous structures by the stimulant-activated neutrophils. These fibrous structures are composed of cytosolic protein brought together by a network of releasing chromatin. This network can inhibit the spread of pathogens by their entrapment. Moreover, NET might damage the host by producing toxic agents and triggering thrombosis. Therefore, this phenomenon may act as a two-edged blade. Regarding the speedy growth of COVID-19, it is crucial to examine the involvement of NETosis in these patients. This study aims to discuss NETosis participation to show its probable association and help develop new therapeutic approaches in the battle against this viral disease.

Methods: Relevant literature was identified by a PUBMED and GOOGLE SCHOLAR search (2000-2021) using key words and the terms such as: "NETosis", "Neutrophil extracellular trap", "SARS-Cov2", and "nCoV-2019".

Results: Significant increase of NET-specific biomarkers like myeloperoxidase-DNA, citrullinated histones H3, cell-free DNA, TNF- α , IL-2, IL-6, IL-10 in COVID patients which play significant role in promoting both neutrophil count and activation, result in neutrophil extracellular trapping and coagulation by initiating contact pathways and cause thrombin formation through enhancing other pro-coagulant pathways.

Conclusion: The increasing rate of clinical trials in the COVID-19 epidemic indicates that it is now the most significant global health concern requiring excessive research. Since no fully-effective therapies have been revealed to date, the present article deals with the critical mechanism called NETosis to suggest new potential therapies for COVID-19. Regarding the role of NETosis in TRALI, there is an excessive possibility of a similar mechanism in COVID-19, particularly in patients with respiratory distress. The present review indicated that CRS, the result of increasing levels of several pro-inflammatory cytokines and chemokines, could be the main reason for the NETosis contribution to COVID-19. Given the critical role of IL-6 in CRS triggering, Tocilizumab, a humanized recombinant monoclonal antibody, was used to prevent IL-6 attachment to its receptor. Besides, Aspirin and notably DNase-1 decrease the MPO-DNA level and prevent NET formation, coagulation



dysregulation, and, subsequently, dangerous adverse consequences such as organ failure or death. Altogether, this review emphasizes that NETosis, as an essential linker between CRS and thrombosis (especially in patients with ARDS), should be considered to develop more effective therapeutic approaches.

Keywords: COVID-19; Neutrophil extracellular traps (NETosis); acute respiratory distress syndrome; (ARDS); Thrombosis; inflammation



VDR and H19 as Potential Biomarkers in the Pathogenesis of Acute Lymphoblastic Leukemia

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Background and Aim: As the most common pediatric cancer, leukemia accounts for 30% of all cancers that occur in children whose three-quarters are acute lymphoblastic leukemia (ALL). Vitamin D receptor (VDR) and related long non-coding RNAs (lncRNAs) signaling have been shown to play a role in the pathophysiology of several cancers. However, their contribution to ALL remains to be elucidated.

Methods: In this case-control study, 30 newly diagnosed ALL patients and 30 age and sex-matched healthy children were selected. Then, the level of 25(OH) vitamin D and the expression of VDR and four VDR-related lncRNAs were assessed.

Results: No significant difference in serum 25(OH) vitamin D was observed between ALL patients (20.42 ± 6.5 ng/ml) and healthy subjects (25.45 ± 11 ng/ml). In addition, the expression of MALAT-1, HOTAIR, and P-21 was not statistically significant between the two groups. However, a significant reduction in VDR and H19 expression was observed in ALL patients ($p < 0.05$).

Conclusion: 25(OH) vitamin D insufficiency was evident in both groups. VDR and H19 signaling might be contributed to the pathogenesis of ALL which needs further investigation.

Keywords: Acute lymphoblastic leukemia; Vitamin D; Vitamin D receptor; lncRNAs.

A Review on Convalescent Plasma Therapy for COVID-19

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Background and Aim: In patients with COVID-19, convalescent plasma (CP) from donors with antibodies may benefit the patient by providing immediate passive immunity via transfusion or by preparing hyperimmune immunoglobulin preparations. The purpose of this study is to provide an overview of current knowledge regarding the efficacy and safety of CP for COVID-19 infection.

Methods: This study is a review study by analyzing articles from databases, including ScienceDirect, PubMed, and Scopus. This research has been done with keywords " Covid-19," and " Convalescent plasma therapy,".

Results: CP can be an effective anti-viral and anti-inflammatory treatment for COVID-19, which can be adversely affected by SARS-CoV2 infection due to severe cytokine storms, leading to respiratory distress syndrome-like symptoms in severe cases. The first report of CP therapy for COVID-19 patients, where 245 patients in China were treated with specific neutralizing antibodies and the therapy was safe and effective. According to another patient study, anti-SARS-CoV2 spike-binding antibodies were detected from days 10 to 15 following the disease onset and suggested the need to explore the relationship between viral load and immune response. Additionally, the study found that anti-SARS-CoV2 antibody titers in elderly and middle-aged COVID-19 patients were significantly higher than in young patients. In another study, plasma therapy showed benefits in COVID-19 patients with severe underlying health problems such as hypertension, renal failure and chronic obstructive pulmonary disease (COPD). Also, they suggested that patients who survived severe illness might have higher levels of anti-SARS-CoV2 antibodies with longer persistence than those with less severe illness.

Conclusion: Even though it is promising to be hopeful, the question of whether CP can be a silver lining in COVID-19 treatment is still far from being answered conclusively, though more research and a deeper understanding of SARS-CoV2 immunopathology and drug response may help us give plasma therapy a clear thumbs-up.

Keywords: Covid-19; Convalescent plasma therapy; SARS-CoV2; COVID-19 Treatment

Association Between Self-Reported Symptoms of COVID-19 and COVID-19 Seropositivity among Blood Donors with Negative Result in SARS-CoV-2 PCR Testing in Iran

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Background and Aim: Corona virus can cause a wide range of clinical symptoms from a mild cold to a severe respiratory illness. It may never cause symptoms in some infected people. The aim of this study was to investigate the association between self-reported symptoms of COVID-19 and blood donors with negative result in SARS-CoV-2 PCR testing.

Methods: This cross-sectional study was conducted on blood donors with previous PCR testing history who referred to 31 main or satellite blood centers in different provinces of Iran from September 2020 to November 2020. A questionnaire was filled out to collect socio-demographic characteristics, history of COVID-19 symptoms. Blood sample was collected from each participant who had for detecting SARS-CoV-2 IgG antibody using EUROIMMUN kit with ELISA method.

Results: During the study period, a total of 844 blood donors had history of SARS-CoV-2 PCR testing. Of them, 615 (72%) had negative result in SARS-CoV-2 PCR testing who were included in the study. Of them 527 (85.7%) were male. The mean age of participants was 36.5 ± 10.3 . Among participants, 139 (22.6%) self-reported having symptoms; however, only 58 (41.7%) of these were seropositive in serological assay. Symptomatic but seronegative individuals had differing symptom profiles than seropositive individuals: fever was the most common symptom in seropositive individuals 12.4% and cough was the most common symptom in seronegative individuals 5.8%.

Conclusion: The results of the study confirmed that laboratory testing is more precise than self-reported COVID-19 symptoms for determining COVID-19 disease in the past. Symptomatic non-COVID-19 individuals cause mistake for COVID-19 during the outbreak.

Keywords: Blood donors; COVID-19 Nucleic Acid Testing; COVID-19 serological testing; Symptom: SARS-CoV-2; Iran

Blood Transfusion During COVID-19 Outbreak: A Systematic Review

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Background and Aim: SARS-CoV-2 has been detected in Wuhan, China, in late 2019 and has set up a worldwide pandemic. The covid-19 outbreak has affected all people's lifestyles and all tasks. This study was aimed to investigate the challenges and situation of blood transfusion during the COVID-19 outbreak.

Methods: This article is a review by searching reputable databases including PubMed, Scopus, Cochrane Library, and web of science with the keywords "Blood transfusion", "COVID-19", "Iran" which were examined to ensure the completion of search results of article sources and after removing duplicate titles from endnote software and reviewing the titles and abstracts, articles related to JBi tools were reviewed.

Results: There was a decrease in the number of donors in the first 2 weeks of the Covid-19 outbreak, then the number of donors gradually increased. There were no official reports that the coronavirus can be transmitted through blood or blood products. Although a precautionary delay from donating blood for 21 days after any exposure to a confirmed patient is suggested by the European Center for Disease Prevention and Control (ECDC). In addition, those who recovered from the disease should avoid any donation for 28 days after recovering and therapy completion. Also, there was no special method for decontaminating blood products altogether.

Conclusion: The decrease of donors in the first two weeks of the Covid-19 outbreak was unavoidable since people were frightened by the outbreak and stayed at home. The upward trend after the first two weeks was simultaneous to the prevalence and increase in the number of patients, demonstrating that the National Committee on Management of COVID-19 outbreak in the Iranian Blood Transfusion Organization (IBTO) could manage the situation and provide the requirements to attract public trust.

Keywords: Blood transfusion; COVID-19; Iran

Procoagulant Effects of Platelet-derived Microvesicles During Platelet Concentrate Storage

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Background and Aim: Platelet-derived microvesicles (PMVs) are small particles that are released from the platelets membrane and increase significantly during platelets storage. PMVs are bioactive agents, and it has been suggested that they can mediate unintended post-transfusion reactions. The aim of this study was to investigate the procoagulant activity of PMVs in stored platelet concentrates at room temperature.

Methods: Twenty random platelet concentrates (PCs) were stored for up to 5 days. Samples were taken at five different times (0, 1, 3, 4, and 5 days) and evaluated for the presence of PMVs. MVs were separated and following filtration, flow cytometry was used to characterize PMVs based on the expression of CD42b, and Annexin V (AnnV) antigens. The hemostatic activity of PMVs was tested by procoagulant activity (PCA), and clotting time (CT) assays. Results were compared before and after filtration.

Results: Flow cytometry revealed an 18.5-fold increase in PMVs after 5 days of storage. Significant correlations were found between AnnV+ MVs and PCA (r equal 0.94; p less than 0.001), and CT (r equal -0.81; p less than 0.001) which was associated with increased PCA and shortened CT throughout PCs storage. Filtration of samples efficiently removed MVs (p less than 0.001), and also reduced PCA (p less than 0.001).

Conclusion: PMVs particularly AnnV+ MVs have procoagulant property. Reduction of MVs from PCs may reduce the risk of transfusion-induced thrombotic complications.

Keywords: Coagulant; Filtration; Microvesicles; Platelet



Immunology



Allergenicity and Immunochemical Characterization of Five Common Regional Plant Pollens

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Background and Aim: The inhalation of plants pollen grains is one of the most important triggers of allergic symptoms which has a high impact on patients' quality of life as well as a highly important economic burden. Knowing the main source of allergenic pollens in each area is the first step in the prevention and management of inhalant allergic disorders. This study aimed to evaluate the allergenicity and immunochemical characterization of five common regional plant pollens

Methods: Pure pollen of five abundant plants in the area, including Eucalyptus tree (blue gum), Rosa damascene flowers (Damask rose), Jasminum sp flowers (Persian Jasmine), Phoenix dactylifera tree (palm), and Pistacia vera trees (pistachio), were collected during pollination season and then aqueous extracts were prepared. After receiving ethics committee approval, the five extracts, as well as several commercial allergenic extracts, were tested on thirty-seven people with a history of allergic rhinitis, by the skin prick test method. The protein profile of domestic extracts was evaluated by SDS-PAGE and, the allergenic profile was investigated by immunoblotting using pooled serum collected from sensitive patients.

Results: The highest and lowest prevalence of skin sensitivity to domestic extracts was for pollens of the Eucalyptus tree (59.45%) and Phoenix dactylifera tree. The largest mean wheal size was for Rosa damascene flowers and, Eucalyptus tree. The antigenic and allergenic profiles of the extracts also varied. The resolved protein fractions on SDS-PAGE ranged from 5–85 kDa. There were several reactive bands for each extract but the main reactive band for each extract was as follows: Eucalyptus 55 kDa, Jasminum sp 45 kDa, Phoenix dactylifera 40 kDa, Rosa damascene 30 kDa and, Pistacia vera 25 kDa.

Conclusion: In conclusion, the analysis demonstrated these five plants as the aeroallergens which could cause IgE induced hypersensitivity reaction

Keywords: Allergens; Allergic rhinitis; IgE; Immunoblotting; Pollen; SDS-PAGE

Allergic Asthma: An Overview of the Importance of House Dust Mites in Asthma and Allergy

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Background and Aim: Asthma and allergies are one of the most common diseases and affect about 300 million people worldwide. One of the effective factors in this disease is House dust mites (HDM). HDM are allergens from Pyroglyphidae family which grow in hot and humid areas and can increase respiratory diseases such as asthma and inflammation of the respiratory tract. The purpose of this study is to review the importance of HDM on asthma and allergies.

Methods: This is a review study that after searching with the keywords Allergy, House dust mites, Asthma in PubMed, Embase, SID, Google Scholar databases from 2010 to 2021, 47 articles related to inclusion criteria were extracted. And then analyzed.

Results: HDM can cause sneezing, runny eyes, runny nose, skin inflammation, respiratory problems. Allergic reactions to HDM vary from nasal allergies to asthma attacks. However, it mainly causes asthma and atopic dermatitis, inhaling dust containing mites, feces, and other related wastes and fungi can cause allergic reactions. HDM also feed on the human body shell and other organic matter.

Conclusion: Implementation educational programs, increasing public awareness in environmental health is also effective in preventing and not exacerbating this disease.

Keywords: Allergy; House dust mites; Asthma

Alteration in HLA-DR, CD64, CD206 and CD163 Marker Expression in Transduced Macrophages with MARCH-1

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Background and Aim: studies have shown that macrophage polarization is a therapeutic target for controlling immune responses especially in chronic inflammatory conditions. MARCH family protein (Membrane-Associated RING-CH-type finger) is known to inhibit dendritic cell maturation and antigen delivery to T lymphocytes. We transduce macrophages with MARCH-1 gene and assay their immunophenotypes to consider the macrophage plasticity to M2. Those could have been beneficial for preventing covid-19 cytokine storm progression.

Methods: Plasmids pCDH-1 MARCH-1- GFP, psPAX2.221 and PMD2.G were used for transferring MARCH-1 into the macrophage. Macrophages derived from THP-1 cells and human monocytes were transduced with viral lentivirus produced from HEK-293T cells. transduced cells were stained with Anti-CD206-PE, Anti-CD163-PE, Anti-HLA-DR-PE, Anti-CD64-PE, Anti-CD14-APC markers for flow cytometry consideration. Mean fluorescence intensity (MFI) was assessed for each marker separately.

Results: MARCH-1GFP+ expressing macrophage population increase by 69.3% compared to non-transduced population. The mean fluorescent intensity (MFI) of HLA-DR expression in MARCH-1+ macrophage population reduced from 331 ± 35 to 134 ± 19 arbitrary unit ($P=0.008$) and CD64 decreased from 366 ± 95 to 284 ± 85 arbitrary unit as well ($P=0.015$). However, CD206 expression in transduced macrophages increased from 100.6 ± 13.7 to 168 ± 22 arbitrary unit ($P=0.008$). The MFI for CD163 expression in transduced macrophages was 215 ± 55 in compared with non-transduced with 199.8 ± 85 arbitrary unit ($P=0.54$).

Conclusion: This study has offered a detailed characterization of the polarizing effect of MARCH-1 on human monocyte-derived macrophages. Our data suggest that an increased expression of MARCH-1 in monocytes/ macrophages could be responsible for macrophage polarization to M2 macrophages. Thus targeting MARCH-1 in monocytes/macrophages could represent a new therapeutic strategy in inflammatory or antiinflammatory conditions such as covid-19 infection.

Keywords: MARCH-1; HLA-DR; M2 macrophages; CD64

Alterations in Serum Levels of Immune Inhibitory Markers in Coronaviruses-Related Anxiety in Animal Model; The Protective Roles of *Rhus Coriaria*

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Background and Aim: Anxiety of corona pandemic via different mechanisms causes immunosuppression. Because of the high potential of *Rhus coriaria* on pathological anxiety, the aim of the present study was to investigate the influence of *Rhus coriaria* inhibitors on blood levels of Interleukin-2 (IL-2), Interferon- γ (IFN- γ), Tumor Necrosis Factor- α (TNF- α) and white blood cells (WBCs).

Methods: Thirty male Wistar rats were randomly divided into 5 groups (n=6 for each group). Group 1 was received with normal saline for 30 consecutive days. Group 2 was received with normal saline for 30 consecutive days and at day 30 rats were injected intraperitoneally with the pentylenetetrazol (PTZ) (20 mg/kg). In groups 3 to 5, the rats were pretreated with different concentrations (20, 60 and 180 mg/kg) of the *Rhus coriaria* extract, respectively. At day 30 groups 3 to 5 were injected with pentylenetetrazol (20 mg/kg) to induce anxiety. Animals were treated once daily over a period of 30 consecutive days. Blood samples were collected 24 hours after injection (i.p) of pentylenetetrazol (20 mg/kg) and levels IL-2, IFN- γ , TNF- α and WBCs were determined. Data were statistically analyzed through one-way ANOVA, followed by Tukey's post hoc test.

Results: Based on the results, a decrease in blood levels of IL-2, IFN- γ , TNF- α and WBCs was observed 24 hours after injection of PTZ compared to control rats (P less than 0.05). Oral administration of *Rhus coriaria* improved the decreased level of IL-2 and TNF- α (P less than 0.05). There was no significant difference in IFN- γ and WBCs between group 2 and group 3 (P greater than 0.05). These changes were ameliorated through treatment with *Rhus coriaria* extract at different doses as compared with the group 2 (P less than 0.05).

Conclusion: The extract was found to be an effective inhibitor that can improve immune system function due to its active compounds with medicinal value.

Keywords: Anxiety; Coronaviruses; Immunomodulation; Pentylenetetrazol; *Rhus coriaria*

Assessment of Circulating Vitamin D in Adult Burned Persons According to Total Body Surface Areas (TBSAs) Burned

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Background and Aim: Regards to few researches, Vitamin D has a significant role in the immune system. This study aimed to evaluate serum concentration of Vitamin D, Calcium, Magnesium, and Phosphorous in adult burned persons according to the different total body surface areas (TBSAs) of burns in the 1st, 3rd, and 7th days.

Methods: In this study, 30 individuals as healthy controls (HC) group and 50 individuals with various degrees of burn injuries were participated. With reference to the burn of TBSAs %, the burned persons were categorized into three groups: G1 (N equal 25): 20%-50%TBSAs burned (mean $\pm$ SD age: 39.5 ± 14.1); G2 (N equal 10): 50%-70% TBSAs burned (mean $\pm$ SD age: 32.4 ± 9.4), and G3 (N equal 15): more than 70%TBSAs burned (mean $\pm$ SD age: 32.3 ± 15.5).

Results: The mean $\pm$ SD of age in burned persons and HC group were 35.9 ± 14.3 and 40.8 ± 10.9 , respectively. In general, burned persons compared with HC group indicated no significant differences for serum total Calcium, Magnesium, and Phosphorous. A significant decrease of Vitamin D serum levels was seen in the burned persons in comparison to HC group (p less than 0.001). However, there was no significant discrepancy during 1st, 3rd and 7th days of total burned persons. In addition, Vitamin D serum levels in three groups of TBSA burned in 1st, 3rd, and 7th days were evaluated. There was a significant decrease in Vitamin D serum concentrations among three TBSAs burned groups (G3 burned in the 3rd and 7th days) (p less than 0.05).

Conclusion: Vitamin D is usually found to be rare in burned persons especially in adult with severe burning. So, an important role of Vitamin D as an immunomodulator in the immune system might be useful in patients with very low levels of serum Vitamin D. More studies are required to prove definite guidelines respecting the supplementation of nutrients in burned persons.

Keywords: Adult; Burn; Serum Vitamin D; TBSAs

Cardiac Immunology: A New Era for Immune System in the Heart

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Background and Aim: The past few decades have generated growing recognition that the immune system makes an important contribution to cardiac development, composition, and function. Data suggest that a dysregulation of the immune system and inflammatory pathways may be the leading mechanisms in a large number of Cardiovascular diseases, including heart failure, pericardial disease, cardiomyopathies, and rhythm disorders. The immune system has a fundamental role in the post-myocardial infarction process. Different immunosuppressive drugs, anti-inflammatory, and immunomodulatory interventions have been studied upon MI. Suppressing the immune system has a negative impact on post-MI wound healing processes. One of the clinical trials has shown that the granulocyte colony-stimulating factor (G-CSF), an endogenous cytokine mobilizer of bone marrow granulocytes, improved cardiac function by decreasing scar size and preventing LV remodeling in post-MI patients. This study indicates that receiving G-CSF (10 microgram/kg of body weight, daily) for continuous 7 days is safe and seems to offer a pragmatic strategy for recovery of myocardial global function. Interferon regulatory factor 3 is another part of the immune system that can play a role in poor outcomes following myocardial infarction. MI-activated IRF3 results from ischemic cell death, the release of dying cell DNA, and subsequent activation of the cGAS/STING/IRF3 axis.

Results: Activated IRF3 in this instance increases expression of inflammatory factors, such as CXCL10, and ISGs, such as IFIT1 and IFIT2, in damaged heart vasculature. More importantly, deletion of the IRF3 gene in an MI mouse model prevents most of the MI-induced expression of inflammatory factors and ISGs in MI heart tissue, thereby preventing post-MI mortality and increasing post-MI heart function. Therefore, pharmaceuticals that reduce IRF3 activation may be a future treatment post-MI. Elevation of inflammatory biomarkers such as TNF alpha, ST2, IL-6, galectin-3, and pentraxin-3 in the circulation is a hallmark characteristic for heart failure. Targeting IL-1 proteins reduce leukocyte activation, supports a resolution of infarct inflammation, and ameliorates HF in animal models.

Conclusion: Understanding how cell-specific and molecular mechanisms of the immune response interfere with the cardiac remodeling in heart failure may open new avenues to design biomarkers or druggable targets.

Keywords: Inflammatory biomarkers; Myocardial infarction; Heart failure; Immune system

Clinical Features Associated with the Magnitude of the Antibody Response in COVID-19 Patients

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Background and Aim: Despite extensive studies on the COVID-19 epidemic, there are no accurate reports on the association between clinical symptoms and the magnitude of immune response in recovered patients. The aim of this study was to determine the relationship between clinical symptoms and serological responses in recovered COVID-19 patients at admission time and after recovery, also to identify risk factors for the lower serological response as a result of the likelihood of disease recurrence.

Methods: Methods: In this descriptive-analytical study, which was performed by call-out method, the serum levels of IgM and IgG antibodies were measured using ELISA in 248 recovered patients. Patients were classified into three groups (IgM+ and IgG+), (IgM- and IgG+) and (IgM- and IgG-), based on the results of serological tests. The patient's signs and symptoms at the onset of the disease as well as serological tests were investigated in the serological groups.

Results: Among the symptoms, only vomiting and appetite showed a significant relationship with IgG response (P value= less than 0.05). Patients who reported normal appetite at the time of test had a significantly higher rate of IgG positivity; patients who reported vomiting at admission and the time of test had significantly lower IgG-positive levels.

Conclusion: Based on the present study's results, report vomiting among other symptoms as well as anorexia after recovery should be considered as a risk factor and these patients need more attention to follow the treatment process and re-infection.

Keywords: COVID-19; Immune response; Clinical features; Vomiting; Anorexia

Clinical Sensitivity of PCR and Serological COVID-19 Diagnostics

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Background and Aim: Today, PCR test is known as the standard gold in the diagnosis of coronavirus infection. This method has shown a sensitivity of nearly 100% by tracking virus-specific nucleic acids in nasopharyngeal samples. But this sensitivity is dependent on viral load and disease stage. To diagnose suspected cases that, lack molecular confirmation, Serology could help. The aim of this study was to investigate and compare the sensitivity of PCR-test and serological tests in different stages of the disease and identify the best method in each period of infection.

Results: Sensitivity gets tested daily, from the onset date of the symptoms or when the first positive test was calculated. 5 days after the onset of symptoms, PCR test showed 90% sensitivity. In the following days, the sensitivity of the test decreased, following the reduction of virus load. Clinical sensitivity during days 9 to 11 after symptom onset, was 70%-71%, and 30% at Day 21. Unlike the PCR-test, which a decrease in susceptibility is seen in the days after the onset of symptoms, serological sensitivity increases following antibody production. After Day 7, >50% of patients had at least one antibody isotype, >80% after Day 12, and 100% by Day 21.

Conclusion: The sensitivity of these techniques depends on the state of the patient's illness. The highest sensitivity of PCR-test is in the stage of symptoms and the highest load of virus. However, serological test is followed by indirect markers, which increases its sensitivity by finding antibodies and after a few weeks of recovery.

Keywords: PCR; SARS-CoV-2; sensitivity; serology

COVID-19 Serological Survey in Referred to Bou-Ali Laboratory, Yazd

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Background and Aim: Coronavirus disease 2019 (COVID-19) is a transmissible disease caused by severe acute respiratory syndrome coronavirus (SARS-CoV-2). An antibody test searches for the presence of antibodies, which are made in response to infection. Antibodies show an immune response to the infection. Antibody test results are important for detecting previous infections in people who had few or no symptoms.

Methods: This descriptive cross-sectional study was carried out on individuals referred to Bou-Ali Laboratory, Yazd Iran. 5 ml of peripheral venous blood from each contributor was collected. The serum of the collected blood samples was used. The presence of anti-SARS-CoV-2 IgM and IgG antibodies in 798 participants were determined by ELISA Method (Pishtaz Teb) and 189 participants were tested by Diazist Kit.

Results: Nine hundred and eighty-seven contributors were 53.4% males and 46.6% females. Mean age of the participants was 41.04 ± 15.41 (range 3 to 96 years). Overall 313 (31.7%) and 149 (15.1%) samples had anti-SARS-CoV-2 IgG and IgM in serum respectively. The mean $\pm$ SD value of anti-SARS-CoV-2 IgG was 30.9 ± 20.5 Au/ml (Min. 10.2, Max. 106.9). The mean $\pm$ SD value of anti-SARS-CoV-2 IgM was 41.8 ± 38.7 Au/ml (Min. 10.3, Max. 136.3).

Conclusion: The low percentage of people who had anti-SARS-CoV-2 antibodies indicates a low level of immunity to the disease in Yazd population. The low anti-SARS-CoV-2 IgG titer may also indicate that the immunity to the disease is short-lived

Keywords: Covid-19; anti-SARS-CoV-2; Yazd

DiABZI as a STING Agonist Could Effectively Protect Cells From SARS-CoV-2 Infection: A Systematic Review

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Background and Aim: Despite relative success in the management of COVID-19, new variants and diverse clinical presentations still are challenging common therapeutic options and indicate the importance of an efficient therapeutic agent. Stimulator of interferon genes (STING), the critical adaptor molecule in innate immune signaling pathways, could be activated by cyclic dinucleotides (CDN) and leads to autophagy and significant IFN secretion which inhibits viral entry and replication. Dimeric Amidobenzimidazole (diABZI) is a new class of synthetic STING agonists whose protective effect on viral infections has been shown. In this systematic review, we will discuss the protective effect of diABZI on infection with SARS-CoV-2 and other properties as a novel therapeutic option in the clinic.

Methods: All studies with the similar subject were detected by searching the MESH terms in the MEDLINE, Scopus, and Google Scholar databases from 2015 to December 2021. After evaluation of the abstracts, just 10 relevant studies were selected among them.

Results: DiABZI is a small synthetic non-nucleotide STING agonist with 18-fold more potent than CDNs in the production of IFN- β . Recent studies relieved that diABZI reduced infected cells with SARS-CoV-2 through activating STING signaling in compression with IFN treatment. Notably, greater EC₅₀ (3 nM) and less CC₅₀ (100 nM) have been reported when compared with Remdesivir, IFN- α , and CDN. Moreover, intranasal administration of diABZI before the infection and even in the first hours of post-infection remarkably reduced lethality compared with when treatment was delayed.

Conclusion: The results show that because of systemic administration of diABZI in the first days of infection has a higher efficacy in comparison with IFN therapy and Remdesivir with low side effects, diABZI not only could be recruited in future clinical trials as a novel potential therapeutic agent in COVID-19 treatment but also could be employed in other viral respiratory infections.

Keywords: DiABZI; COVID-19; STING; Treatment

Diagnostic Accuracy of Complete Blood Cell Count (CBC) Indices and C Reactive Protein (CRP) for COVID-19

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Background and Aim: Diagnosis of COVID-19 is through polymerase chain reaction (PCR) or typical involvement of lung by the virus in CT scan. However, PCR is not always available, and also CT scan has a high dose of radiation. This study was performed to find the role of complete blood cell (CBC) indices and qualitative C reactive protein (CRP) in the screening of symptomatic patients.

Methods: A diagnostic accuracy study was performed on symptomatic cases in Abadan. Four stepwise logistic regression models were designed that the outcomes were PCR positivity, CT scan positivity, PCR and CT scan positivity, and COVID-19 positivity (i.e. PCR or CT scan positivity). Post-estimation receiver operating characteristics (ROC) curve analysis was performed to report the area under the curve (AUC).

Results: A total of 104 patients were studied. The most accurate model was for prediction of CT scan positivity (AUC =0.874) in which the predictors were age (odds ratio [OR] =1.063) and CRP (OR =2.661 for each plus of positivity). The second accurate model was for the prediction of COVID-19 positivity (AUC =0.828) in which the predictors were white blood cell count (OR =0.735 for every 1000 counts per μ L) and neutrophil per lymphocyte ratio (OR =1.248).

Conclusion: Higher levels of CRP are associated with and predictor of lung involvement in COVID-19 infection. CRP qualitative levels can be measured before CT scan if there is no other indication for imaging.

Keywords: COVID-19; CBC; WBC; CRP; CT scan

Differences Between Molecular and Serological Results of Anti-SARS-CoV-2 in Healthcare Workers

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Background and Aim: The serological and molecular tests (PCR) are widely used in the diagnosis of COVID-19. Because its diagnostic efficacy remains unclear, the aim of this study was to evaluate the degree of positive serological and molecular tests among healthcare workers.

Methods: In this descriptive-analytical study, which was performed by recall method, PCR results and the level of serum IgG antibodies of 492 personnel working in hospitals and medical centers of Shahrekord University of Medical Sciences was evaluated based on PCR and ELISA. Chi-square, t-test, and ANOVA tests were used in SPSS-15 software to investigate the relationship between variables.

Results: In this study, 19.2% of the staff had a positive PCR test; However, IgG positivity among them was only 16%.

Conclusion: Based on the results of the present study, the positive serological response in healthcare workers was less than the prevalence of the disease based on molecular tests. We believe that the PCR is a better method for the diagnosis of and follow-up of COVID-19 in healthcare workers.

Keywords: PCR; Immune response; Serological test; Health workers

Early Recognition of the Likelihood of COVID-19 Infection by Measurement of Immunologic Indexes

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Background and Aim: With the possibility of the arrival and spread of SARS-CoV-2 in Iran from February 2020 and investigation of diagnostic procedures for this disease in patients in Rosha medical diagnostics lab, Tehran, Iran, and the significance of screening and diagnosing and ultimately quarantining suspected COVID-19 cases at home or in a hospital to prevent them from entering organizations and offices, their diagnosis took place during the onset of the disease progression. First, the measurement of white blood cell count and the lymphocyte activity percentage and then the assessment of CRP, ESR, LDH, and COVID-19 IgG and IgM was commenced and continued for one year. Clinical symptoms of patients including fever, cough, and fatigue in the early stages of the disease were examined. Moreover, laboratory diagnosis was made possible by the evaluation of CRP and ESR and in the next step quantification of IgG and IgM in patients was reviewed. Analysis of the aforementioned indexes in this study showed significant differences in the comparison of CRP and ESR with COVID-19 IgG and IgM.

Methods: CRP was measured by turbidity method with Hitachi biochemistry analyzer that evaluates the optical opacity of the sample. In this study, the opacity is caused by acute-phase proteins in the patient's serum. ESR was measured from the sedimentation rate of red blood cells in millimeters during a specific amount of time. ESR is an unspecific marker for inflammation, acute and chronic infection, progressive malignancies, tissue necrosis, and infarction.

Results: Analysis of the aforementioned indexes in this study showed significant differences in the comparison of CRP and ESR with COVID-19 IgG and IgM.

Conclusion: In comparison to past diagnostic procedures, in early diagnosis of COVID-19 and after the advent of COVID-19 diagnostic kits we concluded that evaluation of CRP and ESR can help identify the disease sooner and therefore commence the treatment earlier and when it is more effective. In most cases, the CRP and ESR levels of the patient are synchronized with each other because they both show inflammation in the body.

Keywords: ESR; CRP; COVID-19; IgG; IgM

Effects of 50-Hz Magnetic Field on Expression of *T-bet*, *GATA-3*, *IFN- γ* and *IL-4* Genes During 90 Days of Exposure

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Background and Aim: Due to the advancement of technology, the use of electrical appliances is increased. The produced magnetic fields (MFs) by these devices may cause adverse effects on human health. Therefore, this study investigated the effects of 50-Hz magnetic field on gene expression of T-box transcription factor (T-bet) and transcription factor GATA binding protein-3 (GATA-3) related with Th1 and Th2 subsets and their associated cytokine i.e. interferon (IFN)- γ and interleukin (IL)-4 genes.

Methods: Twenty rats were equally divided into exposure and control groups. The exposure group was exposed to 100 μ T magnetic flux density (50 Hz, 2h/day for 90 days). The rat bloods were drawn on all rats on days 0, 7, 14, 30, 60, and 90. The blood expression levels of the genes were measured by reverse transcription-quantitative polymerase chain reaction.

Results: The results showed that the weights of rats, spleens and thymuses were not significantly different compared to controls during exposure. The expressions of the IFN- γ , GATA-3 and IL-4 genes were reduced, respectively at 30, 60 and 90 days of exposure. The expression of T-bet was not changed in the groups at any time.

Conclusion: Magnetic fields may down regulate inflammatory genes and cytokines. This reduction may decline the immune system. On the other hand, the magnetic field can be useful in the treatment of autoimmune and allergic diseases.

Keywords: T-box transcription factor; Transcription factor GATA binding protein-3; Interferon- γ ; Interleukin-4; Th1 cells; Th2 cells; Magnetic field

Evaluaiton of COVID-19 Vaccines (Sputnik V, AZD-1222, Sinopharm, and Covaxin) Efficacy in Inducing Anti RBD IgG Antibody among a Group of Vaccine Receivers

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Background and Aim: Ability of vaccines in inducing humoral immunity against infections agents is an important issue in evaluation of the vaccine efficacy and usefulness. This study was designed to investigate the efficacy of the four common COVID-19 vaccines including Sputnik-V, AZD-1222, Sinopharm, and Covaxin in eliciting an RBD IgG two weeks after the second dose among a group of healthy vaccine receivers.

Methods: An invitation for participating in the study and donating a Venus blood sample was sent for the personnel of Birjand Medical University. Participants were filled a questionnaire about their health status, previous COVID-19 infection as well as type and date of vaccines. The qualitative measurement of anti RBD IgG was performed by a commercial ELISA kit.

Results: In total 214 people (Mean age: 36.5 ± 8.75 , range: 19-64 years, M/F: 0.66) were participated in this study. The prevalence of positive anti RBD IgG was 95%, 93%, 73% and 70% for Sputnik V, AZD-1222, Sinopharm and Covaxin respectively. There was no significant difference between female and male participants.

Conclusion: According to the results, Sputnik V and AZD-1222 vaccines are more effective than Sinopharm and Covaxin vaccines in inducing anti RBD IgG.

Keywords: Vaccine; Covid-19; efficacy; Sputnik V; AZD-1222; Sinopharm; Covaxin

Evaluate the Effect of Mesenchymal Stem Cells Derived Exosomes on Regulatory T-cell Function in COVID-19 Patients PBMCs

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Background and Aim: During the COVID-19 pandemic, much attention was paid to the function of immune cells in the pathogenesis of the disease. During the cytokine storm, which is part of the pathogenesis of this disease, a large amount of inflammatory cytokine is produced, leading to the occurrence of acute respiratory syndrome. Regulatory t cells suppress the immune system cell responses by producing inhibitory cytokines such as interleukin-10 (IL-10) and transforming growth factor-B (TGF-B) and cell-to-cell interaction. Therefore, studying the function of regulatory T cells in patients with Covid-19 is of particular importance and can help to better understand the pathogenesis and find treatments based on the increase of regulatory T cells. Since mesenchymal stem cells-derived exosomes have immunomodulatory properties, the use of these vesicles can improve the function of Tregs in COVID-19 patient PBMCs.

Methods: In this study, mesenchymal stem cells were first isolated from adipose tissue. After culturing these cells, the exosome was isolated from their supernatant by ultracentrifugation. SEM, TEM and DLS confirm isolated exosomes characterization. Then PBMCs were isolated from patients with COVID-19 cultured in the presence of these exosomes. The expression of transcription factors related to these cells and the secretion levels of cytokines was assessed by real-time PCR and ELISA, respectively, before and after treatment with MSCs exosomes.

Results: A significant reduction was observed in the levels of TGF- β , IL-10, and FoxP3 expression in COVID-19 patient PBMCs. Our data revealed the expression of FoxP3, TGF- β , and IL-10, the secretion levels of the TGF- β , and IL-10 in cultured PBMCs are increased in MSCs-derived exosome-treated group compared to the control group.

Conclusion: The current study results indicated that AD-MSCs derived exosomes can restore the function of Treg cells of COVID-19 patient isolated PBMCs.

Keywords: MSCs; Exosome; PBMCs; COVID-19; Cytokine

Evaluate the Effect of Mesenchymal Stem Cells Derived Exosomes on TH-17 Frequency in COVID-19 Patients PBMCs

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Background and Aim: After the outbreak of the Covid-19 pandemic, much attention was paid to the responses of immune system cells and their role in the pathogenesis of the disease caused by the SARS-COV-2 viral infection. Impaired balance of the ratio of regulatory T cells to TH-17 is one of the main reasons related to the immune system in the pathogenesis of this disease. Recently, the use of MSCs in treating COVID-19 disease has improved long-term pulmonary function. Still, the specific mechanisms by which MSCs derived exosomes inhibit the severe inflammatory response induced by SARS-CoV-2 have not been elucidated.

Methods: In this study, we first took 5 ccs of blood from people diagnosed with covid-19. Then PBMCs were isolated from these patients' blood. MSCs isolated from adipose tissue a starved for 3 days to isolate exosomes. Exosomes were then separated from the supernatant by ultracentrifugation. Exosomes were characterized by SEM, TEM, and DLS. PBMCs were then co-cultured these exosomes, and the frequency of TH-17 cells was assessed by flow cytometry before and after treatment by intracellular IL-17A staining.

Results: A significant increase was observed in the proportion of peripheral TH-17 cell frequency ($p=0.029$) in COVID-19 patient PBMCs compared to healthy individuals. Our data revealed that MSCs exosomes considerably influenced the ratio of intracellular IL-17A expressing TH-17 in all the patients.

Conclusion: The current study results indicated that AD-MSCs derived exosomes can decrease the frequency of TH-17 cells of COVID-19 patient isolated PBMCs.

Keywords: MSCs; Exosome; COVID-19; PBMCs; TH17

Evaluation of CD4+ T cells frequencies in women with Hashimoto's thyroiditis

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Background and Aim: Hashimoto's thyroiditis (HT) is the most prevalent autoimmune disease and there is no definitive treatment available for this disease. In order to find the appropriate therapeutic approach, it is necessary to determine the mechanism of this disease. To achieve this purpose, the frequency of CD4+ T cells in HT patient was evaluated and compared to that in healthy individuals.

Methods: Twenty-six female patients with HT, aged 20-45 years enrolled in this study. Based on level of anti-TG and anti-TPO in serum of HT patients, they were divided into two groups. In group 1 (n=13) the serum level of anti-thyroid peroxidase antibody (anti-TPO) was above 100 IU/ml whereas in group 2 (n=13) both the serum levels of anti-TPO and anti-thyroglobulin antibody (anti-TG) were above 100 IU/ml. Eleven healthy women were considered as control group, group 3. Using flow cytometry, the frequency of T helper1 (Th1), T helper2 (Th2), T helper17 (Th17), T regulatory type 1 (Tr1) and LTCD4+IL-4+IL-17+ cells and mean fluorescent intensity (MFI) of their related cytokines were evaluated.

Results: The frequency of Th2 cells in groups 1 (anti-TPO > 100) and 2 (anti-TPO > 100 and anti-TG >100) were more than control group. Only the difference between groups 3 (healthy person) and groups 2 (anti-TPO > 100 and anti-TG >100) was significant (P= 0.022). The frequency of LTCD4+IL-4+IL-17+ cells in group 1 was significantly more than group 3 (P=0.027); However, the difference between group 2 and 3 was not significant (P=0.126). The expression of interferon gamma (IFN- γ) in group 2 (P= 0.001) and group 1 (P= 0.001) is significantly higher than group 3. The frequency of Th17, Th1 and Tr1 cells, and MFI of IL-17 and IL-10 were not significantly different between study groups.

Conclusion: In the present study, no significant differences have been observed in the frequency of Th17 and Tr1 cells and in MFI of IL-17 and IL-10 in comparison with healthy individuals. Therefore, trying to make a change in the population of these cells probably does not have a significant therapeutic effect. Since Th2 cells and the expression of IFN- γ increased in women with HT, reducing

the frequency of Th2 or the expression of IFN- γ may be effective in controlling this disease progression. It may be helpful for these patients and prevent the progression of the disease

Keywords: Hashimoto's thyroiditis; CD4+ T cells; Th17; Tr1; Th1; Th2; LTCD4+IL-4+IL-17+



Evaluation of Lactobacillus Plantarum Effects on Survival and Proliferation of MCF-7 Breast Cancer Cell Line

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Background and Aim: Need for novel preventive and curative approaches with more safety than the present one seems essential for breast cancer as the most common cancer among women. Probiotics are described as a group of live bacteria and yeasts with beneficial effects on human health. They have various effects and recently anti-cancer properties have been shown for them. Lactobacilli are a species of probiotics with antitumor activities. In this study, we aimed to assess the effects of Lactobacillus plantarum (L. plantarum) on the MCF-7 breast cancer cell line

Methods: L. plantarum was cultured in de Man, Rogosa, and Sharpe (MRS) broth medium and human breast cancer cell line (MCF-7) in DMEM high glucose medium containing 10% heat-inactivated fetal calf serum (FBS), and 1% penicillin/streptomycin. Effects of different concentrations of supernatant (5%, 10%, 20%, and 50%) and lysed bacterial pellets (1%, 5%, and 10%) of L. plantarum on MCF-7 cells were examined by MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] assay and Trypan blue exclusion test of cell viability. Also, MRS medium was used as a control.

Results: In this study, supernatants of L. plantarum (10%, 20%, and 50%) decreased the proliferation and cell viability of MCF-7 cells. These effects were not noticeable using bacterial cell pellets

Conclusion: Results of this study suggest that L. plantarum might secrete some metabolites that could have anti-proliferative effects on cancerous cells. Therefore, L. plantarum administration as a preventive or therapeutic agent in combination with common medications of breast cancer may improve patient health and their response to treatments.

Keywords: L. plantarum; MCF-7; Breast cancer;

Evaluation of Practical Strategies in the Diagnosis and Treatment of COVID-19 by Monoclonal Antibodies

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Background and Aim: Coronavirus disease, caused by severe acute respiratory syndrome virus, has emerged as a new infectious disease which has reached pandemic proportions. Monoclonal antibodies (MAbs) are one of the emerging therapeutic agents efficacious for treating infectious diseases such as COVID-19. They are one of the fastest-growing pharmaceuticals and are considered to be highly specific in their action. MAbs are lab-grown antibodies that specifically target the pathogen, causing its destruction immediately. The present study investigates technological innovations in the development of monoclonal antibodies in the diagnosis and treatment of COVID-19.

Methods: Application techniques for MAbs were reviewed using Internet search engines such as PubMed, Google Scholar, Science Direct, and WIPO (World Intellectual Property Organization). We searched Lit Covid, and Med Rxiv using the search terms coronavirus and only those articles containing the required information were selected for the analysis.

Results: The findings indicated that the alteration in the key components such as epitopes might contribute to the development of resistance to most of the preventive as well as therapeutic interventions, including MAbs. Further, the RNA-virus in the past has displayed higher rates of mutation. In this context, efforts are being made to target different sites of the virus by using a combination of drugs.

Conclusion: MAbs have a neutralizing ability against several pathogens including SARS-CoV-2. They are reported to have potential for both a prevention as well as therapeutic purpose. Monoclonal antibodies would be a complement to the vaccines especially during the pandemic, as they offer immediate protection upon administration and lasts for weeks or months. This will, also, help in the treatment of infected patients and may prevent young and elderly people from being infected. However, it is important to study the safety, manufacturing capacities, production time, price and availability of these products monoclonal antibodies in all countries.

Keywords: practical strategies; diagnosis and treatment; COVID-19; monoclonal antibodies

Evaluation of TH1 and TH2 Cytokines in Pregnant Women with a History of Recurrent Abortion and Normal Pregnancy in the First Trimester of Pregnancy and its Relationship with Pregnancy Hormones

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Background and Aim: Recurrent Spontaneous abortion(RSA) is one of the most common complications of pregnancy. Various factors including immunologic factors have been proposed to contribute to the etiology of RSA. We aimed to measure cytokines in women with a history of RSA in the first trimester of pregnancy. Moreover, we assessed the relationship between T-helper cytokines (TH1and TH2), and hormone progesterone, in all groups.

Methods: In this study, normal pregnant women (n=30) and those with history of recurrent abortion that have failed pregnancy (RSA-F) and with recurrent abortion who have a successful pregnancy(RSA-S) (n=43) enrolled. Venous blood (15ml) was obtained from all participants for serum and culture analysis. We assessed IL-4, IL-6, and IL-10 as well as TNF- α and IFN- γ and progesterone hormone, using enzyme-linked immunosorbent assay (ELISA).

Results: The mean concentrations of TH2 cytokines and progesterone level significantly decreased in RSA - F women compared with RSA - S and normal groups ($P < 0.05$) . in addition, the concentration of TH1 cytokines including TNFa and IFN - γ significantly increased in (RSA - F) compared with women with normal pregnancy and RSA - S . ($P < 0.05$) . We found a negative correlation between TH1 / TH2 cytokine in RSA - F women, while a positive correlation was observed between progesterone levels and TH2 cytokines ratio in normal group .

Conclusion: A higher TH2 cytokine level is seen in RSA-S women, while this trend shifted toward a higher TH1 level in the RSA-F group. Cytokine assessment could be proposed as a marker for predicting the success or failure of pregnancy in women with recurrent abortion during the first trimester of pregnancy.

Keywords: Recurrent Abortion; TH1 Cytokines; TH2 Cytokines; Progesterone

Evaluation of the Effect of SB203580, Camptothecin and Metformin on Cell Viability in Human Breast Cancer Cell Lines MDA-MB-231 and MCF-7

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Background and Aim: Breast cancer is the most common malignancy diagnosed in women and leads to death in many countries. Researches has shown that metformin inhibits the proliferation and growth of tumor cells, especially breast tumors, by activation of AMP-activated protein kinase (AMPK). On the other hand, SB203580, as a Mitogen-Activated Protein Kinase- α (P38 α) inhibitor, inhibits cell growth and induces apoptosis in the cancer cell line. The use of camptothecin, as a Nuclear Receptor subfamily 4 group A (NR4A) inhibitor, reduces cancer cell invasion and metastasis by inducing failure in single-stranded DNA, in regard to, there is limited information on the simultaneous effects of metformin, camptothecin and SB203580 on cancer cells, we intended to investigate the single and combined effects of these drugs on the survival rate of breast cancer cell lines.

Methods: The human Breast cancer cell lines MCF7 and MDA-MB-231 were seeded onto 96-well plates in 100 μ L of RPMI 1640, which was supplemented with 10% fetal bovine serum and 1% penicillin–streptomycin, at a density of 3×10^3 cells per well for 24 hours before treatment. Then cell treated with metformin (10 mM), SB203580 (10 μ M) and CPT (25nM) alone and in combination for 24 and 48 hours. The viability of cells was then examined by a colorimetric test that reflects the number of viable cells present. Briefly, 20 μ L of MTT (5 mg/mL in PBS) was added to each well and incubated in a dark place at 37°C for 3 hours. Thereafter evacuating the contents of the plate, 100 μ L of DMSO was added to each well and the plates were incubated for additional 20 min. The absorbance was measured by an Elisa reader at 590 nm.

Results: The viability of MDA-MB-231 cells in group of metformin was 60% and 24% after 24 and 48 hours' treatment, respectively while in the SB group, respectively, 96% and 86% and in the CPT group 100% and 91%. Combination of metformin/SB decreased cell viability compared to single treatment after 24 and 48 hours but there wasn't significant decrease in combination of metformin/CPT and also in combination of SB/CPT after 24 and 48 hours. Our results indicated that combination of metformin/SB/CPT leads to significant decrease of cell viability after 24 and 48 hours. The viability of MCF7 cells in group of metformin was 76% and 48% after 24 and 48 hours' treatment, respectively while in the SB group, respectively, 93% and 104% and in the CPT group 95% and 91%. There wasn't significant decrease in combination of metformin/CPT and also in



combination of SB/CPT after 24 and 48 hours. Our results indicated that combination of metformin/SB/CPT also didn't leads to decrease of cell viability after 24 and 48 hours.

Conclusion: In this study, we observed that metformin decreases MDA-MB-231 cell viability especially after 48 hours' incubation while SB and CPT were not able to inhibit cancer cell growth, but in combination with metformin, either as a double combination (with the exception of the Metformin/CPT group, which was seen as a slight decrease due to metformin) or as a triple combination, greatly enhances the effect of metformin. We observed that metformin decreases MCF7 cell viability especially after 48 hours' incubation while SB and CPT alone and in combination together even in combination with metformin were not able to inhibit cancer cell growth.

Keywords: Camptothecin; Metformin; P38 α MAP kinase; NR4A; SB203580



Evaluation of the Expressed MiR-129 and MiR-549a in Patients with Multiple Sclerosis

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Background and Aim: Background: The expression of microRNAs (miRNAs) as circulating biomarkers has been underlined in multiple sclerosis (MS) in the last decade. Due to the presence of a possible relationship between expressed miRNAs and heterogeneous appearances of the pathological processes in MS, the present study attempts to evaluate the expression of miR-129 and miR-549a in patients with MS in comparison with healthy control (HC) group.

Methods: Materials and Methods: Peripheral blood mononuclear cells were separated from fifty patients with MS (subtypes including relapsing-remitting MS and secondary progressive MS) in the Kashani Hospital, Isfahan, Iran, and fifty people as HC group. After RNA extraction and complementary DNA synthesis, the expression of miR-129 and miR-549a was evaluated in patients with MS in comparison with the HC group using a quantitative real-time polymerase chain reaction assay. The data were analyzed using the Kolmogorov–Smirnov, and Mann–Whitney tests. Spearman’s correlation coefficient was used to examine the relationship between miR-129 and miR-549a with age.

Results: Results: The results showed that the expression of miR-129 and miR-549a was not significant in patients with MS in comparison with the HC group. The relationship between such miRNAs and age and gender was not significant.

Conclusion: We suggest the expression of miR-129 and miR-549a as circulating miRNAs in peripheral blood mononuclear cells could not be considered a biomarker for diagnosis and Para clinical.

Keywords: MicroRNAs, miR-129, human, miR-549a, multiple sclerosis

Evaluation of the Immunogenicity Effect of Chitosan Nanoparticles Containing Recombinant Protein of ml1-stxb in Mice

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Background and Aim: Ribosome inactivating proteins (RIPs) are toxic proteins with characteristic N-glycosidase activity. They remove a specific adenine (depurination) from the 28S rRNA of the eukaryotic ribosome, preventing its binding with elongation factor, and thereby inhibit translation. One of these proteins is called ML1 (Mistletoe lectin 1) which is a member of RIP 2 family. These combinations can be used as active biological factors and anticancer because of their ability in destroying cells by stopping protein production. StxB (Shiga toxin B-subunit) connects to its own cell receptor, globotriaosylceramide (Gb3). There is Gb3 On the surface of many cells in our body. Researches have shown that the binding domain of the ml1-stxb a high immunogenicity effect that could be used as an efficient recombinant vaccine. The recombinant vaccines are not potent enough to stimulate the immune responses, therefore, the use of biocompatible and safe adjuvants is inevitable. In recent years, there have been many studies on the adjuvant effect of nanoparticles and their role as delivery vehicles of recombinant vaccines. This study investigated the vaccine delivery effect of chitosan nanoparticles for administration of a recombinant candidate vaccine against ml1-stxb.

Methods: The expression and purification of ml1-stxb was done in *E. coli* BL21 (DE3). Chitosan nanoparticles were synthesized by ionic gelation method. The protein containing nanoparticles were administered to mice subcutaneously. At the same time, a control group received the candidate vaccine in combination with the Freund adjuvant. Finally, the IgG titers of immunized mice were assayed by ELISA.

Results: The recombinant protein was successfully expressed and purified and was subsequently confirmed by western blot analysis. The loading capacity of the recombinant antigen into nanocapsules was calculated as 89%. ELISA results showed an increase in the IgG titer after each administration.

Conclusion: The present study shows that the chitosan nanoparticles containing a recombinant vaccine could efficiently stimulate the humoral immune responses.

Keywords: Immunogenicity; toxins; cytotoxicity; ML1-STxB

Evaluation of the Immunomodulatory Properties of Curcumin on the Cytokines Level in Patients with Osteoarthritis; a Double Blind Randomized Clinical Trial

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Background and Aim: Osteoarthritis (OA) is the most common joint disease that its incidence directly related with aging. Although the underlying cause of the disease has not yet been determined, the results of various studies have shown the important role of T lymphocytes and their cytokines in the progression of this disease. Given the anti-inflammatory effects of curcumin, the aim of this study was to investigate the effects of this compound in nano particle form on T cells cytokines involved in the immune system functions of OA patients.

Methods: Thirty female patients with OA were randomly divided into the two groups, intervention group who received nano-curcumin (15 patients) and placebo group (15 patients). After three months of study, the concentration of cytokines IL-4, IL-10, IL-17, IL-23, TNF- α , CCL2 and CCL5 in the serum of patients were measured via ELISA.

Results: Serum levels of cytokines including IL-4 and IL-10 increased insignificantly in the curcumin receivers, while serum levels of IL-17, IL-23, TNF- α , CCL2, and CCL5 had an insignificant decrease. In the placebo group, serum levels of IL-4, IL-10, and CCL5 showed an insignificant decrease, but the level of IL-17, IL-23, TNF- α and CCL2 chemokine, showed an insignificant increase.

Conclusion: Curcumin effects on the production of these cytokines, indicating a change in the cytokine profile from inflammatory towards anti-inflammatory conditions. Therefore, it can be considered that this drug can play a role in improving the inflammatory conditions in the joints of osteoarthritis patients. Since the changes were not significant, it might be concluded that measuring the systemic level of these cytokines and chemokines cannot show an accurate image of the effects of curcumin on these factors.

Keywords: Osteoarthritis; Curcumin; T lymphocyte; Cytokine

Evaluation of the Performance of Corona Genomic Vaccines and Comparison with Other Types.

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Background and Aim: Vaccines usually need years of research and examination before reaching the clinic, vaccines that deliver numbers of the coronaviruses own genes into human cells to stimulate an immune response. vaccines are divided into genetic and non-genetic in terms of type. each of which follows a different method of operation.

Methods: we used research sites such as PubMed, and ScienceDirect and applied time filters to articles from the last five years by importing keywords: Corona; Corona vaccine; Corona genetic vaccine; DNA; RNA.

Results: This review is about examining the types of genetic and non-genetic coronavirus vaccines (SARS-CoV2). Various platforms for the development of Covid 19 vaccines are being explored including RNA, DNA, and inactivated vaccines. RNA-based vaccines like Moderna; this mRNA vaccine encodes the spike protein. Protein S is directed by host cell proteases and is subdivided into two subunits: S1 and S2. Protein S acts as the most vulnerable target for the development of the Covid vaccine. SARS-CoV-2S (S-Trimer) protein smoothing device, specific regions of the S protein such as subunits S2, RBD, NTD, and FP as potential targets. SARS-CoV2 binds to the same host cell receptor, ACE-2.

Conclusion: In this paper, we conclude that a vaccine is a biological preparation designed to protect humans from viral and bacterial infections. There are some genomic and non-genomic vaccines are being studied for corona. DNA and RNA, both platforms do not require bioreactor culture techniques, vaccines based on inactivated virus technology, on the other hand, are already licensed but do not produce a strong immune response unless used in conjunction with, for example, an aluminum adjuvant. DNA and RNA-based vaccines are made from the viral sequence of the virus. Viral vaccines use another virus, such as adenovirus, and insert the SARS-CoV-2 genetic material into their genome.

Keywords: Genetic vaccines; RNA vaccine; Inactivated vaccine; DNA vaccine; SARS-CoV2

Evaluation of the Relationship Between Alleles in Rheumatoid Arthritis Patients Based on the Response Pattern to Treatment

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Background and Aim: Rheumatoid arthritis (RA) is an autoimmune inflammatory rheumatic disease that influences about 0.5–1 % of the population and causes chronic synovial inflammation bring about joint damage and disability. Although the cause of RA is still unknown, genetic, environmental, and autoimmune factors cause the onset and persistence of RA underlying synovitis. One of most important genetic factors is HLA that it involved in many autoimmune diseases especially RA. This study aimed to explore the associations of HLA-DRB1/-DQB1 alleles and haplotypes with RA risk and the response pattern to treatment in these patients.

Methods: A total of 167 RA patients and 153 ethnically matched healthy controls were enrolled. the genotypes of HLA-DRB1 and -DQB1 alleles for all patients and healthy control subjects were determined by PCR-SSP method by using low-resolution HLA-DR-DQ SSP kits. Then, the genotype of some of most important HLA-DRB1 and -DQB1 alleles such as HLA-DRB1*04, HLA-DRB1*10, HLA-DQB1*03, HLA-DQB1*04 and HLA-DQB1*05 for patients were determined by PCR-SSP method by using high-resolution HLA-DR-DQ SSP kits. In addition, DAS28 was used as an indicator of disease activity and clinical response to therapy. Patients with $\text{DAS28} \leq 3.2$ were classified as good-responders (GR) and patients with $\text{DAS28} > 3.2$ were categorized as poor-responders (PR) groups.

Results: Analysis of HLA-DRB1 and -DQ alleles frequencies among RA patients and healthy controls revealed higher significantly frequencies of DRB1*04 and DRB1*10 alleles and lower frequencies of DRB1*13 allele in the RA patients versus healthy controls. Also, among DRB1-DQB1 haplotypes, DRB1*04-DQB1*03 ($p=0.004$, $\text{OR}=2.081$), DRB1*04-DQB1*04 ($P=0.03$, $\text{OR}=1.935$) and DRB1*10-DQB1*05 ($p=5 \times 10^{-4}$, $\text{OR}=3.947$) haplotypes showed significant statistical increase in RA patients compared to healthy controls. In contrast, we found that DRB1*13-DQB1*06 ($p=0.01$, $\text{OR}=0.467$) haplotype significant statistical decrease in RA patients compared to healthy controls. furthermore, among HLA-DRB1 and -DQB1 alleles and haplotypes, we found significant statistical association between HLA-DRB1*04:05 ($p=0.006$, $\text{OR}=0.445$), HLA-DQB1*06:02 ($p=0.01$, $\text{OR}=0.502$) in PR group compared to GR group. In contrast, among HLA-DRB1 and -DQB1 alleles and haplotypes, only DRB1*11-DQB1*03 haplotype have a significant relationship in GR group compared to PR group.



Conclusion: We identified DRB1*04 and DRB1*10 alleles (SE alleles) as risk alleles and DRB1*13 allele with a decreased risk for RA disease. Moreover, we found a close link between genetic susceptibility for RA and the response pattern to treatment that was more evident for DRB1*04, DQB1*06 alleles in PR response and DRB1*11 allele in GR group.

Keywords: Rheumatoid arthritis; HLA-DRB1; HLA-DQB1; Good Response; Poor Response



Evaluation of *TNF α IP3* Gene Expression in PBMCs of Patients with Coronary Artery Disease

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Background and Aim: Coronary artery disease (CAD) which is a multistep, inflammatory disease, has become the leading cause of death worldwide. Inflammation has long been recognized as a key factor in the progression of CAD. Some crucial inflammatory genes are regulated post-transcriptionally in this pathway, which may aid in the formation of atherosclerotic plaques. Therefore, the aim of this study was to evaluate the expression of *TNF α IP3* in the peripheral blood mononuclear cells (PBMCs) of CAD group compared with the control group.

Methods: A total of 168 subjects (84 CAD subjects and 84 control subjects) were included in this study. Expression level of *TNF α IP3* in PBMCs was measured using the real-time PCR technique.

Results: A comparison of the CAD group with the control group indicated a significantly reduced expression level of *TNF α IP3*. Also, there was a statistical correlation between the numbers of clogged arteries with the expression level of *TNF α IP3* in the CAD group.

Conclusion: In patients with CAD, decreased expression of *TNF α IP3* gene may have a role in the progression of atherosclerotic plaques.

Keywords: Coronary Artery Disease; Atherosclerosis; Inflammation; *TNF α IP3*

Evaluation the Gene Expression of MDA-5 and Type I Interferons in Patients with Mild and Severe Forms of COVID-19

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Background and Aim: Severe coronavirus disease-2019 (COVID-19) is associated with dysregulated immune response and hyper inflammation, which may lead to severe pneumonia as well as acute respiratory distress syndrome and subsequently death. Cytosolic nucleic acid sensors belonging to the RIG-I-Like Receptor (RLR) family have been recognized as the main pattern recognition receptors (PRRs) that sense coronaviruses. Cytosolic RNA sensor melanoma differentiation-associated protein 5 (MDA5) is one signaling receptor in this family. It is essential for inducing type I interferons (IFNs). IFN remarkably inhibited viral replication and production of infectious virions. Considering the prominent role of MDA-5 and IFN-I, this study was conducted to investigate the MDA-5, IFN- α , and IFN- β contribution to the pathogenesis of patients with mild and severe forms of COVID-19.

Methods: forty-five patients with mild and 45 patients with a severe form of COVID-19 were enrolled in this study. Peripheral blood was taken, RNA was extracted from sample patients and then converted to cDNA. The gene expression levels of MDA5, IFN- α , and IFN- β were measured using a real-time PCR.

Results: The gene expression levels of MDA5 and IFN- β were significantly lower in the severe form of COVID—19 compared to the patients with mild form COVID-19 ($P=0.021$, $P=0.032$, respectively). In contrast, IFN- α had no remarkable difference between the two studies mentioned above groups ($P=0.088$).

Conclusion: In sum, MDA5 can be considered as an important cellular sensor for SARS-CoV-2 infection that induced IFN- I, implicating clearance infection.

Keywords: Keywords: MDA5; IFN- α ; IFN- β ; COVID-19

Exosomes Derived from Serum of Patient with Glioblastoma Showed Immune Regulatory Properties on Lymphocytes and Monocytes in Vitro.

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Background and Aim: Exosomes are bilayer extracellular vesicles with a diameter about 30-150 nm that contain miRNAs, proteins, and other intracellular matrix components. These vesicles are secreted by nearly all of the live cells as an intercellular connection mechanism. Depended on their source and the type of targeted cells, exosomes have different effects on their targets. By this way, exosomes have conventional effects on cancers. They can support or suppress cancer growth. In this study, we assessed the effect of exosomes derived from the serum of patients with Glioblastoma on lymphocyte proliferation and phagocytosis ability of monocytes.

Methods: We collected serum from patient with glioblastoma and extracted exosomes. We characterized the exosomes by Dynamic Light Scattering for their size, transmission and scanning electron microscopy for their morphology and flow cytometry for assessment of common exosome markers. Then we obtained PBMCs from blood sample of healthy donors by ficoll and separated monocytes by adherent properties to cell culture plate. We also cultured lymphocytes and assessed the effect of serum derived exosomes on their proliferation by MTT assay kit. We cultured monocytes in 96 well plate and treated them by the exosomes. After 48 hours, we measured phagocytosis ability of the monocytes.

Results: Our data showed that exosomes from serum of patients with Glioblastoma lead to down regulation of in vitro lymphocytes proliferation. These exosomes also reduced phagocytosis of yeast by monocytes.

Conclusion: In these cases, exosomes derived from serum of patients with glioblastoma had suppressive effects on immune system. Additional studies would be needed to determine the immunoregulatory effect of these exosomes on other parts of immune system.

Keywords: serum derived exosomes; Glioblastoma; immune system; immune regulatory;

Exosomes from Glioblastoma Derived MSCs Showed in Vitro Immune Regulatory Properties against Lymphocytes and Monocytes

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Background and Aim: Mesenchymal stem cells (MSCs) can be found in already all tissues. MSCs have also been discovered in the majority of solid tumor tissues. These cells contribute to various biological functions such as wound healing and tissue hemostasis by interacting with other cells and releasing secretory factors such as cytokines, growth factors, and exosomes. Exosomes are bilayer extracellular vesicles with a diameter of 30-150 nm. These vesicles contain miRNAs, proteins, and other intracellular matrix components. Deferent studies have been shown that exosomes have controversial effects on immune system according to their source.

Methods: In this study, we isolated MSCs from Glioblastoma samples and cultured them. MSCs were characterized by their ability to differentiate into adipocytes and osteocytes. We also confirmed the presence of the most common MSC markers on these cells. We collected serum free culture media of MSCs and then extracted exosomes. Exosome was characterized by dynamic light scattering, transmission and scanning electron microscopy. PBMCs were obtained from circulating blood of healthy donors by ficoll and monocytes were separated by their adherent properties to cell culture plate. We cultured monocytes in 96 well plate and treated them by the exosomes. After 48 hours, we measured phagocytosis ability of these monocytes. We also assessed the effect of MSCs derived exosomes on the proliferation of lymphocytes by MTT assay kit.

Results: Our data showed that exosomes from Glioblastoma derived MSCs significantly reduced phagocytosis of yeast by monocytes. These exosomes also led to downregulation of in vitro lymphocytes proliferation.

Conclusion: In these cases, exosomes derived from MSCs of Glioblastoma had suppressive effects on immune system. Additional studies would be more effective to determine the correct effect of these exosomes on other parts of immune system.

Keywords: tumor associated mesenchymal stem cells; MSC derived exosomes; Glioblastoma; immune system; immune regulatory;

Expression Analysis of ANRIL and H19 lncRNAs in Peripheral Blood Mononuclear Cells from Relapsing-Remitting Multiple Sclerosis Patients

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Background and Aim: Long noncoding RNAs (lncRNAs) have important roles in regulating autoimmunity, and immunity balance. In multiple sclerosis (MS) disease, the ability of immune system to set a balance between pro-inflammatory and anti-inflammatory responses is lost. Disturbance in the balance between Th17/Treg cells plays a key role in development and progression of MS. In this study, we evaluated expression levels of lncRNAs involved in induction of Th17 development including ANRIL and H19; as well as expression of IL-6, in patients with MS.

Methods: The quantitative Real-time PCR method was used to assessment of the relative expression of lncRNAs and IL-6 mRNA in peripheral blood mononuclear cells (PBMCs) of 60 relapsing-remitting MS (RRMS) patients in comparison with 30 control subjects.

Results: It was evidenced, H19 was up-regulated in relapse phase patients compared to remitting phase patients and controls. However, ANRIL was increased in relapse phase patients compared to controls, but was decreased compared to remitting phase patients.

Conclusion: Findings of this study revealed that, H19 was related to disease attacks in patients with RRMS, while ANRIL was related to both relapse and remitting phases of disease. Importantly, at relapse phase of RRMS, up-regulation of ANRIL and H19 was positively correlated with the overexpression of IL-6.

Keywords: Relapsing-remitting MS (RRMS); Peripheral Blood Mononuclear Cells (PBMCs); ANRIL; H19

Ferroptosis Plays an Important Role in Triggering Inflammation in lung Ischemia-Reperfusion Sterile Injury

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Background and Aim: Lung ischemia-reperfusion (IR) injury is a form of sterile inflammatory lung injury that can cause acute respiratory distress syndrome. Ferroptosis is a novel form of cell death and has been studied in various diseases including cancer. Ferroptosis is known to be caused by the accumulation of lipid peroxides. However, the role of ferroptosis in IR, especially in the lung, is not well understood. Considering the unique conditions required for ferroptosis and those caused by IR, we hypothesized that ischemic conditions cause the accumulation of lipid peroxides, leading to ferroptosis in lung alveolar epithelium immediately after IR.

Methods: A mouse surgical model was applied to induce lung IR injury in vivo. In the IR group, the left pulmonary artery was occluded for 1 h without bronchial involvement (and therefore no hypoxia) and then lung was reperfused for 30 min. In the liproxstatin-1 (ferroptosis inhibitor) group, 10 mg/kg liproxstatin-1 was given intraperitoneally 1 h before ischemia induction. IR injury in vitro was mimicked by depriving normal human bronchial epithelial cells (NHBE) of nutrients for 1 h and then feeding the cells for 2 h. Liproxstatin-1 (200 nM) was administered in vitro 10 h before ischemia induction.

Results: Lung IR injury in vivo significantly enhanced IL-6 levels in plasma and lung tissue. Furthermore, the induction of IR in vitro markedly increased CXCL-1 levels in the supernatants of NHBE cells. The images were taken with a confocal microscope also showed robust probe oxidation of NHBE cells subjected to IR injury. The inhibition of ferroptosis by liproxstatin-1 in vivo and in vitro significantly returned all to the levels measured before ischemia induction.

Conclusion: The findings reveal that ferroptosis may play a crucial role in lung IR injury since the inhibition of ferroptosis attenuates inflammation. Therefore, this study suggests a unique mechanistic approach for lung IR injury treatment.

Keywords: Ferroptosis; Inflammation; Ischemia-reperfusion

Genetic Variations in Chemerin Receptor 23 in Allergic Rhinitis

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Background and Aim: The most common upper inflammatory disorder, allergic rhinitis (AR), is caused by aberrant immune responses to allergens in genetically predisposed individuals and is characterized by symptoms of nasal congestion, nasal itch, and sneezing. Inflammation is resolved by essential fatty-acid-derived immunoresolvents such as lipoxins, resolvins, protectins, and maresins. Resolvins play a significant role in resolving allergic inflammation by binding to Chemerin Receptor 23 (ChemR23). As a result, genetic studies of ChemR23 polymorphisms may provide valuable insight into AR pathogenesis. In this study, we assessed the possible correlation between single nucleotide polymorphisms (SNPs) of the ChemR23 gene and AR risk in the Kermanshah population of Iran.

Methods: Using polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP), we genotyped four SNPs in the chemR23 gene (rs4373981 G>C, rs73201532 C>T, rs35121177 G>A, rs4964676 G>A) in 130 patients with AR and 130 healthy controls.

Results: The distribution of allele and genotype frequencies of four SNPs in the ChemR23 gene (rs4373981 G>C, rs73201532 C>T, rs35121177 G>A, rs4964676 G>A) did not show any association with AR ($p>0.05$).

Conclusion: We investigated the association between SNPs in the ChemR23 gene and AR for the first time. We did not observe any correlation between four genetic variants of ChemR23 and AR in an Iranian population; however, it is vital to repeat our study with additional SNPs and design a more expensive study with larger sample sizes from different ethnic groups.

Keywords: Allergic rhinitis; Chemerin Receptor 23; Single nucleotide polymorphisms; PCR-RFLP

Immunogenicity of the Inactivated SARS-CoV-2 Vaccine (BBIBP-CorV) in Hemodialysis Patients: a Case-Control Study

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Background and Aim: The vaccines were known as the most important way to protect people against COVID-19 and perhaps the ultimate solution to this pandemic. Neutralizing antibodies (NA) are suggested to correlate with the degree of protection against COVID-19. This study aimed to evaluate the efficacy of Sinopharm vaccines in hemodialysis patients in Birjand.

Methods: The study population included all hemodialysis patients in the Special Diseases Center of Birjand. Inclusion criteria were signing written informed consent as well as completeness of vaccination with Sinopharm vaccines (two doses at standard intervals). Exclusion criteria were consumption of immunosuppressive drugs. The control group was also selected from the staff of Birjand University of Medical Sciences immunized with Sinopharm vaccines, who had no history of chronic diseases. To assess the level of anti-SARS-CoV-2 NA, a commercial IgG-specific antibody detection kit against SARS-CoV-2 RBD was used. This kit can detect RBD and ACE-2 reaction inhibitory antibodies by indirect ELISA. The specificity and sensitivity of this kit were reported 100%. The obtained information was entered into SPSS v.16. Seropositivity significant differences were analyzed by chi-square test at the significant level of P-value less than 0.05.

Results: This study was performed on 63 controls, and 101 hemodialysis patients. Hypertension (69.3%) and diabetes mellitus (37.6%) were the most common causes of hemodialysis. In case group, the rate of seropositivity for NA two weeks and six weeks after receiving the second dose were 54.2 %and 46.7%, respectively (P-value equal 0.001). However, in the control group, the rate of seropositivity two weeks after receiving the second dose was 73.0%. The difference in seropositivity rates two weeks after receiving the second dose in controls and cases was statistically significant (P-value equal 0.017).

Conclusion: Anti-SARS-CoV-2 NA didn't increase significantly in hemodialysis patients after receiving two doses of the Sinopharm vaccine; moreover, seropositivity rate decreased rapidly. As a result, the injection of the booster dose might be necessary for hemodialysis patients.

Keywords: COVID-19 Vaccines; Hemodialysis; Neutralizing antibodies

Immunomodulatory Effects of Curcumin in Systemic Autoimmune Diseases

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Background and Aim: Systemic autoimmune diseases like rheumatoid arthritis, multiple sclerosis, and systemic lupus erythematosus represent various autoimmune conditions identified by immune system dysregulation. The activation of immune cells, auto-antigen outbreak, inflammation, and multi-organ impairment are observed in these disorders. The immune system is an essential complex network of cells and chemical mediators, which defends the organism's integrity against foreign microorganisms and its precise operation and stability are compulsory to avoid a wide range of medical complications. Curcumin is a phenolic ingredient extracted from turmeric and belongs to the Zingiberaceae, or ginger family. Curcumin has multiple functions, such as inhibiting inflammation, oxidative stress, tumor cell proliferation, cell death, and infection.

Methods: We observed studies that evaluated the relationship between curcumin and systemic autoimmune diseases. The following databases were searched up to May 2021: PubMed, Google Scholar, Scopus, Ovid-Medline, and Web of Science, and no limits about period or type of work were utilized. Case reports, clinical trials, original research, and review articles were thus evaluated

Results: Nevertheless, the immunomodulatory influence of curcumin on immunological reactions/processes remains mostly unknown.

Conclusion: In the present narrative review, we sought to provide current information concerning the preclinical and clinical uses of curcumin in systemic autoimmune diseases

Keywords: Immunomodulatory Effect; Curcumin; Systemic autoimmune diseases; Inflammation

Introducing a New Biomarker for the Diagnosis and Monitoring of Breast Cancer

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Background and Aim: Breast cancer is one of the most lethal diseases among women in the world. Breast cancer cells invade and metastasize in a variety of ways, one of which is the secretion of small vesicles or exosomes. By secreting exosomes containing microRNAs, cancer cells can transmit their messages to distant sites and can also help metastasize during the epithelial-mesenchymal transition (EMT) process. Early detection of this type of cancer is helpful in its therapy, and finding new biomarkers will help monitor the process of this cancer. This study aimed to determine the level of miRNA-10b-5p expression, which is associated with EMT, in serum-derived exosomes of breast cancer patients in different grades to introduce a new biomarker.

Methods: In this study, blood was taken from 30 cases of breast cancer in three different grades (I, II, III) and five healthy individuals as a control group. Exosomes were purified from serum samples and miRNA-10b-5p expression was measured by real-time PCR method.

Results: miRNA-10b-5p expression was significantly increased in patients with breast cancer grades I, II, and III compared to the control group ($p < 0.0001$), ($p < 0.0001$) and ($p < 0.0001$), respectively. Also, miRNA-10b-5p expression was significantly increased in patients with grade III compared to the patient with grades I, II ($p < 0.0001$) and ($p < 0.0001$) respectively. And no significant difference was seen between the group of patients with grade I and the group of patients with grade II.

Conclusion: According to our study, miRNA-10b-5p in serum-derived exosomes of cancer patients could be a new biomarker in the diagnosis and monitoring of breast cancer.

Keywords: breast cancer; exosome; miRNA-10b-5p

Introducing a Simple High Yield Wharton's Jelly Of The Human Umbilical Cord-Derived Mesenchymal Stem Cell Isolation Technique

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Background and Aim: Mesenchymal stem cells (MSCs) are used in the treatment of many diseases due to their low immunogenicity and immunomodulatory properties. MSCs can be isolated from different tissues. The umbilical cord is becoming a viable alternative to bone marrow, as a source of MSCs. In order to meet the requirements of cell therapy, it is important to produce stem cells on a clinical scale, with less time, cost and contamination. Here is a simple and efficient protocol for isolating MSC from Wharton's jelly (WJ-MSC) by Explant method. In this study, the ability of cells to proliferate in the presence of various supplements including Basic fibroblast growth factor (bFGF), Non-essential amino acids (NEA) and L-Glutamine(L-Gln) in Alpha Minimum Essential Medium (α -MEM) and Dulbecco's Modified Eagle Medium/Nutrient Mixture F-12 (DMEM-F12) basic culture media were investigated.

Methods: Human umbilical cord Wharton's jelly was cultured in small pieces in a flask according to the Explant method. The nature of these cells was characterized by phenotyping through surface markers CD34, CD45, CD73, CD90 and CD105 as well as their ability to differentiate into adipocyte and osteocyte lineage. Then, in order to evaluate the ability of cells to proliferate by adding various supplements including bFGF, NEA and L-Gln to α -MEM and DMEM-F12 basic culture media.

Results: WJ-MSCs isolated by explant method was removed from the tissue after 7 days and entered the culture medium. These cells were positive for the expression of CD73, CD90 and CD105 and negative for CD34 and also differentiated into adipocyte and osteocyte lineage. The following results showed that relative to the control, the addition of bFGF in α -MEM and DMEM-F12 medium resulted in a significant increase in the proliferation of MSCs but no significant differences were observed for NEA and/or L-Gln.

Conclusion: This study reveals that although the presence of bFGF greatly improves cell proliferation, proper preparation of Wharton's Jelly of the human umbilical cord tissues can also contribute to the growth and proliferation of MSCs.

Keywords: Mesenchymal stem cell; Umbilical cord; Wharton's jelly; Explant; bFGF

Investigation of CAR T Cell Therapy Effect on Acute Lymphoblastic Leukemia: A Review Study

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Background and Aim: Acute lymphoblastic leukemia (ALL) is a relapsed and refractory disease. There are some therapeutic methods for acute lymphoblastic leukemia. One of these treatment procedures is CAR T cell therapy. In this study, we want to explain the effects of CAR T cell therapy on acute lymphoblastic leukemia.

Methods: This article is a review study by searching reputable databases including Google Scholar, PubMed, Scopus, Cochrane Library, Web of science and with the keywords "Chimeric antigen receptor", "CAR T cell", "Immunotherapy", "Acute lymphoblastic leukemia" to ensure the completion of the search results, the sources of the articles were reviewed, and after removing duplicate titles from the endnote software and reviewing the titles and abstracts, the articles related to the JBI tools were reviewed. The desired checklist was entered.

Results: Chimeric antigen receptor (CAR) is infused into T cells to express them. For acute lymphoblastic leukemia therapy, CAR T cells dedicated to CD19 link to the pan-B-marker CD19 to inhibit B cells. Based on recent studies, CAR T cell therapy has increased the rate of improvement in acute lymphoblastic leukemia patients.

Conclusion: CAR T cell therapy has promising acute lymphoblastic leukemia therapy results. We should study this method in a large number of people with different age categories and some more cancers to make more use of it.

Keywords: Chimeric antigen receptor; CAR T cell; Immunotherapy; Acute lymphoblastic leukemia

Kinetic of COVID-19-Specific Antibodies During Time among Recovered Patients

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Background and Aim: The COVID-19 pandemic has caused more than three million deaths worldwide. Although the exact mechanisms of immunity against SARS-COV2 are not yet fully understood, SARS-COV2 specific IgG and IgM antibodies are detectable in most of patients shortly after the start of the infection and as such considered a valuable tool for confirming infection, particularly in sero-epidemiological studies. Studying the kinetics of specific antibodies against SARS-COV2 gives valuable information about the durability of immune protection after COVID-19 infection or vaccination. The aim of the current study was to evaluate the kinetic of SARS-COV2 specific IgM and IgG and affected factors in a group of recovered patients.

Methods: COVID-19 patients confirmed by RT-PCR and positive IgM and IgG titers were enrolled in this study. The levels of SARS-COV2 specific IgG and IgM antibodies were measured by ELISA three and six months after the primary test.

Results: In total, 87 patients were enrolled in this study (mean age= 42.87 ± 12.29 , range = 14-85 years, M/F ratio=2.48). The mean titer of the first COVID-19 IgM and IgG after infection was 4.77 ± 4.09 and 10.12 ± 4.85 respectively. The mean titer of COVID-19 IgG was significantly reduced after three and months intervals; the severity and duration of disease as well as the primary titer of IgG was significantly higher in females than males but females showed a higher percentage of decline in IgG over time.

Conclusion: The results this study confirmed steady decline in the level of COVID-19 specific IgG during the time after natural infection.

Keywords: COVID-19; diagnosis; ELISA; IgM/IgG antibody; immune response; SARS-COV2; severe disease

Mesenchymal Stem Cells Derived Exosomes Decrease TH-17 Function in COVID-19 Patients PBMCs

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Background and Aim: The study of immune cell responses in the pathogenesis of Covid-19 disease is of particular importance. The production of inflammatory cytokines by these cells and some other cells in the body leads to a cytokine storm (CS). This overproduction of cytokines is responsible for a part of the pathogenesis of a disease called acute respiratory distress syndrome (ARDS), which involves the lung and impairs its function. TH-17 cells are responsible for at least part of the production of cytokines in ARDS. Modulating the responses of these cells can help prevent disease progression. Due to their immunomodulatory properties, mesenchymal stem cells (MSCs) can suppress the inflammatory response in PBMCs isolated from patients with Covid-19. However, due to the prohibitions on the use of cells as a treatment, their exosomes can be used as a substitute.

Methods: For this purpose, blood samples were first taken from patients with Covid-19, and their PBMCs were isolated. PBMCs were cocultured with MSCs exosomes, and the TH17 cells were evaluated before and after treatment. The expression of the transcription factor, associated cytokines, and their concentration by real-time PCR and ELISA, respectively.

Results: Our data revealed that MSCs exosomes considerably influence The expression levels of ROR γ t ($p < 0.0001$) and IL-17 ($p=0.0044$), and also secretion level of IL-17 ($p=0.0011$) was decreased in the treated group compared to the control group.

Conclusion: The current study results indicated that AD-MSCs derived exosomes can decrease the function and cytokine production of TH-17 cells in COVID-19 patient isolated PBMCs.

Keywords: MSCs; Exosome; PBMCs; TH-17

Mesenchymal Stem Cells MSCs-Derived Exosomes Improve Regulatory T-cell Frequency in COVID-19 Patients PBMCs

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Background and Aim: After the COVID-19 virus epidemic, many studies were conducted to find the role of immune system cells in this disease. One of these cells is Treg, which is important due to its role in regulating immune system responses' homeostasis. An imbalance occurs in Treg/Th17 cells' ratio during the pathogenesis of COVID-19 and shifts to Th17, resulting in uncontrolled inflammatory responses. Therefore, it can be beneficial to study the factors that can increase Treg responses and differentiation. Since adipose tissue-derived mesenchymal stem cell (AD-MSCs) exosomes have immunomodulatory properties, the use of these vesicles can be an excellent option to reduce inflammatory responses and induce anti-inflammatory responses in COVID-19 peripheral blood mononuclear cells.

Methods: In the present study, we isolated AD-MSCs exosomes by ultracentrifugation and investigated their immunomodulation effect on peripheral blood mononuclear cells (PBMCs) of covid-19 patients. For this purpose, after culturing the PBMCs with MSCs exosomes, the frequency of Treg lymphocytes was assessed by flow cytometry. PBMCs were washed and incubated with FITC-conjugated anti-CD4, PE-conjugated anti-CD25, and PE-cy7-conjugated anti-CD127 monoclonal antibodies isotype-matched IgG controls before staining. The exosomes were by and characterized by SEM, TEM, and DLS.

Results: A significant reduction was observed in the proportion of peripheral Treg cell frequency in COVID-19 patient PBMCs. Our data revealed that the ratio of CD4⁺ CD25⁺ CD127⁻ Treg was considerably influenced by MSCs exosomes in all the patients.

Conclusion: The current study results indicated that AD-MSCs derived exosomes can restore the frequency of Treg cells of COVID-19 patient isolated PBMCs.

Keywords: MSCs; Exosome; COVID-19; PBMCs; Treg

Micro RNA-34a Sensitizes MCF-7 Breast Cancer Cells to Carboplatin via the Apoptosis Induction

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Background and Aim: Breast cancer (BC) has been classified among the main causes of death owing to females' cancer. Carboplatin is a platinum-based chemotherapeutic drug that is an important treatment option for BC. But high and frequent doses of carboplatin usually reduce the reaction of cancer cells to medication. There is an immediate need to establish methods for increasing the carboplatin susceptibility to BC cells. For instance, micro RNAs (miRNAs) such as MiR34a demonstrate significant potential. Considering that, this research was planned to explore the better clinical effect and underlying mechanism of miR-34a as a possible tumor inhibitor and drug resistance regulator in comparison with carboplatin chemotherapy drug in the cell lines of BC in humans.

Methods: MCF-7 cell line was transfected with miR-34a to perform functional analyses. Subsequently, the MTT assay was applied to assess cell viability. Cell viability and cell death-associated gene expression amounts including Bax, Bcl-2, caspase-3, MDR1, P53, and mir34-a, were examined through quantitative real-time PCR.

Results: Findings showed that miR-34a upregulation significantly decreased MCF7 cell viability in comparison with the control group. Furthermore, the single treatment of cells with miR-34a mimics and carboplatin could significantly increase Bax, Caspase-3, P53, and decrease Bcl-2 mRNA expression levels compared to the non-treated group. Moreover, by reducing the expression levels of the MDR1 gene, BC cells' reaction to carboplatin has been increased via miR-34a.

Conclusion: This study showed, that miR-34a restoration might improve the responsiveness of breast cancer cells to carboplatin chemotherapy through the downregulation of MDR1.

Keywords: Breast cancer; miR-34a; carboplatin; Apoptosis; Chemosensitivity

MicroRNA-30c Delivered by Bone Marrow-Mesenchymal Stem Cells Induced Apoptosis and Diminished Cell Invasion in U-251 Glioblastoma Cell Line

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Background and Aim: Glioblastoma multiform (GBM) is the most belligerent and prevalent brain malignancy among adults. Due to the blood brain barrier (BBB), drug administration is confronted by massive challenges which make the resectional surgery the only treatment pipeline. MicroRNAs have recently absorbed the attention of studies for their ability to correlate with the progression of various malignancies. miR-30c has been reported to play role in cell proliferation, metabolism, and apoptosis process. For instance, miR-30c has been reported to regulate apoptosis through the TNF-related apoptosis-inducing ligand (TRAIL). miR-30c also targets IL-6 which further induces apoptosis. Besides, miR-30c inhibits glioma proliferation and their migratory ability. In addition, the overexpression of miR-30c arrested cells at G0 as well as dampening their migration and invasion. However, it has been shown that the expression level of miR-30c was low in glioma. MSCs have the ability to migrate toward tumor cells which is called tumor-tropism, in which they are capable of delivering engineered miR-30c based on gap junction and non-intimacy mechanisms.

Methods: MiR-30c was cloned into pCDH-CMV-MCS-EF1-copGFP vector utilizing XbaI and EcoRI in order to construct pCDH-miR-30c. Then psPAX2, pMD2.G, and pCDH-miR-30c were co-transfected into Hek-293T to yield lenti-miR-30c virus particles. Next, bone marrow-mesenchymal stem cells (BM-MSCs) were Transduced with lenti-miR-30c. Thereafter, we co-cultured U-251 cell line with BM-MSCs-miR-30c and evaluated the apoptosis rate and the relative expression level of IL-6, Klf4, Sox2, c-Myc, and Oct4 using Real-Time PCR and flow cytometry.

Results: Wound healing assays represented low migratory ability in U-251 cells treated with BM-MSCs-miR-30c. Plus, apoptosis assay using Annexin V/7AAD showed an increased number of apoptotic U-251 cells following the treatment. miR-30 targeted IL-6 and induced apoptosis. It also impacted on self-renewal and anti-apoptotic cluster of genes, namely Klf4, Sox2, c-Myc, and Oct4, to induce apoptosis and dwindle the migration and invasion.

Conclusion: In conclusion, therefore, in this study we have demonstrated that delivering miR-30c using BM-MSCs machinery is a successful tool regarding the possible gene therapy targets. We could manage to represent data in the context of apoptosis induction in U-251 cells which were co-cultured with BM-MSCs-miR-30c using various in-vitro approaches.

Keywords: Glioblastoma multiform (GBM); miR30c; apoptosis; migration; IL-6; Klf4; Sox2; c-Myc; Oct4

Modulatory Effect of Probiotics on Proinflammatory Cytokine Levels in Acrylamide-Treated Rats

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Background and Aim: The aims of this study are to investigate the effect of acrylamide on the level of proinflammatory cytokines in the blood of acrylamide-treated rats and to find the modulatory impact of probiotics on those cytokines.

Methods: Thirty-two rats were divided into four groups: rats which received 20 mg acrylamide, acrylamide with 20 mg probiotics, acrylamide with 200 mg probiotics, and standard water and food (groups 1–4, respectively). The serum levels of cytokines were measured on days 0, 15, and 30.

Results: Group 1 showed an increased serum level of IL-1 β , IL-6, and TNF- α after 15 days, and they decreased on day 30. Serum IL-6 level was significantly decreased on days 15 and 30 in rats in group 2 compared to the controls. TNF- α and IL-1 β levels were not statistically different after treated with probiotics.

Conclusion: The exposure of rats to acrylamide led to increased systemic inflammation as evidenced by higher levels of proinflammatory cytokines, and probiotics can modulate this inflammation.

Keywords: Acrylamide; Cytokines; Inflammation; Microorganisms; Probiotics; Rats

NGAL as a New Biomarker for Diagnosis and Prognosis of COVID-19 Patients with AKI: A Systematic Review

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Background and Aim: Despite the improvement in COVID-19 management, optimal treatment for critically ill patients still has remained controversial. One of the major risk factors and complications of critically ill patients is acute kidney injury (AKI) so that about 10-30 % of ICU patients develop AKI. Moreover, neutrophil gelatinase-associated lipocalin (NGAL) is an early biomarker for inflammation which has been introduced as a biomarker for the diagnosis and evaluation of COVID-19. This systematic review was performed to understand the capability of NGAL as a predictor of AKI in COVID-19.

Methods: Data were collected by searching keywords in PubMed, Scopus, and Google Scholar (2015- December 2021), and then about 15 relative studies were selected to evaluate the details of findings.

Results: Among different biomarkers, it is shown that urinary NGAL strongly correlates with AKI in ICU admitted patients with higher specificity and sensitivity rather than previous common laboratory tests like serum creatinine. Moreover, length of mechanical ventilation and mortality rate have a direct relation with urinary NGAL value in the first hours from ICU admission. Furthermore, as NGAL is a marker for distal tubular injury, it could indicate the location of specific segments injury in the kidney.

Conclusion: Our study demonstrates the predictive role of NGAL in the rate of kidney injury as well as the severity of clinical presentation of COVID-19 and higher properties rather than other traditional methods (like serum creatinine). In this regard, the evaluation of the NGAL level in severe cases could be critical for a proper and opportune decision for an efficient treatment strategy.

Keywords: NGAL; COVID-19; AKI; biomarker

Prognostic Power of Tumor Infiltrating Lymphocytes in Larynx Squamous Cell Carcinoma

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Background and Aim: The prognostic importance of tumor-infiltrating lymphocytes (TILs) in the tumor microenvironment of various cancers is increasingly recognized. In this regard, we retrospectively evaluated the prognostic value of CD3, CD4, CD8, and CD45RO positive cells infiltration in larynx squamous cell carcinoma (LSCC).

Methods: Formalin-Fixed and Paraffin-Embedded (FFPE) tissue blokes from 63 primary LSCC patients were immunostained for CD3, CD4, CD8, and CD45RO expression. An expert pathologist acquired digital micrographs from center of tumor (CT) and the invasive margin (IM) of each stained marker. Quantification of TILs staining was performed semi-automatically using ImageJ software. Association of stained markers expression with clinicopathologic parameters were evaluated by Non-parametric Mann-Whitney U test and their prognostic effect on disease-free survival (DFS) and overall survival (OS) were analyzed by univariate and multivariable Cox regression analysis.

Results: Among patients clinicopathologic parameters, tumor grade was associated with the CD3+ TILs level in IM ($P=0.037$) and CD4+ TILs in CT ($P=0.006$) in a way that patients with well differentiated tumors had a higher TILs level. Also higher level of CD4+ TILs associated with lower TNM stage ($P=0.043$). Results of univariate Cox regression indicated that higher infiltration of CD4+ cells in IM region had positive effect on both OS ($HR=3.706$, $P=0.045$) and DFS ($HR=3.588$, $P=0.050$). Also CD45RO+ cell in IM (OS: $HR=3.936$, $P=0.021$, DFS: $HR=3.927$, $P=0.021$) was significantly associated with disease outcomes. From multiple analyses, CD45RO+ cells in IM had independent prognostic significance on OS ($HR=5.136$, $P=0.016$) and DFS ($HR=5.102$, $P=0.016$).

Conclusion: TILs associated with patients clinicopathologic parameters in a way that higher infiltration links with better clinicopathologic reports, and affect their survival. CD45RO+ TILs are a valuable biomarker for predicting prognosis of LSCC patients and their assessments has the potential to be integrated into clinicopathological prognostic models for patients with LSCC.

Keywords: tumor-infiltrating lymphocytes; prognostic marker; larynx squamous cell carcinoma; CD45RO+ cells

Receptor-Binding Domain-Based Vaccine Induces Neutralising Antibodies against SARS-CoV-2: An Immunoinformatics Approach

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Background and Aim: Recently the outbreak of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has become a pervasive threat to public health so it is an emergency to vaccine development. The spike (S) glycoprotein SARS-CoV-2 contains two subunits, S1 with presents the receptor-binding domain (RBD) is responsible for binding, and then conformation change is necessary for fusion through the S2 subunit.

Methods: Considering the multi-epitope peptide vaccines are capable of inducing the specific humoral or cellular immune responses we investigated immunoinformatics analysis of the RBD-surface exposed B-cell epitope and then constructed with S1/S2 protease cleavage site and moreover, PADRE was applied as a universal T-helper activator.

Results: The designed vaccine showed appropriate physicochemical properties furthermore molecular docking and molecular dynamics of vaccine candidate with immunological receptor was satisfying.

Conclusion: The designed multi-epitope vaccine candidate showed up a promising potential to induce neutralizing antibodies against SARS-CoV-2.

Keywords: immunoinformatics; RBD; fusion; B-cell multi-epitope; SARS-CoV-2

Recombinant CD137-Fc, its Synthesis, and Applications for Improving the Immune System Functions, such as Tumor Immunotherapy and to Reduce the Inflammation due to the Novel Coronavirus

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Background and Aim: CD137 (ILA/4-1BB), a member of the tumor necrosis factor receptor superfamily, is one of the most important T cell costimulatory molecules. Interaction of this molecule with its ligand transmits a two-way signal that activates both T lymphocyte and antigen-presenting cells. The soluble form of CD137 (sCD137) reduces the activity of its membrane isoform and is associated with T lymphocyte activation induced cell death. Recombinant CD137-Fc may be used to treat cancers, autoimmune disorders and viral infections. It may also be useful for the management of coronavirus infection.

Methods: The 1276 bp DNA sequence encoded CD137-Fc recombinant protein was prepared and subcloned into the lentiviral vector and expressed in transduced CHO-K1 eukaryotic cells.

Results: The sodium dodecyl sulfate-polyacrylamide gel electrophoresis, Western blot analysis, and enzyme-linked immunosorbent assay analysis results demonstrated that the expression of the 70-kDa CD137-Fc molecule was detectable without any degradation.

Conclusion: This study helps to confirm previous research suggesting the use of this recombinant protein as a promising solution for the treatment of virus infections. CD137-Fc fusion protein could also make immunotherapy more effective for some diseases. This product is widely used in novel medical treatments, including cell-based immunotherapy such as dendritic cell, CAR T and CAR NK therapy. Its production and usage in research and treatment is noticeable also in current coronavirus disease 2019 pandemic.

Keywords: autoimmune disorders; cancer immunotherapy; coronavirus inflammation; recombinant protein CD137-Fc; sCD137

Recurrence of SARS-CoV-2 Infection in Recovered COVID-19 Patients

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Background and Aim: After the global SARS-CoV-2 pandemic, the amount of antibodies available after infection and the possibility of re-infection is an important concern for the whole community. The aim of this study was to investigate the recurrence of SARS-CoV-2 infection in recovered COVID-19 patients.

Methods: Re-infection was investigated in 248 recovered patients that we had evaluated their humoral immune responses. For re-infection evaluation, we did follow-up the patients up to 18 months and the PCR results of them were gathered through information drawn from the Hospital Information System.

Results: Among all patients, only four cases of re-infection were confirmed. The re-infection rate was 1.61%. Out of whom, three had positive serology tests and one had a negative serology test.

Conclusion: Based on the present study's results, it is possible to achieve a correct estimate of re-infection of the disease to manage the disease efficiently.

Keywords: Coronavirus; COVID-19; Seroprevalence; Immune response; Re-infection

Regulatory T Cell Landscapes in Corona Virus Disease 2019

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Background and Aim: Recently, several studies have been conducted regarding the role of regulatory T cells (Tregs) in immunopathogenesis of viral infections. Considering the anti-inflammatory nature of Tregs response, detailed understanding of the immunopathology of COVID-19 infection may give us more insight into the curative strategies. To broaden our knowledge, we systematically reviewed the current literature on the role of Tregs in COVID-19 disease.

Methods: PubMed and Google Scholar were searched. Data extraction, and quality assessment of the study were conducted. 133 studies assessed for eligibility and 14 studies entered the quality check. Finally, 12 studies were included in this systematic review.

Results: The number and function of Treg cells were mostly studied in peripheral blood in extremely severe, severe, moderate/mild COVID-19 patients. Treg cells were identified based on their cell surface markers including CD3, CD4, CD25, FoxP3 and CD127^{low}. Although the results of different studies were to some extent inconsistent, but a trend towards the decrease of Treg levels in severe COVID-19 patients was clearly seen.

Conclusion: This revealed that the function of Treg cells played a role in the pathogenesis of COVID-19. More studies with focus on the functional aspects of immune cells especially Tregs are required to unravel the potential role of Tregs in COVID-19 disease.

Keywords: COVID-19; Immune response; Regulatory T cell

Relevance of Autoantibody Profile with HLA-DRB1 and -DQB1 Alleles in Rheumatoid Arthritis Patients

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Background and Aim: One of the most relevant genetic components in Rheumatoid arthritis (RA) is human leukocyte antigen (HLA) gene complex which plays a central role in autoimmune responses. This study aimed to explore the associations of HLA-DRB1/-DQB1 alleles and haplotypes with RA risk and the appearance of autoantibodies such as Anti-CCP (ACPA) and Rheumatoid factor (RF) in RA disease.

Methods: A total of 167 RA patients and 153 ethnically matched healthy controls were enrolled. the genotypes of HLA-DRB1 and -DQB1 alleles for all patients and healthy control subjects were determined by PCR-SSP method by using low-resolution HLA-DR-DQ SSP kits. Then, the genotype of some of most important HLA-DRB1 and -DQB1 alleles such as HLA-DRB1*04, HLA-DRB1*10, HLA-DQB1*03, HLA-DQB1*04 and HLA-DQB1*05 for patients were determined by PCR-SSP method by using high-resolution HLA-DR-DQ SSP kits. In addition, autoantibodies titers were measured among RA patients by ELISA method. Finally, HLA alleles and haplotypes frequencies were compared between two groups and among the patients in terms of autoantibodies spectrum.

Results: Analysis of HLA-DRB1 and -DQ alleles frequencies among RA patients and healthy controls revealed higher significantly frequencies of DRB1*04 and DRB1*10 alleles and lower frequencies of DRB1*13 allele in the RA patients versus healthy controls. Also, among DRB1-DQB1 haplotypes, DRB1*04-DQB1*03 ($p=0.004$, $OR=2.081$), DRB1*04-DQB1*04 ($P=0.03$, $OR=1.935$) and DRB1*10-DQB1*05 ($p=5 \times 10^{-4}$, $OR=3.947$) haplotypes showed significant statistical increase in RA patients compared to healthy controls. In contrast, we found that DRB1*13-DQB1*06 ($p=0.01$, $OR=0.467$) haplotype significant statistical decrease in RA patients compared to healthy controls. furthermore, among HLA-DRB1 and -DQB1 alleles and haplotypes, we found significant statistical association between HLA-DRB1*11 ($p=0.006$, $OR=0.445$), HLA-DQB1*03 ($p=0.01$, $OR=0.502$), DRB1*04-DQB1*03 ($p=0.03$, $OR=2.133$) and DRB1*11-DQB1*03 ($p=0.004$, $OR=0.428$) with status of ACPA in RA patients.

Conclusion: We identified DRB1*04 and DRB1*10 alleles (SE alleles) as risk alleles and DRB1*13 allele with a decreased risk for RA disease. More importantly, we found a close link between genetic susceptibility for RA and status of autoantibodies that was more evident for DRB1*04 and DRB1*11 alleles.

Keywords: Rheumatoid arthritis; HLA-DRB1; HLA-DQB1; ACPA; RF

Rheumatoid Arthritis is Associated with KIR2DS4-Full among the KIR Genes in Lur Population of Iran

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Background and Aim: Among the approaches to autoimmune disease, especially Rheumatoid Arthritis (RA), we can mention the important role of immune system and the molecules involved. Previously, different types of genetic elements were studied in connection with this disease, however, few studies have been performed on Killer cell Immunoglobulin-like Receptors (KIR) and Human Leukocyte Antigen (HLA) molecules, and in particular their compounds. Therefore, we intend to examine KIR genes, their HLA ligands, and KIR-HLA compounds in patients with RA and healthy controls.

Methods: In this case-control study, 50 patients with RA and 100 healthy individuals were included in the study. DNA samples were evaluated based on PCR method with sequence specific Primers (PCR-SSP). Odds ratio (OR) with 95% confidence interval (CI) were reported.

Results: Results: Among the inhibitor genes KIR2DL5A ($P = 0.0255$, OR = 0.389, 95% CI = 0.210-0.722), and among the activating genes KIR2DS4-full ($P < 0.0001$, OR = 6.163, 95% CI = 3.174-11.968) had a significant association with susceptibility to RA. No significant association was found between KIR genotypes, HLA ligands, and KIR-HLA compounds with a susceptibility to RA.

Conclusion: In our population, KIR2DS4-full increased susceptibility to RA were as KIR2DL5A was a protecting factor based on both cross table and regression analyses. This study should be repeated in other populations.

Keywords: Rheumatoid Arthritis, NK cells, KIR, HLA

Seroepidemiology Survey of SARS-COV-2 IgG, IgM Antibodies in Patients of Ghiassi Hospital (Shahrak-e Valiasr, District 18, Tehran) in COVID-19 Pandemic from Tir to Esfand 1399/July to March 2021

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Background and Aim: The world is facing a new scenario of the COVID-19 pandemic in which there is an increasing request for antibody testing. **Material and Methods:** In our study we had 1000 blood specimen from patients who were admitted to Ghiassi Hospital, Tehran, Iran, or were required performance of serological SARS-COV-2 IgM, IgG test by their physicians, due to COVID-19 disease. The antibody levels were assessed via Enzyme Immunoassay using S and N antigens, during peaks of COVID-19 in Iran.

Methods: Elisa assay method Enzyme Immunoassay using S and N antigens

Results: IgG showed a significant mean increase among the patients during peak 3, while IgM showed a significant mean increase during peak 2. This might be because of the high disease activity in each peak and/or the time of patient's admission.

Conclusion: Detection of SARS-COV-2 IgG, IgM has potential significance for evaluating the severity and prognosis of COVID-19. In addition, all and every seroepidemiology data in each community can help Health Commissions all over the world for controlling this pandemic.

Keywords: SARS-COV-2 IgG, IgM, Covid-19, Elisa, Wave, Peak.

Suggesting a Bioinformatic Method to Design Epitope Vaccines

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Background and Aim: Nowadays, Bioinformatic and Immunoinformatic strategies suggested for the identification of structures that could trigger an effective immune response able of protecting against a specific disease. Epitope prediction is one of the most important bases of in-silico vaccine designing, nevertheless it depends on antigen identification, and most crucially epitope selection for an efficient immune response.

Methods: The first step is downloading specific genome sequences from Refseq in NCBI. By ORF finder we can find ORFs. In order to indicate localization of protein, PSORTb server can be used to find candidate extracellular and outer membrane proteins. Signal peptide sequence detection and removal were done by CBS server. Prediction of allergenic proteins and mapping of IgE epitope Allpred was used based on K nearest neighbors. For prediction SVM module based on amino acid composition, SVM module may be used in order to Allergenicity evaluation. Protein-Protein blast was used to check protein homology in human and mouse. VaxiJen and ToxinPred server apply to evaluate immunogenicity and toxicity. To remove transmembrane helix from the protein TMHMM server is applicable. Linear and conformational epitope prediction were done by IEDB and CBTOPE. Finally, appropriate linker and adjuvant was used. PRABI server was used to validate second structure of protein. Third structure validation was done by SWISS-MODEL based on homology modeling and according to Main-Ramachandran-Plot. PortParam server determines the instability Index (II), physical and chemical properties of protein, MW and half-life. The latest validation is docking in order to check ability to protein binding to MHCI, II.

Results: Rapid identification of immune epitopes is based on computational predictions, which apply advance patterns and increasing epitope data base.

Conclusion: This approach processes many advantages over traditional methods, it diminishes time and cost in vaccine development; corrects the number of proteins to be studied, simplify the selection process can identify antigens present in small scales or expressed only at certain stages, which would prevent or hamper their purification; and permits for the research of non-cultivable or precarious microorganisms.

Keywords: Bioinformatic; Immunoinformatic; epitope vaccines

Survey of Inflammatory Parameters Extracted from Complete Blood Count in Cystic Fibrosis Patients

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Background and Aim: Cystic fibrosis (CF) known as an autosomal recessive genetic deficiency in childhood is a systemic disease following complicated chronic lung infections. This study aimed to investigate the inflammatory parameters extracted from Complete Blood Count (CBC) test in CF patients hospitalized from 2010 to 2020 in a teaching hospital, Sanandaj, Kurdistan, Iran.

Methods: A cross-sectional study was planned based on patient's documents in Sanandaj's hospitals from 2010 to 2020. 57 patients suspected of CF with a positive family history or symptoms of growth retardation, chronic cough, fatty stools, and meconium ileus were examined. 19 and 38 patients were with and without CF, respectively. The extracted CBC test parameters, including the NLR (Neutrophil-to-lymphocyte ratio), PLR (Platelets-to-lymphocyte ratio), and RPR parameters (RDW-to-platelets ratio), were measured and compared in two groups using an independent T-test or its corresponding nonparametric test, Mann-Whitney U test.

Results: The mean and standard deviation (SD) age of patients with and without CF were 9.8 ± 11.1 and 11.31 ± 9.46 , respectively. 14 (73.7%) of 19 CF patients were male. The mean of NLR (2.2 ± 1.4) and PLR (11.2 ± 4.93) in patients with CF was significantly higher than patients without CF. However, there was no significant difference between RPR in patients with and without CF ($P=0.2$). The odds ratio and its corresponding 95% confidence interval (CI) of the NLR and PLR indexes were 1.99 (1.12 - 3.54) and 1.22 (1.04 - 1.42), respectively. The RDW and WBC parameters were also statistically significant differences between the two groups.

Conclusion: The NLR and PLR parameters can be useful as a predictor for cystic fibrosis and patients' clinical course with the disease.

Keywords: Cystic fibrosis, Neutrophil-to-Lymphocyte Ratio; Platelets-to-Lymphocyte Ratio; RDW-to-Platelets Ratio; Inflammatory Biomarkers; Inflammatory Activity

Systemic Allergic Contact Dermatitis to Heavy Metals (Chromium, Lead and Copper), Mechanism, Clinical Manifestations, and Prevalence

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Background and Aim: Contact dermatitis is an inflammatory reaction of the skin due to direct contact with chemical agents in the environment, skin changes arise after re-contact with this substance. The clinical condition of contact dermatitis is regarded as local skin rash, itching, redness, swelling, and lesions, which are mainly detected by the patch test. Today, 15 to 20% of people have some degree of severe contact dermatitis. Immune responses in allergic contact dermatitis (ACD) are due to the interaction of cytokines and allergen-specific CD4⁺ and CD8⁺ T cells. Common causes of contact dermatitis include acids, alkalis like drain cleaners, body fluids, plants such as poinsettias, hair dyes, nail polish remover, and heavy metals. Heavy metals refer to any metallic chemical element that has a relatively high density and is toxic at low concentrations.

Methods: We observed studies that evaluated the relationship between exposure to heavy metals (“Chromium”, “Lead”, “copper”) and allergies. The following databases were searched up to September 2021: PubMed, Google Scholar, Scopus, Ovid-Medline, and Web of Science, and no limits about period or type of work were utilized. Case reports, clinical trials, original research, and review articles were thus evaluated. Metal allergy may cause ACD and also systemic contact dermatitis (SCD). We can detect contact dermatitis through various laboratory tests such as patch testing, lymphocyte stimulating test (LST), or evaluation of cytokine production by primary cultures of peripheral blood mononuclear cells

Results: Chromium, lead, and copper, are among the most common heavy metals used in various industries. Analysis of data from patch testing, lymphocyte stimulation (LST) testing, or evaluation of cytokine production by primary cultures of peripheral blood mononuclear cells, in numerous studies, confirmed the occurrence of contact dermatitis due to exposure to these heavy metals.

Conclusion: This article presents an updated observation on the epidemiological and clinical characteristics of systemic contact dermatitis to three heavy metals

Keywords: Systemic contact dermatitis (SCD); Allergic contact dermatitis (ACD); heavy metals; clinical manifestations; prevalence

Targeted Knockdown of Tim3 by Short Hairpin RNAs Improves the Function of Anti-Mesothelin CAR T Cells

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Background and Aim: T-cell immunoglobulin mucin 3 (Tim3) is an immune checkpoint receptor that plays a central role in chimeric antigen receptor (CAR) T cell exhaustion within the tumor microenvironment. This study was aimed to evaluate the effects of targeted-knockdown of Tim3 on the antitumor function of anti-mesothelin (MSLN)-CAR T cells.

Methods: To knockdown Tim3 expression, three different shRNA sequences specific to different segments of the human Tim3 gene were designed and co-inserted with an anti-MSLN-CAR transgene into lentiviral vectors. To investigate the efficacy of Tim3 targeting in T cells, expression of Tim3 was assessed before and after antigen stimulation. Afterwards, cytotoxic effects, proliferative response and cytokine production of MSLN-CAR T cells and Tim3-targeted MSLN-CAR T cells were analyzed.

Results: Our results showed that activation of T cells and MSLN-CAR T cells led to up-regulation of Tim3. Tim3 knockdown significantly decreased its expression in different groups of MSLN-CAR T cells. Tim3 knockdown significantly improved cytotoxic function, cytokine production and proliferation capacity of MSLN-CAR T cells.

Conclusion: Our findings indicate that targeted knockdown of Tim3 allows tumor-infiltrating CAR T cells that would otherwise be inactivated to continue to expand and carry out effector functions, thereby altering the tumor microenvironment from immunosuppressive to immunosupportive via mitigated Tim3 signaling.

Keywords: Mesothelin; Chimeric antigen receptor; Tim3; Targeted knockdown; Tumor microenvironment

The Association Between covid-19 and Blood Groups: a Review

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Background and Aim: ABO blood groups are actually carbohydrate epitopes that exist on the surface of all human cells. ABO blood group antigens are presented not in red blood cells but in respiratory epithelial cells and only in endothelial cells below blood vessels that also synthesize ABH carbohydrate epitopes. In this review article, we study the association between COVID-19 and blood groups.

Methods: This article is a review of reputable databases including PubMed, Scopus, Cochrane Library and Web of Science with the keywords "ABO blood group", "covid-19" and "disease susceptibility". By removing duplicate titles from endnote software and reviewing titles and abstracts, articles related to the JBi tool were reviewed.

Results: It is hypothesized that protein S from virions produced by individuals A or B could be decorated with carbohydrate A or B epitopes, respectively. Natural anti-A or anti-B antibodies from people in blood groups O, B, and A can bind these epitopes to the S protein of viral particles and block their interaction with ACE2 protein receptors in the host cell membrane, resulting in Prevent infection. also Patrice et al. Found that anti-A antibodies specifically inhibited the adhesion of SARS-CoV S-expressing cells to ACE2-expressing cell lines. Due to the similarity of nucleic acid 8 sequence and the similarity of angiotensin 2 (ACE2) receptor binding receptor binding between SARS CoV and SARSCoV-2, lower sensitivity of blood group O and higher sensitivity of blood group A for COVID-19 can be considered despite anti Natural anti-blood group antibodies, especially anti-A antibodies, in the blood

Conclusion: According to findings and studies, there is a relationship between blood types and the risk of COVID-19. As a result, people with blood type O are at lower risk and people with blood type A are at higher risk.

Keywords: ABO blood group; covid-19; disease susceptibility

The Duration of Antibody Persistence in Recovered COVID-19 Patients

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Background and Aim: Despite extensive studies on the COVID-19 epidemic, there are inconsistencies in the duration of antibody stability and adequate immune response in patients. The aim of this study was to evaluate the durability of IgG antibodies in recovered COVID-19 patients.

Methods: In this descriptive-analytical study, the serum levels of IgG antibodies were measured using ELISA in 248 recovered patients. Chi-square, t-test, ANOVA, and regression analysis in SPSS-15 and Stata-14 were used to investigate the relationship between variables.

Results: The data showed that 81% of discharged patients with over three months having passed since their admission had positive IgG levels. Moreover, five recovered patients with over seven months having passed since their admission had positive IgG levels. The highest prevalence of IgG positivity was observed in recovered patients with four weeks to two months having passed since their admission.

Conclusion: These results indicate that the immune response develops in most COVID-19 patients and can last up to seven months. However, it is undoubtedly advisable to vaccinate the patients even recovered patients to prevent reinfection.

Keywords: Coronavirus; Seroprevalence; IgG level; Immune response; Humoral response.

The Effect of Interferon-Beta Therapy on T-Helper 17/miR-326 and T-Helper 1/miR-29b-3p Axis in Relapsing-Remitting Multiple Sclerosis Patients

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Background and Aim: We aimed to evaluate the therapeutic effects of interferon-beta (IFN- β) on hsa-miR29b-3p and hsa-miR326 in isolated T-helper (Th)1 and Th17 cells expressed by relapsing-remitting multiple sclerosis (RRMS) patients before and after 1 year of treatment with IFN- β .

Methods: The study was done on 19 RRMS patients pre- and posttreatment with IFN- β to evaluate the frequency of Th1 and Th17 cells by flow cytometry. The expression level of hsa-miR-29b-3p and hsa-miR-326 in isolated Th1 and Th17 cells was assessed by quantitative polymerase chain reaction. Enzyme-linked immunosorbent assay was also used to measure the plasma levels of I interferon - gamma and interleukin (IL)-17A.

Results: Th17 cells and plasma levels of IL-17A decreased in RRMS patients after IFN- β therapy but hsa-miR-29b-3p and hsa-miR-326 expression had no significant change in treated RRMS patients versus baseline. MxA gene expression was significantly induced upon IFN- β therapy in patients with RRMS.

Conclusion: IFN- β therapy is more effective on Th17 than Th1, but it does not reform altered expression of hsa-miR-326 and hsa-miR-29b-3p in Th17 and Th1, respectively.

Keywords: Hsa-miR-29b-3p; Hsa-miR-326; Interferon-beta; MxA

The Effect of Micronutrients and Immune System on Obese Covid-19 Patients

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Background and Aim: Coronavirus 2 (SARS-CoV-2) is a violent virus that causes acute respiratory syndrome pandemic (COVID-19). ARDS is the consequence of covid-19 which is stimulated by the cytokine storm. The pattern of cytokine storm in obesity people is usually similar. This pattern (IL-1 β , IL-6, TNF- α , and IL-17) can increase the severity of this disease. Therefore, BMI is an important factor for evaluation of the severity. People with BMI > 30 kg/m² are almost twice in the disease intensity risk. (OR 1.8; 95% CI: 1.20–2.7, p = 0.006). Obese people who are on extreme diets for losing weights are usually deficient in numerous micronutrients such as: vitamins A, B, C, D, and E, Selenium, Copper, Zinc, and Iron. These elements have an essential role in function of immune cells and anti-SARS-CoV-2 defense. The micronutrients deficiency is another stimulant in developing ARDS and increasing mortality rate.

Methods: Obesity is related to high risk of COVID-19 infection and increased severity. Clinical studies on the effect of obesity on virus invasion and inflammatory response to the infection. Systematic researches have shown the obesity and covid-19 correlation and claimed that obese individuals are at a higher risk of covid-19 infection. COVID-19 positive patients with high BMIs manifest more severe and critical symptoms. The obesity prevalence is constantly increasing among populations and malnutrition is considered as an important key stimulator for obesity. A recent study predicted that more than 51% of the world population would involve in obesity by 2030. Moreover, Popkin et al. has reported that there is an association between obese patients and covid-19 by discussing 35 studies. Although the exact mechanism of disease progression is still unknown, inflammation appears to play an important role in exacerbating the condition. The data from these 35 studies showed that individuals with obesity have 48% higher death rate (OR=1.48; 95% CI, 1.22-1.80; p< 0.001) (123). Genetic researches have suggested that some genes associated with the comorbidity disease such as hypertension, (COPD) chronic obstructive pulmonary disease, (CVD) cardiovascular disease, (CLD) chronic liver disease and (CKD) chronic kidney disease were also directly associated.

Results: In this review, we discussed the association of pandemic COVID-19 and immunologic inflammation. During this pandemic, people with obesity are at a higher risk due to deleterious effect of cytokine storm. The excessive and prolonged cytokine response, which is triggered by obesity, is associated to COVID-19. In certain conditions increased migration of immune cells leads to



inflammation and causes ARDS and death. Some interventions and recommendations may help to overcome the RNA viral infection.

Conclusion: Hence, healthy lifestyle, regular exercise, a healthy diet and consumption of micronutrients such as vitamins (B, C, D, E, Iron, Copper, Zinc and Selenium) are essential for the boosting of immunity. It has been suggested that patients with underlying diseases, such as obesity, should undergo an optimal nutritional program before initiating any general treatment. Moreover, anti-inflammatory treatments that target cytokines and block them are part of the optimal intervention during the COVID-19 pandemic which successfully reduce the inflammation and improve the treatment.

Keywords: Obesity; inflammation; micronutrients; Covid-19



The Effects of Extremely Low Frequency Magnetic Fields (ELF-MFs) on the Pro/Anti-Inflammatory Cytokines of Innate and Acquired Immunity: A Review

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Background and Aim: Extremely low frequency magnetic fields (ELF-MFs) which are abundantly used in the modern life have raised some concerns about its possible adverse effects on our health. Several reports have been published on the different effects of ELF-MFs on the immune system. This review focuses on the effects of ELF-MFs on pro/anti-inflammatory cytokines functions of the immune system.

Results: It seems that physical (e.g. frequency, field direction, density, exposure length, etc.) variables of ELF-MFs and biological (e.g. cell type, species, etc.) factors are able to cooperate together to make positive/negative effects on the cytokine functions. According to the results of some in vivo and in vitro experiments, ELF-MFs exposure increased pro-inflammatory cytokines in the innate immunity. In contrast, a series of in vivo studies revealed a reduction of the pro-inflammatory functions of adaptive immunity cytokines due to ELF-MFs exposure. Furthermore, mainly of in vivo and in vitro reports have indicated that ELF-MFs reduce anti-inflammatory cytokines.

Conclusion: Therefore, it can be concluded that at the beginning of the immune system response (by innate immunity), ELF-MFs stimulate inflammation process. However, at the end stage of the immune system reaction (by adaptive immunity), the inflammation will be attenuated by ELF-MFs.

Keywords: Innate immunity; adaptive immunity; pro/anti-inflammatory cytokine; Extremely low frequency magnetic fields

The Evaluation of Three Iranian ELISA Kits for Detecting Anti-SARS-COV-2 Antibodies

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Background and Aim: Following the outbreak of the Covid-19, different methods were introduced for diagnosing Covid-19 infected patients and evaluating of immune response after natural infection or vaccines. This study aimed to compare efficacy of three domestic ELISA kits in measuring the level of COVID-19 specific IgG in a group of patients who recovered from the COVID-19.

Methods: This was a cross-sectional study conducted on serum of patients who recovered from an RT-PCR-confirmed COVID-19, in Birjand city, South Khorasan Iran. The level of COVID-19 anti N and S protein IgG was measured by using commercial ELISA kits. Comparison between groups was done using the Mann–Whitney U test.

Results: The mean titer of anti-N IgG was significantly higher for the PishtazTeb kit than IdealTashkhis (P less than 0.0001). There was no correlation between the titer of anti-N IgG (PishtazTeb and IdealTashkhis) and anti-S IgG (Chemibind) antibodies.

Conclusion: The results of this study indicate that the domestic ELISA kits have variable but acceptable sensitivity for detection of COVID-19 specific IgG antibodies.

Keywords: COVID-19; SARS-CoV2; ELISA; Kit; serologic assays

The Role of Autoantibodies against N-methyl-D-Aspartate Receptor in Schizophrenia

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Background and Aim: Schizophrenia (SCZ) is a debilitating mental disorder. The immune system is now known to play vital roles in nervous system function through regulating neuronal and synaptic plasticity. Dysregulation of the immune system and the possibility of autoantibody production against ion channels in the central nervous system, including N-Methyl-D-aspartate receptor (NMDAR) can lead to the development of SCZ. The aim of this study was to investigate the role of autoantibodies against NMDAR in patients with SCZ.

Methods: A total of 40 patients with SCZ and 40 healthy subjects were enrolled in this study. Antibodies against NMDAR in patients with SCZ were measured using the indirect immunofluorescence assay and analyzed in the Image program. The chi-square and Fisher's exact tests were used to compare the frequency of autoantibodies between patients with SCZ and healthy subjects.

Results: Among 40 SCZ patients, 6 (15%) patients showed serum IgG antibodies against NMDAR, and a significant increase in serum anti-NMDAR IgG levels was found in patients with schizophrenia ($P = 0.026$).

Conclusion: Autoantibodies against NMDAR exist in patients with SCZ. Dysregulation of the immune system and the production of autoantibodies against NMDAR can impair the function of the nervous system and affect the development of SCZ. Therefore, autoantibody testing is recommended in people with a history of mental illness as a screening and early diagnosis.

Keywords: Schizophrenia; Immune system; Autoantibodies; NMDAR; Pathophysiology

The Role of Cytokine Profile and Lymphocyte Subsets in the Severity of Coronavirus Disease 2019 (COVID-19): A systematic Review and Meta-Analysis

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Background and Aim: This study aimed to make a comparison between the clinical laboratory-related factors, complete blood count (CBC) indices, cytokines, and lymphocyte subsets in order to distinguish severe coronavirus disease 2019 (COVID-19) cases from non-severe ones.

Methods: Relevant studies were searched in PubMed, Embase, Scopus, and Web of Science databases until March 31, 2020. Cochrane's Q test and the I² statistic were used to determine heterogeneity. We used the random-effect models to pool the weighted mean differences (WMDs) and 95% confidence intervals (CIs).

Results: Out of a total of 8557 initial records, 44 articles (50 studies) with 7865 patients (ranging from 13 to 1582), were included. Our meta-analyses with random-effect models showed a significant decrease in lymphocytes, monocyte, CD4⁺ T cells, CD8⁺ T cells, CD3 cells, CD19 cells, and natural killer (NK) cells and an increase in the white blood cell (WBC), neutrophils, neutrophil to lymphocyte ratio (NLR), C-reactive protein (CRP)/hs-CRP, erythrocyte sedimentation rate (ESR), ferritin, procalcitonin (PCT), and serum amyloid A (SAA), interleukin-2 (IL-2), IL-2R, IL-4, IL-6, IL-8, IL-10, tumor necrosis factor-alpha (TNF- α), and interferon-gamma (INF- γ) in the severe group compared to the non-severe group. However, no significant differences were found in IL-1 β , IL-17, and CD4/CD8 T cell ratio between the two groups.

Conclusion: Decrease in total lymphocytes and lymphocyte subsets as well as the elevation of CRP, ESR, SAA, PCT, ferritin, and cytokines, but not IL-1 β and IL-17, were closely associated with COVID-19 severity, implying reliable indicators of severe COVID-19.

Keywords: COVID-19; SARS-CoV-2; Cytokine; Lymphocyte subsets; Meta-analysis.

The Role of Heat Shock Proteins in Viral Infections and COVID-19; A systematic review

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Background and Aim: The Heat Shock Proteins are important factors in regulating cell survival, differentiation, and cell death. The HSPs expression is rapidly induced in the face of physiological or environmental attacks. Some of them participate in innate and adaptive immunity. They are crucial factors in many signal transduction pathways such as inflammation, so the control of HSP expression would be a powerful tool for the invading virus to manipulate the fate of the host cell. Currently, humans affected by COVID-19 may suffer from dysregulated immune responses, resulting in excessive inflammation, known as cytokine storm. Studies show there are interactions between HSP and viruses and they can be involved in viral infections.

Methods: This systematic review was conducted to outline comprehensive studies published in PubMed, Scopus, and Google scholar databases from 2020 to 2021 by using 4 keywords 32 articles were screened, and 11 were totally included.

Results: We comprehend that HSPs may be associated with the outcome of infection with SARS-Cov2 and can attract great interest as potential antiviral targets. The researchers stated that HSP60 has enhanced inflammatory response with proinflammatory cytokine induction and pharmacological inhibition of HSP60 could potentially ameliorate inappropriate inflammatory reaction in severe COVID-19 patients. Recent studies have also shown that Hsp90 is involved in the replication of various human coronaviruses and that inhibition significantly reduces the propagation of SARS-CoV and SARS-CoV-2. Another HSP called GRP78 may stabilize the interaction between the viral spike protein and the cellular host receptor to facilitate entry or serve as an alternative host factor for viral entry.

Conclusion: Understanding the innate immune system against SARS-CoV-2 and the role of HSPs in COVID-19 infection and resolution of inflammatory pathways may provide information to the low SARS-CoV-2 mortality rates.

Keywords: Heat Shock Protein; COVID-19; innate immunity; cytokine storm

The Role of Immunotherapy by Using NK Cell Therapy and Interleukin Therapy in COVID-19

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Background and Aim: Vaccination suitable solution for controlling the coronavirus disease (COVID-19) pandemic, but still there are some challenges in COVID-19 vaccination. Also, another medicinal option is Immunotherapy. Restoration of NK cell effector processes has the potential to correct the temperate immune balance required to effectively overcome SARS-CoV-2 infection. transfer of NK or CAR-NK cells with useful cytolytic potentials and the lowest capacity of cytokine-release to severe COVID-19 patients may provide an efficacious treatment mainly in cases suffering from cytokine storms.

Methods: Interleukins (ILs) 2, 12, 15, and 18 released by targets infected with the virus or active antigen-presenting cells can modulate NK cell function while increasing survival, proliferation, cytotoxicity, and cytokine production. IL-12 increases the production and secretion of IFN- γ by NK cells while IL-15 induces NK proliferation. Interleukin treatment can improve the effective function of NK cells. Given the importance of IL-15 signaling and NK cell function, researchers have created an "alternative" to IL-15, which is IL-15: IL-15R heterodimers that have better stability and bioactivity than IL-15 monomers. They have inside the body. Interleukin-6 (IL-6) is also a prognostic marker for COVID-19 infection, and tocilizumab blocks its receptors, the drug of choice for COVID-19 infection.

Results: the supplementation of IL-2 and IL-15 supplies more NK cell maturing (CD56dim phenotype) with an applicable cytolytic effect. with the technologies available to create chimeric antigen receptor (CAR)-NK cells, it is nowadays too possible to design specific cell lines expressing any receptor. However, because these interleukins also act as pro-inflammatory cytokines, their therapeutic use should be associated with severe alert to control the worsening of COVID-19 patient's inflammatory conditions because of expanded levels of IL-15 also chronic pulmonary inflammatory diseases, and Middle East respiratory syndrome coronavirus (MERS-CoV) infection.

Conclusion: Immunotherapy has a remarkable effect on reducing mortality and morbidity of patients with COVID-19. Given the significance of maintaining the innate immune system, particularly the function of NK cells to halt cytokine storms, be effective in preventing either acute diseases of COVID-19 or in the cure of severe conditions of this disease. appears that if the cytokine therapy is successful, their use in the cure of COVID-19 patients can be beneficial than NK cell therapies.

Keywords: Immunotherapy; NK cell therapy; Interleukin therapy; COVID-19; Vaccine

The Role of MicroRNA-30c in Targeting Interleukin 6, as an Inflammatory Cytokine, in the Mesenchymal Stem Cell: A Therapeutic Approach in Colorectal Cancer

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Background and Aim: Colorectal cancer (CRC) is the third most prevalent cancer and the second significant cause of cancer-associated death worldwide. The microRNA-30 is a substantial member of the miRNA family and plays a vital role in the expansion of several cancers. This microRNA potentially targets interleukin-6 as an inflammatory cytokine in CRC.

Methods: MSCs were isolated from mice bone marrow and identified via the related markers. Then transduced with lentiviruses containing miR-30C. Transfected MSCs were collected to evaluate IL-6 levels, CT-26 cells were also co-cultured with MSCs, and the effect of apoptosis and IL-6 on the supernatant was assessed.

Results: Our result showed the expression of IL-6 and the level of protein were decreased in the supernatant of miR-30- transduced MSC cells compared to the control group. The rate of apoptosis was assessed, and the obtained data revealed the induction of apoptosis in CT-26 cells when they are in the vicinity of miR-30c transduced-MSCs.

Conclusion: We demonstrated that downregulation of miR-30c was significantly correlated with CRC progression and survival. So, the present study elucidated the anticancer effects of miR-30c in CRC and presented a novel target for CRC therapy.

Keywords: Mesenchymal Stem Cells; Interleukin-6; microRNA-30

The Serum Levels of IL-35 in Patients with Schizophrenia

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Background and Aim: Schizophrenia (SCZ) is a chronic and severe mental disorder. One of the approaches in schizophrenia is the role of the immune system and inflammation. IL-35 is a regulatory and anti-inflammatory cytokine, which plays an important role in the prevention of autoimmune diseases and inflammatory diseases. The aim of this study was to evaluate serum IL-35 levels in subjects with SCZ.

Methods: Forty patients diagnosed with SCZ and 40 healthy subjects were enrolled in the research. The serum levels of IL-35 were measured using the enzyme-linked immunosorbent assay (ELISA). An independent-samples t-test was used to compare the IL-35 concentration between patients with SCZ and healthy subjects.

Results: Serum levels of IL-35 were significantly decreased in patients with SCZ compared to the control group ($P=0.001$).

Conclusion: Based on the present study's results, serum levels of IL-35 were decreased in patients with SCZ. IL-35 through inhibition Th1 and Th17-dependent immune responses and also supporting regulatory T lymphocytes play an important role in inhibiting immune system and inflammation. We think IL-35 can be a therapeutic target for reducing inflammation and the severity of symptoms in patients with SCZ.

Keywords: Schizophrenia; Inflammation; Interleukin 35; Mental disorders; Immune system

The Use of Vitamin D, a New Way to Relieve and Improve Asthma

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Background and Aim: Asthma and allergies are one of the most common diseases and affect about 300 million people worldwide and can reduce the quality of life. The purpose of this study is to review the effect of supplements, especially vitamin D supplements in improving and relieving asthma.

Methods: The present study is a review. A comprehensive search of PubMed, Embase, Scopus, UpToDate electronic resource databases with the keywords Asthma, Vitamin D, Allergy, and Supplements was performed from 2015 to 2021. 42 Related Articles Extracted and Analysed.

Results: The results of studies indicate that the treatment of asthma with specific methods and standard treatment is vital. Also, taking vitamin D with other supplements such as folic acid, selenium, zinc, and copper can be effective so that it causes better lung function, Control asthma, and increase the quality of life of the patient.

Conclusion: Taking supplements, especially vitamin D supplements along with standard and common asthma treatments, can be effective in improving the clinical symptoms and quality of life of patients with asthma.

Keywords: Asthma; Vitamin D; Allergies; Supplements

Transmission of Inflammatory MicroRNAs by Exosomes in Patients with COVID- 19

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Background and Aim: COVID- 19 disease has already killed thousands of people around the world. This disease can cause severe and widespread inflammation and can kill the patient. Measurement of inflammatory parameters and inflammatory microRNAs that are transmitted by nanoscale vesicles or exosomes to monitor and prognosis the disease and also Understanding and investigating the pathogenic mechanisms of COVID- 19 is of great importance.

Methods: In this study, blood samples were taken from 25 patients with COVID- 19 and 5 healthy someones, then the exosomes were isolated from the serum of the participants in the study. miRNA-21 expression and miRNA-155 expression were examined by real-time PCR test.

Results: The results were as follows, the relative expression of miRNA-21 and miRNA-155 in serum-derived exosomes of patients with COVID-19 had a significant increase (p less than 0.0001) compared to the control group.

Conclusion: It can be concluded that increasing the expression of these inflammatory microRNAs enclosed in exosomes is one of the ways to increase inflammation in the patient's body and transfer the inflammation to different parts of the patient's body. This could help in the design of new anti-inflammatory drugs against COVID- 19 disease.

Keywords: COVID-19; miRNA-21; miRNA-155; anti-inflammatory drugs

Use NK-Cell Instead of T-Cell in CAR-NK Therapy-Review

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Background and Aim: Natural killer cells (NK cells) and cytotoxic CD8 + T cells are two types of immune cells that can kill target cells through similar cytotoxic mechanisms. With the remarkable success of CAR-T-engineered chimeric antigen receptors (CARs) for the treatment of blood malignancies, there is a rapid interest in the growth of CAR-NK cells for the treatment of growing cancer. The study reviewed recent advances in the clinical application of CAR-NK cells and the current and future challenges of CAR-NK cells as new cell immunotherapy in cancer.

Methods: The present review study was performed by analyzing articles in databases such as Science Direct, PubMed, and google scholar with the keywords "CAR-T" and "CAR-NK" in the last 10 years. According to the study criteria, 5 articles were included in the study.

Results: Compared to CAR-T cells, CAR-NK cells have significant benefits, including better immunity, such as the absence of minimal cytokine release syndrome and neurotoxicity in the autologous environment, and host versus host disease in allogeneic conditions, Have high production potential and are ready for consumption. CAR-NK cells can be engineered to target a variety of antigens, increase proliferation and stability in vivo, increase penetration of solid tumors, overcome tumor-resistant micro resistance, and ultimately achieve an effective antitumor response.

Conclusion: According to the findings of the study, since this method is a new method of treatment, a decision, in this case, is not yet possible and further studies are recommended.

Keywords: NK-Cell; T-Cell; CAR-NK

Valproic Acid Inhibits Cell Proliferation and PD-L1-Mediated Tumor Immune Escape Through Targeting CIP2A and c-MYC/PI3K/Akt/mTOR Signalling Molecules in Breast Cancer

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Background and Aim: Resistant cells are a critical problem that reduce treatment efficacy of breast cancer. Nowadays, CIP2A and PD-L1 are considered as therapeutically challenges in breast cancer, because of responsible for drug resistance and immune evasion respectively. Hence, identifying agents to suppress these factors is great of interest. Specifically, epigenetic drugs can be an effective approach to alter the behavior of genes. Although valproic acid (VPA) as a HDAC inhibitor has certain anticancer properties but molecular mechanism of VPA in breast cancer cells remains to be explored. In this study, we investigated drug effects and molecular mechanisms of VPA, particularly its effect on CIP2A and PD-L1 in breast cancer MCF-7 cell line.

Methods: In this study, MCF-7 cells were treated with various concentration of VPA for 24 h, 48 h, and 72 h. The rate of cell viability was measured by MTT assay. Finally, gene expressions of CIP2A, c-MYC, PI3K, Akt, mTOR and PD-L1 were analyzed by real time PCR and Δ CT method.

Results: VPA showed a growth inhibitory effect in a dose and time-dependent manner in MCF-7 cell line. This effect is achieved by decreasing the expression levels of CIP2A oncogene and its downstream signaling molecules i.e. c-MYC, PI3K, Akt and mTOR. Furthermore, VPA decreased the expression levels of PD-L1 in relation to CIP2A inhibition.

Conclusion: These results indicated that VPA exerts its growth inhibitory effect through targeting the CIP2A and its downstream signaling molecules. In addition to being a CIP2A targeting agent, VPA also inhibits PD-L1 through CIP2A inactivation in MCF-7 cell line. Our findings suggest that VPA can be a novel approach to combat with challenges caused by CIP2A and PD-L1, thereby alone or in combination with existing therapies could be promising strategy to get more efficiencies in treatments for breast cancer patients.

Keywords: Valproic acid (VPA); histone deacetylase inhibitor (HDACi); CIP2A, PD-L1; c-MYC; PI3K; Akt; mTOR; breast cancer

Wharton's jelly of the Human Umbilical Cord-Derived Mesenchymal Stem Cell Exosomes Alleviate Liver Fibrosis in Mouse Model of Liver Fibrosis

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Background and Aim: liver fibrosis is a wound healing response characterized by accumulation of extracellular matrix(ECM) following liver injury. Inflammatory factors such as cytokines and chemokines lead to the migration of immune cells, including monocytes and macrophages, to the site of injury. Macrophages play an important role in chronic liver injury. Some subsets are pro-fibrotic while others are anti-fibrotic. Mesenchymal stem cells such as human umbilical cord Wharton's jelly mesenchymal stem cells can be used for treatment of liver fibrosis. Exosomes have similar functions to their parent cells; However, they are safe and, as a cell free treatment, have controllable and predictable results. The aim of this study was to investigate the effect of WJ-MSCs on the improvement of hepatic fibrosis through the expression of Ly6Clow macrophages in liver and spleen tissues.

Methods: Mouse model of hepatic fibrosis was developed by injecting carbon tetrachloride(CCL4), then WJ-MSCs derived exosomes were injected into mice intraperitoneal. Then, using H&E and Masson's trichrome staining, biochemical analysis of liver enzymes, Real- time PCR and flowcytometry, the effects of exosomes on improving liver function were investigated.

Results: In contrast to phosphate-buffered saline(PBS) treated mice, CCL4-induced damage was reduced in exosome-treated mice, which included improved liver function, decreased collagen accumulation, decreased expression of inflammatory Ly6Chigh macrophagesic (pro-fibrotic), and increased restorative Ly6Clow macrophages(anti-fibrotic).

Conclusion: These results suggest that treatment with WJ-MSC-derived exosomes can improve CCL4-induced liver fibrosis by improving liver function and increasing anti-fibrotic Ly6Clow macrophages.

Keywords: Liver fibrosis; WJ-MSCs; Exosomes; Macrophages



Medical mycology



Prevalence of Fungal Infections of the Scalp and Hair in Patients Referred to Zahedan Medicine Mycology Laboratory

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Background and Aim: Tinea capitis is a prevalent childhood infection caused by a type of fungus called *Dermatophyte*. This infection includes three types of Ectothrix, Endothrix, and Favus. The purpose of this study was investigating the prevalence of tinea capitis in patients referred to the mycology laboratory of Zahedan Bu-Ali Hospital.

Methods: This descriptive cross-sectional study was conducted on 74 patients suspected for tinea capitis referring to the medicine mycology laboratory of Zahedan Bu-Ali Hospital during 11 months (November 2018 - September 2020). Samples were obtained by scraping the infected head area with a sterile scalpel. Microscope examination was considered positive in the presence of spores (both ectothrix and endothrix) and hyphae (favus).

Results: 93.15% of patients referred to the medical mycology laboratory (n = 69) had Tinea capitis; Ectotrix and Endotrix types. The prevalence of Tinea capitis was 64.8% in males and 28.35% in females patients, which was statistically significant ($P \leq 0.05$).

Conclusion: The findings of this study showed that the highest prevalence of Tinea capitis is higher in male patients in comparison female patients.

Keywords: Tinea capitis; Dermatophytosis; Ectothrix

A Review About CAM in India

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Background and Aim: Increase incidence of invasive mucormycosis in India is associated with high incidence of COVID-19, so that by June 21, 2021, more than 47,000 cases of mucormycosis related to Covid-19 (CAM) have been reported from India. According to reports, the most common site of infection was Rhino-Orbital / Rhino-Cerebral. A high percentage of these patients had diabetes mellitus, Covid-19 and were treated with corticosteroids. Since that India is known as the diabetes capital of the world, seems diabetes mellitus was the most common underlying risk factor for CAM. Co-occurrence of fungal, bacterial and viral infections has been reported in patients with COVID-19, and especially outbreak of the Delta strain has caused deaths, and unfortunately the situation has become more difficult with the advent of the new Omicron strain.

Results: The three factors of SARS-CoV-2, corticosteroid use and uncontrolled diabetes are the main underlying factors in the development of invasive mucormycosis in India. CAM in India compared with other parts of the world shows that mortality rates in India were lower than other of the world. It was also found that increased incidence and mortality from pulmonary or diffuse mucormycosis, with admission to the intensive care unit, non-vaccination against COVID-19, previous pneumonia, HbA1C greater than 8.0 (uncontrolled diabetes), The use of ventilators and adjuvant oxygen, the use of antiviral drugs, the use of corticosteroids and the age of patients (age range around 55 years) are related.

Conclusion: The incidence of mucormycosis in India due to diabetes, uncontrolled hypertension Socio-economic, genetic and health conditions of the community and the best way of prevention of CAM is regular hand washing, control of blood sugar and blood pressure, not taking drugs arbitrarily, using masks, not using leftovers, boosting the immune system, adequate rest and exercise will be very useful, especially in the current situation.

Keywords: Covid Associated Mucormycosis; Rhinoserebral; CAM in India

Candiduria in Patients Admitted to Beheshti Hospital in Hamadan, West of Iran

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Background and Aim: The presence of *Candida* species in urine (candiduria) is a common clinical finding that indicates colonization or urinary tract infection (UTIs). *Candida albicans* is the most frequently isolated fungal species, although today an increase in the prevalence of non-albicans *Candida* species-NACS has been reported.

Methods: In this retrospective study, which was performed from the beginning of April 1400 to the end of September 1400, all urine cultures of patients admitted to different wards of the hospital were evaluated for urinary tract infections.

Results: Out of 2391 urine cultures, 554 samples (23.17%) were positive for urinary tract infections and 66 samples (11.91%) were identified as candiduria. 22 species of *Candida albicans* (33.3%) and 44 species of non-albicans. (66.6%) were identified. Out of the total number of patients with candiduria, 25 were male and 41 were female.

Conclusion: The presence of 11.91% of urinary tract infections due to *Candida* species and the higher prevalence of non-albicans species and the 1.64-fold increase in this infection in women draw more attention to this opportunistic yeasts.

Keywords: *Candida albicans*; Candidiasis; Urinary tract infection; candiduria

A Systematic Review and Meta-Analysis Study on Antifungal Susceptibility of Clinical *Candida albicans* to Conventional Antifungal Drugs (Azoles, Polyenes, and Echinocandins) in Iran

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Background and Aim: Antifungal susceptibility patterns of *Candida* infectious can play an essential and decisive role in the treatment outcome. The present systematic review and meta-analysis aimed to investigate the drug susceptibility pattern of Iranian clinical *Candida albicans* isolates to conventional antifungal drugs (azoles, polyenes, and echinocandins).

Methods: Six electronic databases including “PubMed,” “Scopus,” “Web of Science,” “IranDoc,” “SID,” “Magiran” were searched from May 2000 to June 2021.

Results: The susceptibility of 6322 *C. albicans* strains from 19967 patients against 14 antifungal drugs were evaluated according to Clinical and Laboratory Standards Institute (CLSI) methods. The pooled prevalence of antifungal resistance was ranged from 0 to 26%. The lowest resistance levels among azoles were observed in luliconazole with a frequency of 0% and voriconazole of 3.94% (95% CI: 2.24 to 6.09%). An increase in trend of itraconazole (7-16.65%), voriconazole (1.4 to 6.52%), and amphotericin B resistance (7.5 to 9.11%) among *C. albicans* strains since 2006 in Iran, while a decreasing trend was observed in fluconazole resistance (23.42 to 14.42%).

Conclusion: In conclusion, due to the resistance of *C. albicans* species to common antifungal drugs, antifungal stewardship strategy combining therapeutic drug monitoring to reduce the emergence of resistant multi-drug *Candida* species.

Keywords: Antifungal resistance; antifungal susceptibility; *Candida albicans*; systematic review; CLSI

Analysis of Seven Loci to Identify Most Suitable Genes for MLST of Major Pathogenic Dermatophytes

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Background and Aim: Dermatophytes are a group of fungi categorized by several genera, namely *Trichophyton*, *Microsporum*, *Epidermophyton*, *Nannizzia*, *Paraphyton*, *Lophophyton*, and *Arthroderma* based on new taxonomy. Demographic changes in recent decades have led to the spread of dermatophyte infections. Considering that there is no database for multilocus sequence typing (MLST) dermatophytes, this study aimed at introducing suitable genes for MLST of major pathogenic dermatophytes was developed.

Methods: We performed MrBayes, RAxML analyses based on the MLST scheme (loci: BT2, TEF-1 α , ACT, CaM, HSP70, D1/D2, and ITS), were performed to the evolution of 99 dermatophytes isolates including four species *Trichophyton interdigitale*/ *T. mentagrophytes* complex, *T. tonsurans*, *T. rubrum*, and *E. floccosum*.

Results: The findings of the present study indicated that three genes: BT2, ITS, and TEF-1 α , which showed the greatest diversity among dermatophyte species, were suitable for MLST. The HSP70 sequences did not show any intraspecies variation in all species. Also, the individual CaM tree failed to separate the clade of *T. mentagrophytes*/*T. interdigitale* complex from the clade of *T. tonsurans*. The phylogenetic tree, constructed by the combination of the three genes, shows a new topological pattern that confirms the derivation of the anthropophilic and zoophilic genera from the geophilic genus.

Conclusion: Results of this study showed that MLST is very effective in determining the boundaries between species and taxonomy. This study was a new starting point to the foundation for a dermatophyte MLST database. It is suggested that other genes with high polymorphism be examined to select the genes used for MLST.

Keywords: Dermatophytes; dermatophytosis; multilocus sequence typing

Antifungal Activity of Curcumin Nanoparticle Against Clinical Isolates of *Aspergillus*

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Background and Aim: *Aspergillus* is best known as a saprophyte fungus responsible for primary to the most devastating lung infections in normal to immunocompromised hosts. In terms of their ubiquitous characteristic, its spores easily transmit and contaminates the hospital ventilation systems. Due to drug resistance, the provided study covers the efficacy of Curcumin (CUR) nanoparticle on clinical isolates of *Aspergillus* as a fungicidal agent.

Methods: CUR nanoparticle was examined against a total 17 clinical isolates of *Aspergillus* including *A. fumigatus* (n= 3), *A. flavus* (n=8), *A. welwitschiae* (n=4), and *A. tubingensis* (n=1); and laboratory strain of *A. fumigatus* (TIMML 025) based on the M38-A2 guideline.

Results: At the concentration of 0.031-10000 µg/ml, CUR nanoparticle didn't show significant minimum inhibitory concentration (MIC); except *A. fumigatus* which showed the inhibitory range from 2500 to 10000 µg/ml.

Conclusion: Overall, the inhibitory effects of CUR nanoparticle was not noticeable against clinical isolates of *Aspergillus*. Due to the fact that it showed antifungal effect for just two, it is considerably not much among 17 isolates. There will be need to investigate more about CUR nanoparticle antifungal effects against other filamentous fungi.

Keywords: Antifungal; Nanoparticles; *Aspergillus*; MIC

Antifungal Activity of Curcumin Nanoparticle against Clinical Isolates of *Candida* Retrieved from Patients with Candidemia

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Background and Aim: The incidence of candidiasis is increasing due to the growing complexity of patients and acquire resistance to antifungal agents. Having said that, the research has oriented to focus on using nanoparticles and extracts of herbal plants as fungicidal agents. In this study, the efficacy of curcumin (CUR) nanoparticles has investigated on *Candida* species.

Methods: CUR nanoparticle was examined against a total 23 *Candida* isolates from patients with Candidemia including *C. albicans* (n=13), *C. dubliniensis* (n=2), *C. parapsilosis* (n=3), and *C. tropicalis* (n=1); and laboratory strains of *C. albicans* (TIMML 1292 and TIMML 183), *C. parapsilosis* (TIMML 2201), and *C. tropicalis* (TIMML 731) based on the M27-A3 guideline.

Results: It showed somehow significant minimum inhibitory concentration (MIC) in mostly the range of concentration from 625 to 10000 µg/ml. The MIC of CUR nanoparticle was 625-10000 µg/ml for *C. albicans*, 625-2500 µg/ml for *C. dubliniensis*, 10000 µg/ml for *C. parapsilosis*, 10000 µg/ml for *C. tropicalis*.

Conclusion: CUR nanoparticle showed the inhibitory affect in most strains of *Candida* in concentration of 10000 µg/ml. As a result, it wasn't that significant against *Candida* species and should be evaluated among other yeasts.

Keywords: Antifungal; Nanoparticle; *Candida*; MIC

Antifungal Susceptibility Patterns of *Aspergillus terreus* Complex Isolated from Clinical Specimens

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Background and Aim: *Aspergillus* species are responsible for the highest mortality rates among patients with fungal infections. *Aspergillus terreus* is the uncommon cause of invasive aspergillosis and pulmonary aspergillosis (PA) in immunocompromised patients. Millions of people worldwide are estimated to be at risk of aspergillosis each year, especially individuals with impaired immune function and those with low numbers of neutrophils, solid organ transplant recipients. Identifying these fungi as infectious agent is crucial because of the resistance to amphotericin B and increased lethality. Therefore, in this study, was determined the antifungal susceptibility profile of the *Aspergillus terreus* species.

Methods: In vitro antifungal susceptibility to amphotericin B, itraconazole, voriconazole, caspofungin, and posaconazole test was performed according to the CLSI M38-A2 guidelines. Thirty-four *Aspergillus terreus* species were identified based on morphological and molecular (using the sequencing beta-tubulin gene) methods.

Results: *Aspergillus terreus* complex included *A. terreus* and *A. citrinoterreus*. MIC/MEC range for caspofungin was 0.007–0.5 µg/ml, compared to 0.03–16 µg/ml for itraconazole, 0.06–1 µg/ml for voriconazole, 0.01–0.6 µg/ml for posaconazole, and 0.5–16 µg/ml for amphotericin B. *Aspergillus terreus* was the most susceptible to caspofungin (MEC₅₀ = 0.007 µg/ml) and the most resistant to amphotericin B (MIC₅₀ = 8 µg/ml). Among all the isolates, only 12.75% were susceptible to AMB. Caspofungin exhibited the lowest G mean (0.01 µg/ml), followed by posaconazole (0.14 µg/ml), voriconazole (0.31 µg/ml), itraconazole (0.57 µg/ml), and amphotericin B (7.8 µg/ml).

Conclusion: This information is necessary to improve patient management and when an outbreak dealing with drug-resistant infections occurs.

Keywords: *Aspergillus terreus*; *Aspergillus citrinoterreus*; antifungal susceptibility test; molecular identification

Comparison of in Vitro Antifungal Activity of Royal jelly, Propolis and Aloe vera with Available Antifungal Agents against Common Species of Dermatophytes

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Background and Aim: Dermatophytes are keratinophilic fungi cause mostly superficial diseases of the hair, nails and stratum corneum on humans and other mammals. We investigated the minimum inhibitory concentrations (MIC) and *mdr2* gene expression between natural compounds such as royal jelly, propolis and aloe vera with terbinafine and itraconazole on resistant and susceptible common species of dermatophytes isolated from clinical specimens

Methods: A total of 52 dermatophytes species were isolated from the patients. Identification of species was done by morphological methods and internal transcribed spacer (ITS1/4 rDNA). In vitro antifungal susceptibility testing with drugs (terbinafine and itraconazole) and natural compounds (royal jelly, propolis and aloe vera) was done in accordance with the Clinical and Laboratory Standards Institute M38-A2 method. Quantitative changes in the expression of the *mdr2* gene were calculated using real-time PCR.

Results: Forty-two percent (22/52) dermatophyte isolates were resistant to both terbinafine and itraconazole. Propolis with MIC range 25-800 µg/ml demonstrated most antifungal activity than royal jelly (50-3200 µg/ml) and aloe vera (400-6400 µg/ml) on susceptible and resistant species dermatophyte. In addition, the level of *mdr2* gene expression was significantly decreased after exposure with royal jelly and propolis extracts.

Conclusion: According to the data obtained from this study, it can be concluded that propolis extract and then royal jelly and aloe vera in general can be inhibitory effects on the growth of dermatophyte species sensitive and resistant to common antifungal drugs and *mdr2* gene expression.

Keywords: Dermatormycoses; Arthrodermataceae; royal jelly; propolis; aloe vera; terbinafine; itraconazole

Covid 19 and Fungal Infections

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Background and Aim: As the human-to-human transmitted disease, coronavirus disease (COVID-19), caused by severe acute respiratory syndrome coronavirus (SARS-CoV-2), has been an emergency global public health events. it is important to notice that COVID-19 patients can develop further fungal infections during the middle and latter stages of this disease, especially severely ill ones. after we search on a different article we found COVID-19 patients, especially severely ill ones or accompanied with immunocompromised state, had co-infections of fungi. in France Lanjuan Li academician and her colleagues reminded clinicians should focus on patients' fungal infections, especially severely ill or immunocompromised ones. *Aspergillus* species could be an important cause of life-threatening infection in COVID-19 patients, especially in those with high risk factors. The potential risk factors for the patients include GC use, prolonged neutropenia, chronic obstructive pulmonary disease (COPD), allogeneic hematopoietic stem cell transplant. For the severe COVID-19 patients who have more opportunities to be treat with broad-spectrum antibacterial drugs, parenteral nutrition and invasive examinations, or the patients accompanied with prolonged neutropenia and other immune impairment factors, the risk of infection with *Candida* species may significantly increase. COVID-19 patients with trauma, diabetes mellitus, GC use, HM, prolonged neutropenia, allo-HSCT, SOT are more likely to develop mucormycosis.

Methods: To design a systematic search strategy to investigate fungal infection in COVID-19 positive patients; PubMed, Google Scholar, and other online web sources were searched from the last 3 months and analyzed using terminologies such as "COVID-19," "SARS-CoV-2," "Fungal infection," "*Mucormycete*," "Black Fungus," "mucormycosis," "Immune signaling," "Immunotherapy," "Diagnosis," and "Treatment." at the end try to use important data of articles that we found.

Results: By analyzing retrospective analysis of SARS and influenza data from China and worldwide, we surmise that the fungal co-infections associated with global COVID-19 might be missed or misdiagnosed. Further, as a life-threatening infectious disease, COVID-19 patients showed overexpression of inflammatory cytokines, and impaired cell-mediated immune response with decreased CD4 + T and CD8 + T cell counts, indicating its susceptibility to fungal co-infection. Moreover, COVID-19 patients accompanied with immunocompromised state, such as prolonged neutropenia, HSCT, GC use, SOT, inherited or acquired immunodeficiencies, and tumor are more likely to develop fungal co-infection. Here, we summarized updated diagnostic information (histopathology, direct microscopic examination, culture, (1,3)- β -d-glucan, galactomannan, PCR-based assays, MALDI-TOF technology, etc.) and treatment recommendations of invasive mycosis.



Conclusion: According to the study of recent articles, at the end we come to the point that in patients who are in severe state of the covid 19 disease due to a weakened immune system and in cases that have hyperglycemia or people who has problems like these, they are prone to fungal infections. physicians need to consider this in severe state of covid 19 patients, and this is a very important point.

Keywords: fungal infections; covid19 and mycosis; Fungal co-infection; SARS-CoV-2 fungal infections; Invasive fungal infections



Effects of Dielectric Barrier Discharge Plasma on the Clinical Nystatin Resistant *Candida glabrata* Strains-Associated Denture stomatitis

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Background and Aim: Denture Stomatitis (DS) is an inflammatory lesion in which there is redness of the oral mucosa underneath a removable denture. That most cases of DS are *Candida* species related. The antifungal resistance has led to a reduce in the effectiveness of current therapies, prolonging remedy time and rising health care costs. The Dielectric Barrier Discharge (DBD) plasma used as a dental therapy. In this study, we examined the inhibitory activity of DBD plasma against resistant *Candida glabrata* strains isolated from patients with denture stomatitis.

Methods: DBD plasma was employed to treat three nystatin resistant *C. glabrata* strains and *C. glabrata* standard (ATCC 90030) for 1, 2, 4, 8 minutes. In order to assess the inhibitory activity of DBD plasma, we used CFU counting method. Besides, drug susceptibility was evaluated by using CLSI (M27.A3) guideline on strains treated with DBD plasma.

Results: By using CFU counting method, it was observed that the DBD plasma can be reduce colony count of all three resistant strains in 4 minutes. The resistant strains that treated with DBD plasma showed a lower Minimum Inhibitory Concentration (MIC) value to nystatin when compared with control groups.

Conclusion: DBD plasma alone or assisted with drug therapy can provide an appreciated tool for resistant Candidal infections, and it may be effective for protecting denture in stomatitis.

Keywords: *Candida glabrata*; Drug Resistance; Stomatitis; Denture; Plasma Gases

Epidemiology, Risk Factors, Species Distribution, and Antifungal Susceptibility of Candidemia among Hospitalized Patients with COVID-19

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Background and Aim: The pandemic of COVID-19 has caused a worldwide health crisis. Candidemia is a potentially lethal condition that has not yet been enough discussed in patients with COVID-19. The current study aimed to investigate the prevalence of candidemia among Iranian COVID-19 patients and characterize its causative agents and the antifungal susceptibility pattern.

Methods: The present cross-sectional survey was carried out from March 2020 to March 2021 at Imam Khomeini hospital, Tehran, Iran. Blood specimens were obtained from patients with confirmed coronavirus infection who also had criteria for candidemia and were examined for any *Candida* species by conventional and molecular techniques. Susceptibility of isolates to amphotericin B, voriconazole, itraconazole, fluconazole, caspofungin, and 5-flucytosine was tested using the CLSI broth dilution technique.

Results: one hundred fifty-three patients with COVID-19 were included. Candidemia was confirmed in 12 of the 153 (7.8 %) patients. The majority of patients were ≥ 50 years of age (n=9) and female (n=8). Six of the 12 patients were diabetic. The presence of central venous catheters, broad-spectrum antibiotic therapy, ICU admission, and mechanical ventilation were observed in all patients. *C. albicans* (n=7, 58.3 %) and *C. dubliniensis* (n=2, 16.7%) were the most common isolated species. Amphotericin B and 5-flucytosine were the most active drugs. Despite antifungal treatment, 4 out of 12 patients (33.3 %) died.

Conclusion: Due to the high mortality, the early diagnosis and proper treatment of candidemia are essential requirements for optimal clinical outcomes in COVID-19 patients.

Keywords: Candidemia; *C. albicans*; *C. dubliniensis*; COVID-19; Iran

Evaluation of Anti-*Candida* Effects of Moronecidin Peptide and Myrtus Extract against Drug-Resistant Isolates of *Candida albicans* and *glabrata*

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Background and Aim: In the last decades, *Candida* species have been causing severe cases of fungal infections. But the therapeutic effect of antifungal drugs has been limited by the emergence of drug-resistant strains. In the present study, the anti-*candida* effect of Moronecidin antimicrobial peptide (MAP) and *Myrtus communis* hydroalcoholic extract (MHE) on drug-resistant *Candida* isolates was investigated.

Methods: Five clinical isolates of *Candida glabrata* with GenBank Accession No: KT763087 (FLC, ITC, KTZ, VCZ resistant), KT763091 (ITC, KTZ, CASP resistant), KT763111 (FLC, ITC, KTZ, VCZ, AMB resistant), KT763123 (FLC, ITC, KTZ, AMB resistant), KT763084 (FLC, ITC, KTZ, VCZ, CASP, AMB sensitive) and 5 clinical isolates of *Candida albicans* approved by PCR-RFLP and duplex PCR (FLC resistant) were used. MAP was synthesized by the solid-phase synthesis by Pepmic (Suzhou, China). Subsequently, the standardized yeast suspension was inoculated into 96-well micro-titer plate containing MAP from 0.1 to 1.0 ug/uL and MHE from 0.23 to 0.00045 ug/uL. Positive and negative controls were placed in columns 11 and 12 of the microtiter plate for each isolate, respectively. Then plates incubated at 35° C for 48 h according to the reference microdilution method established by the CLSI (M27 4th ed).

Results: The MICs were defined as the lowest MAP or MHE dilution with no visible growth. MHE and AMP showed good overall activity against *C. albicans* and *C. glabrata* with MIC range, 0.9-1.8 ng/ul and 0.5-0.8 ug/ul for MHE and MAP, respectively.

Conclusion: High antifungal side effects and the increasing emergence of multidrug-resistant isolates are among the most important challenges in treating candidal infections, and the results of the present study showed that MAP and MHE can be good candidates for new antifungal compounds.

Keywords: Moronecidin; antimicrobial peptide; *Myrtus communis* extract; *Candida Spp.*; antifungal susceptibility

Evaluation of Antifungal Effects of Hydro-Alcoholic Extract of *Nigella Sativa* and *Urtica Dioica* on Fungal Agents Isolated from Clinical Specimens

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Background and Aim: Due to the indiscriminate use of chemical drugs to treat infectious diseases, multidrug resistance of human pathogenic microorganisms has developed and caused many problems that always threaten human health. The occurrence of fungal species resistant to antifungal drugs and also their numerous side-effects have persuaded researchers to work on new therapeutic methods. For this reason, today, medicinal plants have received more attention than ever in the treatment and pharmacological process; So far, many researchers have tried to find natural antifungal agents with fewer side effects. In this regard, the antifungal activity of some medicinal plants has been proven. Therefore, this study was designed to evaluate the antifungal effects of hydro-alcoholic extract of *Nigella sativa* and *Urtica dioica* on fungal agents isolated from clinical specimens.

Methods: In the current study, antifungal effects of hydro-alcoholic extract of *Nigella sativa* and *Urtica dioica* against 11 *Candida* isolates (*Candida albicans*, *Candida parapsilosis*), 13 *Aspergillus* isolates (*Aspergillus flavus*, *Aspergillus terreus*) and 5 Mucoral agents (*Mucor circinelloides*, *Rhizopus oryzae*) were examined using broth microdilution method. The tested fungal isolates were associated with different concentrations of the plant extracts, and then, the Minimum Inhibitory Concentration (MIC) of these extracts were determined separately along with amphotericin B.

Results: Both soaked and Soxhlet hydro-alcoholic extracts of *Nigella sativa* and *Urtica dioica* showed antifungal effects against *Candida*, *Aspergillus* and *Mucorales* isolates. The MIC₉₀ values of soaked and Soxhlet hydro-alcoholic extracts of *Nigella sativa* in *C. albicans* and *C. parapsilosis* were 25, 12.5 mg/ml respectively. For *Aspergillus flavus* in both methods were 25 mg/ml but, in *Aspergillus terreus* in soaking and Soxhlet methods were 12.5 and 25 mg/ml respectively and in mucoral species (*Mucor circinelloides* and *Rhizopus. oryzae*) were obtained 12.5 mg/ml. The MIC₉₀ values of soaked and Soxhlet hydro-alcoholic extracts of *Urtica dioica* in *Candida* and mucoral species in both methods were 12.5 mg/ml, but in the *Aspergillus* species were obtained 25 mg/ml.

Conclusion: The Hydroalcoholic extracts of *Nigella sativa* and *Urtica dioica* have acceptable antifungal activity on *Candida*, *Aspergillus*, *Mucor* and *Rhizopus* species. *Urtica dioica* extract has better efficacy against *Candida* species and *Mucorales*.

Keywords: *Nigella sativa*; *Urtica dioica*; *Candida*; *Aspergillus*; *Mucorales*

Evaluation of Biofilm Formation in *Candida parapsilosis* Isolated from Catheter in COVID-19 Patients

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Background and Aim: *Candida parapsilosis* is an important cause of fungal bloodstream infections and can form biofilms on catheter. Hence, biofilm formation may be an important clinical challenge for therapeutic choices. The objective of this study was to characterize biofilm formation by clinical isolates of *C. parapsilosis* isolated from catheter of COVID-19 patients.

Methods: Twenty isolates of *C. parapsilosis* isolated from indwelling catheter patients with COVID-19 in different units (neonatal, oncology, intensive care units). The biofilms were permitted to form in microtiter plate structure to mature (48 h) phases and quantified by MTT assays.

Results: Our finding showed that all the strains produced different level of biofilms but the biofilm formation ability of *C. parapsilosis* isolated from neonate catheters were significantly higher than other units (73%), followed by, intensive care units (55%%) and oncology 8 (42%) units.

Conclusion: The finding highlights the importance of *C. parapsilosis* biofilm formation as an important factor especially in neonates in association with indwelling catheters.

Keywords: MTT; *Candida parapsilosis*; Biofilms

Evaluation of Hematological Parameters in Patients with Coronavirus Disease 2019-Associated Mucormycosis(CAM) , Northeastern Iran

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Background and Aim: There are increasing reports of rhino-orbital mucormycosis in people with coronavirus disease 2019 (COVID-19). Significant laboratory findings such as lymphopenia and leukocytosis were highly reported in COVID-19 patients. Based on the important correlation between COVID-19 and fungal infections, this study provided the evaluation of hematologic parameters in patients with COVID-19-associated Mucormycosis in northeastern, Iran.

Methods: During an eight-month period, in two tertiary referral hospitals of Mashhad, hematological parameters such as White Blood Cell (WBC) count of 40 patients affected to COVID-19 with mucormycosis were precisely evaluated to check the results of leukopenia and leukocytosis.

Results: The patients with the range of 21 to 76 years old, median age of 58, assessed in this study. Of the 40 patients, 19 (47.5%) had diabetes mellitus, and 17 (42.5%) them died. The number of the patients with normal leukocyte count were 10. The 12, 3, and 1 patients showed the result of leukocytosis (neutrophilia), lymphopenia and leukopenia, respectively. However, the combination of leukocytosis and lymphopenia were significant in 7 patients. In between, the coincident presence of leukopenia and lymphopenia were investigated in just 5 patients. Above all, the results of leukopenia, lymphopenia, and neutropenia were coincided in 2 patients.

Conclusion: The hematological parameters of leukocytosis and lymphopenia were significant in evaluated patients. Clinicians should be aware that Mucormycosis may be a complication of COVID-19 in high-risk patients. Also, poor control of diabetes mellitus is an important predisposing factor for coronavirus disease 2019-associated mucormycosis (CAM). However, more studies should be done to conclude a more justifiable result.

Keywords: Mucormycosis; COVID-19; Infection; Mortality; Hematological parameters

Evaluation of Hematological Parameters in Patients with Coronavirus Disease 2019-Associated *Aspergillus rhinosinusitis*, Northeastern Iran

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Background and Aim: *Aspergillus rhinosinusitis* is a potentially lethal complication in patient with with underlying immunodeficiencies. *Aspergillus rhinosinusitis* can be a opportunistic fungal infection, caused by various *Aspergillus* species. Significant laboratory findings such as lymphopenia and leukocytosis were highly reported in COVID-19 patients. Based on the important correlation between COVID-19 and fungal infections, The aim of this study is to evaluate the hematologic parameters in patients with COVID-19-associated Mucormycosis in northeastern, Iran.

Methods: During an eight-month period, in two tertiary referral hospitals of Mashhad, hematological parameters such as White Blood cell (WBC) count of 14 patients affected to COVID-19 with *Aspergillus rhinosinusitis* were precisely evaluated to check the results of leukopenia and leukocytosis.

Results: The patients with the range of 42 to 67 years old, median age of 59, assessed in this study. Of the 14 patients, 8 (57.4%) cases had diabetes mellitus, and 9 (64.2%) cases of them died. We had 2 patients with normal leukocyte count. The 3, 2, and 1 patients showed the result of leukocytosis, lymphopenia and leukopenia, respectively. However, the combination of leukocytosis and lymphopenia were significant in 4 patients. In between, the coincident presence of leukopenia and lymphopenia was investigated in just 1 patient. Above all, the results of leukopenia, lymphopenia, and neutropenia was coincided in 1 patient.

Conclusion: *Aspergillus rhinosinusitis* had a high mortality among COVID-19 patients, high rate of diabetes mellitus is an important predisposing factor for coronavirus disease 2019-associated *Aspergillus rhinosinusitis*. Most of these patients received amphotericin B liposomal and caspofungin, but no significant difference was observed between recovery and death. The hematological parameters of leukocytosis and lymphopenia were significant in evaluated patients.

Keywords: *Aspergillus rhinosinusitis*; COVID-19; Infection; Mortality; Hematological parameters

Evaluation of the Apoptosis Effect of Titanium Dioxide Nanoparticles (TiO₂-NPs) on Different *Candida* spp.

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Background and Aim: Cell death divides into necrosis and apoptosis. Candidiasis is a fungal infection due to *Candida* species. The purpose of the study was to measure the apoptosis effect of titanium dioxide nanoparticles (TiO₂-NPs) on different *Candida* spp.

Methods: Apoptosis induced by MIC concentration of TiO₂-NPs for each *Candida* species was measured by the flowcytometric method.

Results: The flow cytometry results showed that TiO₂-NPs induced apoptotic effects on all *Candida* species. The highest apoptotic of TiO₂-NPs was seen on *C. albicans*. The lowest apoptosis effect of TiO₂-NPs was observed on *C. tropicalis*.

Conclusion: Our finding showed that TiO₂-NPs induced variable apoptotic effects on all *Candida* species.

Keywords: *Candida* spp; titanium dioxide nanoparticles; apoptosis

Evaluation of the Relationship Between Demographic Factors and the Incidence of Drug Resistance on *Candida albicans* Clinical Isolates

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Background and Aim: Vulvovaginal candidiasis (VVC) is the second most common infection of the female genital tract. *Candida albicans* (*C. albicans*) is the most frequently agent which identified from VVC. Following the widespread and long-term use of azole drugs, drug resistance and recurrence of the disease is increasing. Awareness of the risk factors affecting VVC and its association with azole resistance helps us in effective prevention and treatment. In this study attempted to determine the role of the risk factors in the development of resistance of isolates to azole drugs.

Methods: This cross-sectional study was performed on 60 *C. albicans* isolated from patients suspected to VVC. The demographic information participants recorded by questioner. Antifungal susceptibility was tested for 4 azoles (fluconazole, itraconazole, voriconazole, clotrimazole) via the microdilution method as per the testing standard M27-A3 of the Clinical and Laboratory Standards Institute. Then, logistic regression model was used to investigate the relationship between risk factors and resistance of each azole drug. The significance level was set at less than 0.05.

Results: The highest drug resistance was seen for fluconazole (64.4%) and clotrimazole (54.2%). A significant correlation was found between risk factor for use of contraceptive methods and resistance to fluconazole and voriconazole ($P=0.02$ and $P=0.04$ respectively).

Conclusion: Due to the high level of resistance to fluconazole and clotrimazole, followed by increased recurrence and treatment failure, we will need alternative drugs. The use of contraceptive methods, which is itself a risk factor for vulvovaginal candidiasis, was associated with the prevalence of resistance.

Keywords: Risk factor; *Candida vulvovaginitis*; Azoles; drug resistance

Evaluation of Virulence Factors of *Candida albicans* Isolated from COVID-19 Patients with Oropharyngeal Candidiasis

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Background and Aim: Some oral pathological diseases can be worsened in COVID-19 patients. Based on recent experiences with handling severe COVID19 patients, *Candida* species particularly *Candida albicans* (*C. albicans*) may be a possible cause related to development of oropharyngeal candidiasis (OPC) in these individuals which begins with the colonization of the yeast, on the oral mucosa. Several factors, such as pathogen factors and host conditions, can impact yeast formation in the oral cavity. Therefore, our investigation was taken up with an aim to evaluate of *C. albicans* virulence factors which were obtained from COVID-19 patients with OPC.

Methods: to evaluate virulence attributes, forty-six *C. albicans* were isolated from oral lesions of COVID-19 hospitalized patients who were clinically suspected of OPC. Extracellular enzymatic activity, cell surface hydrophobicity (CSH), and biofilm formation (evaluated using MTT assay) were all investigated as virulence factors. In addition, the correlation between the biofilm formation and underlying conditions and risk factors was analyzed.

Results: All *C. albicans* isolates showed hemolysin activity, and CSH, 44 (95.7%) produced biofilm, and 43 (93.6%) produced proteinase. While, thirty-six (78%) of isolates were esterase positive (36/46), 65% of isolates were coagulase-positive (30/46) and 63% were phospholipase-positive (29/46). In addition, Biofilm development rates were significantly higher among cases that used central venous catheters.

Conclusion: Our study highlights the importance of *C. albicans* virulence factors as crucial attributes in pathogenesis of *C. albicans* in OPC patients as concurrent secondary infections in COVID-19 patients.

Keywords: Oropharyngeal candidiasis; Virulence factors; COVID-19; Biofilm

First Molecular Report of Causative Agent of Otomycosis Due to *Aspergillus luchuensis*

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Background and Aim: Otomycosis is a fungal infection of the external auditory canal with worldwide distribution. It is often reported from hot, humid, and dusty areas of tropical and subtropical regions. Among a broad spectrum of saprophytic filamentous fungi, yeasts, and dermatophytes, *Aspergillus* was reported as the most common genus responsible for otomycosis. *Aspergillus* currently includes 25 sections. The most common cause of otomycosis was reported as *Aspergillus* section Nigri (black aspergilli). Section Nigri comprises 27 species of which 11 were reported from different human clinical entities, including otomycosis, pulmonary aspergillosis, aspergilloma, and onychomycosis. Also, *Aspergillus* section Nigri, including *Aspergillus niger* and other cryptic species, has received increasing attention worldwide because of their role in industrial exploitation, prevention of food spoilage, and toxin production. To our knowledge, in this case, we report the first clinically relevant isolation of *Aspergillus luchuensis* identified by molecular technique as a causative agent of otomycosis.

Methods and results: A 43-year-old female patient with a left ear infection was admitted to a specialty ear, nose and throat (ENT) hospital (Amir Alam Hospital of Tehran University of Medical Sciences (TUMS), Tehran, Iran). The clinical presentations included otalgia (pain in the ear), otorrhoea (purulent discharge from the ear), itching and the otorrhoea had persisted for one month while she remained afebrile. On otoscopic examination, an eclipse was seen that occupied her left external ear canal. The audiogram showed a hearing loss in the left ear. The patient had a history of frequent swimming, the use of cotton swab and seasonal allergy. Otoscopic examination revealed inflammation in the external auditory canal. Debris samples were collected from the outer ear with aseptic precautions by sterile cotton swabs and sterile forceps. Collected samples underwent microscopic examination and culture immediately. Direct examination of the specimen, using 10% potassium hydroxide, revealed septate hyphae and fruiting bodies consistent with *Aspergillus* species. The specimen's culture onto sabouraud dextrose agar (SDA, Merck, Germany) plates yielded growth of colonies compatible with *Aspergillus* section Nigri. The amplicons were sequenced (Macrogen, South Korea), and the results were visually checked and compared with the GenBank sequences using nucleotide blast analysis (<https://www.blast.ncbi.nlm.nih.gov/Blast.cgi>).



Conclusion: Besides the necessity for accurate identification, evaluating susceptibility profiles of clinical *Aspergillus* isolates is also needed. This would support treatment, as well as enhance understanding of the epidemiology of *Aspergillus* species.

Keywords: *Aspergillus*; Section nigri; molecular; antifungal; otomycosis



Frequency of Bacterial and *Candida albicans* Secondary Infections in Sputum Samples of Patients with Covid-19 Admitted to Vali-e-Asr Hospital in Zanjan from March 2019 to March 2020.

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Background and Aim: Since simultaneous bacterial and fungal infections in hospitalized adults with Covid-19 virus caused by SARS-CoV-2 are not well known clinically, the distinction between isolated Covid-19 viral infection and the bacterial or fungal infection is challenging, and further research on secondary and associated infections is necessary. The aim of this study was to evaluate the frequency of bacterial and *Candida albicans* secondary infections in sputum samples of patients with Covid-19 admitted to Vali-e-Asr Hospital in Zanjan from March 2019 to March 2020.

Methods: The statistical population of this descriptive cross-sectional study included patients with Covid-19 admitted to Vali-e-Asr Hospital in Zanjan from March 2019 to March 2020. All subjects tested for secondary infection were included in the study by census method. Data were extracted from patients' files and entered into SPSS 20 statistical software and analyzed by descriptive and inferential methods.

Results: The results showed that laboratory findings were more likely positive in elderly men, people with disease, using medications and long hospital stay compared to young women, healthy people, without medication, short-term hospitalization showed less clinical symptoms (p-value of less than 0.05). It was also observed that the bacteria with the highest frequency in the studied patients were *Streptococcus* and *Staphylococcus* strains with 21.5% and 16.7% and the lowest frequency was *Acinetobacter* with 4.2%. The results showed that in fungi the highest frequency was in patients with *Candida albicans* strain with 6.7% and the lowest frequency with *Aspergillus* with 2%.

Conclusion: It is recommended that men, the elderly, the sick, and people with more clinical symptoms pay more attention to isolation and risk assessment of infectious factors.

Keywords: Covid-19; Secondary infection; Nosocomial infection

Frequency of *Candida* Species in the Oral Cavity of Narcotics and Stimulants Smokers in Isfahan, Using Polymerase Chain Reaction-Restriction Fragment Length Polymorphism Method

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Background and Aim: *Candida* species are a group of coexistent microorganisms in the oral cavity that opportunistically cause disease in vulnerable people, including addicts. This study is aimed at identifying and determining the frequency of *Candida* species in the oral cavity of the addicts to inform the epidemiological panel and to warn to prevent the resistance of the species.

Methods: In this study, sampling was done from the oral cavity of 83 addicts who referred to Addiction Treatment Centers in Isfahan, Iran, using moist sterile swab. The presence of yeast on the direct microscope slides of 58 samples was confirmed. To carry out culture and the primary identification, Sabouraud dextrose agar medium with chloramphenicol as well as HiCrome *Candida* agar medium was used. Isolates were identified by polymerase chain reaction (PCR) restriction fragment length polymorphism (using specific primers ITS1-ITS4 and MSP I enzyme) and Duplex PCR test (using specific primers CDU1-CDU2/CAL1-CAL2).

Results: Out of 93 *Candida* isolates which had been isolated from 58 culture-positive samples of the oral cavity, *Candida albicans* (41.93%) were the most prevalent species, followed by *Candida dubliniensis* (24.73%), *Candida glabrata* (21.50%), *Candida kefyr* (8.60%), *Candida tropicalis* (2.15%), and *Candida parapsilosis* (1.07%), respectively. In 33 cases (56.90%), the presence of more than one *Candida* species was observed.

Conclusion: Compared to the studies conducted on the oral cavity of healthy controls, smoking certain drugs can have a significant effect on the presence and frequency of *Candida* species, particularly *C. dubliniensis* and *C. glabrata*.

Keywords: Addiction; *Candida* oral cavity; polymerase chain reaction; restriction fragment length polymorphism

Fungal Colonization of Respiratory Tract in Symptomatic Patients Hospitalized in Pulmonary Units of Khorramabad Hospitals

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Background and Aim: During recent decades, pulmonary fungal diseases are being diagnosed with increasing frequency in parallel with the increase in immunocompromised patients. Pulmonary fungal diseases include fungal colonization, allergies, and infections of the lungs. In most cases, colonization is the first step in the progression to fungal lung infection. This study was to investigate the colonization of fungal elements in the respiratory tract of symptomatic patients admitted to the pulmonary ward of Khorramabad hospitals in 2021.

Methods: The samples included lavage alveolar bronchial, sputum and tracheal bronchus. Diagnosis was based on observation of fungal elements in direct microscopy using KOH 10% and culture.

Results: A total of 127 lung specimens (36 bronchoalveolar lavage (BAL) and 79 sputum and 12 tracheal bronchus samples) were obtained from symptomatic patients hospitalized in pulmonary units. Patients included 80 men and 47 women in the age group of 7 to 96 years. In this study, 90% of fungi were observed in patients who had at least one underlying factor for the development of fungal lung disease. Of these, 67 cases of fungal infections were positive in direct examination and culture. Organisms isolated from pulmonary specimens included: *Candida* species (34.6%) and *Aspergillus* species (11%) represent the two most commonly isolated species in the current study. *Candida albicans* subspecies had the highest frequency with 18.1%. Also, 1 case of invasive pulmonary aspergillosis and in other hand 2 case of Mucormycosis rhinocerebral and 11 case *Candida* colonization in the respiratory tract in patients with COVID-19, were diagnosed.

Conclusion: In this study *Candida* and *Aspergillus* was the most common fungal species isolated from symptomatic. Chronic Obstructive Pulmonary Disease (COPD) lung cancer, Covid-19 and diabetes mellitus were important underlying conditions for pulmonary fungal colonization and/or infection.

Keywords: colonization; Covid-19; respiratory tract

Fungal Keratitis Caused by *Pseudallescheria boydii*: Clinical and Mycological Characteristics

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Background and Aim: *Pseudallescheria boydii* keratitis is rare but important type of fungal keratitis because of the inherently resistance of the organism to many existing antifungal agents. We present the clinical characteristics, risk factors, treatment, and prognosis of patients with *Pseudallescheria boydii* keratitis, and also present the antifungal sensitivities of the isolated strain.

Methods: Slit-lamp and confocal microscopy were used for clinical examinations. Fungal isolates were identified based on morphological characteristics and DNA sequence of the internal transcribed spacer region (ITS). In vitro antifungal susceptibility testing for fungal isolates was performed according to the Clinical and Laboratory Standards Institute (CLSI).

Results: *Pseudallescheria boydii* identified in four patients. All patients had a history of ocular trauma. In clinical examination hypopyon were seen in three patients. The main antifungal medications were oral and topical voriconazole. After treatment the visual activity of all patients improved in 2–3 weeks.

Conclusion: These patients bold the importance of determining causative organism of fungal keratitis and their antibiotic susceptibility. Culture findings are limited in identifying organisms. Sequencing of polymerase chain reaction-amplified DNA is good for accurate and rapid identification of species that can be helpful for optimize treatment.

Keywords: Fungal keratitis; *Pseudallescheria boydii*; Antifungal

How Dose Chronic Periodontitis Affect the Average Number of *Candida Albicans* Colonies in the Periodontal Pockets of Improved Covid-19 Patients before and after Using Herbal Plant, Neem (*Azadirachta indica*) as a Mouthwash

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Background and Aim: This study has tried to raise the level of oral health literacy in improved Covid-19 patients by using a mouthwash made by an old herbal plant named Neem, so it can reduce the severity of periodontitis. Chronic periodontitis is an inflammatory, infectious disease of tissues that support the teeth and is caused by a particular microorganism or group of specific microorganisms. Periodontitis associated with other conditions, such as candidiasis caused different and resistant clinical signs. Candidiasis is an infection caused by *Candida* species such as *Candida albicans*.

Methods: 30 periodontitis patients with candidiasis who were improved from Covid-19 were selected. After sampling the periodontal pockets of patients using paper point No. 45. Samples of a paper point were placed in a 1ml sterile saline centrifuged and then were transferred to medium CHROMagar. The green colonies of *Candida albicans* (*C. albicans*) were found in the culture medium, were counted for each patient then all of the patients received cured leaves of Neem as mouthwash three times a day. After 6 weeks, the patient re-sampling of periodontal pockets was planted just like before and *C. albicans* were counted.

Results: Before treatment, the average number of colonies derived from the periodontal pockets of patients was 25.3, while the average number of colonies in periodontal pockets after treatment was 15.8. The average number of colonies after treatment was significantly reduced. Gender and age cannot affect the number of fungus *C. albicans* after treatment.

Conclusion: Conclusion: It seems that using the Neem plant as a herbal mouthwash can reduce the inflammation and bleeding caused by the fungus *C. albicans* in the mouth.

Keywords: Covid-19; Periodontitis; Candidiasis; *Candida albicans*; Neem

Identification and species distribution of *Candida non-albicans* in oral Candidiasis

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Background and Aim: *Candida* species normally exist as oral flora in humans. Non-albicans species are shown to be contributed in oral infection in immunocompromised patients. Sever oral infections can cause life-threatening contagion in these patients. Here we identified and measured distribution of *Candida non-albicans* isolates from oral candidiasis.

Methods: 85 oral clinical isolates of *Candida non-albicans* from 73 immunocompromised patients with oral infection were collected. All isolates were identified by culturing on CHROMagar and carbohydrate assimilation methods. ITS sequencing was carried out for final determination.

Results: The results showed that 56 isolates (65.88%) of all clinical isolates were *C. glabrata*. Followed by 18 isolates (21.17 %) *Candida parapsilosis*, 4 isolates (4.7 %) *Candida dubliniensis*, 4 isolates (4.7 %) *Candida tropicalis* and 3 isolates (3.52 %) *Candida krusei*.

Conclusion: Our data revealed that among non-albicans species, *Candida glabrata* is the most common cause of sever oral infection in immunocompromised patients.

Keywords: *Candida non-albicans*; oral infection; species distribution

Immunological Response to COVID-19 and Its Role as a Predisposing Factor in Invasive Aspergillosis

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Background and Aim: The world is involved with a pandemic coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2. The clinical manifestations of reported COVID-19-associated pulmonary impairments range from asymptomatic infections to a pneumonia-induced acute respiratory distress syndrome that requires mechanical ventilation. Fungal superinfections complicating the clinical course remain underexplored. Angiotensin-converting enzyme 2, the receptor for COVID-19 that is mainly expressed in airway epithelia and lung parenchyma, is considered an important regulator of innate immunity. With regard to the viral-cell interaction, imbalanced immune regulation between protective and altered responses caused by the exacerbation of inflammatory responses should be considered a major contributor to secondary pulmonary aspergillosis. In addition, the complex inherited factors, age-related changes, and lifestyle may also affect immune responses.

Results: The complication and persistence of invasive aspergillosis have been well described in patients with severe influenza or COVID-19. However, there is a scarcity of information about the immunological mechanisms predisposing patients with COVID-19 to fungal co-infections. Therefore, this study was conducted to investigate the aforementioned domain.

Conclusion: It can be concluded that the immune responses caused by SARS-CoV-2 can increase the risk of fungal superinfections in patients with COVID-19 which may lead to an increase in mortality rate.

Keywords: Aspergillosis; *Aspergillus fumigatus*; Immune responses; SARS-CoV-2

In vitro Antifungal Activity New Azole Against Mold and Yeast Strains Isolated from Otomycosis

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Background and Aim: *Aspergillus* and *Candida* species are the most commonly identified fungal pathogens in otomycosis. However, we usually encounter some difficulties in its treatment because many patients show resistance to antifungal agents, and present a high recurrence rate. The current research was conducted to compare the *in vitro* activities of Luliconazole (LUL) and Efinaconazole (EFN) and the nine comparators on these strains isolated from otomycosis.

Methods: The *in vitro* activities to nine common antifungal drugs (amphotericin B (AMB), voriconazole (VRC), fluconazole (FLU), itraconazole (ITC), ketoconazole (KTO), clotrimazole (CLO), nystatin (NYS), terbinafine (TRB), and caspofungin (CAS)) and two novel new azoles (LUL and EFN) against of 108 clinical isolates of *Aspergillus* and *Candida* species, that obtained from otomycosis, was performed according to CLSI broth microdilution document.

Results: The LUL and EFN had the geometric mean minimum inhibitory concentrations (GM MICs) of 0.098 and 0.109 µg/mL against all *Aspergillus* strains, respectively. Furthermore, the GM MICs of all *Candida* isolates for LUL, EFN, CAS, CLO, VRC, AMB, ITC, KTO, FLU, NYS, and TRB were calculated at 0.133, 0.144, 0.194, 0.219, 0.475, 0.537, 0.655, 1.277, 4.905, 9.372, and 13.592 µg/mL.

Conclusion: As the findings indicated, LUL and EFN showed the lowest GM MIC values against the examined species. Accordingly, these novel imidazoles and triazole antifungal agents can be concluded as proper candidates for the treatment of otomycosis caused by *Aspergillus* and *Candida* strains.

Keywords: Otomycosis; antifungal susceptibility; Efinaconazole; Luliconazole

In vitro Antifungal Activity of Luliconazole, Efinaconazole, and Nine Comparators Against *Candida dubliniensis* and *Candida africana* Isolated from Vulvovaginal Candidiasis

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Background and Aim: Vulvovaginal candidiasis (VVC) is a common and debilitating long-term illness affecting a million women worldwide. This disease is caused mainly by *Candida albicans* and a lesser extent by other species, including the two phylogenetically closely related pathogens *Candida africana* and *Candida dubliniensis*. In this study, we provide for the first time, data on the antifungal activity of two new drugs, Efinaconazole (EFN) and luliconazole (LUL).

Methods: Evaluation of the in-vitro susceptibility profiles of *C. dubliniensis* and *C. africana* to various antifungal drugs was performed according to the CLSI M27-A3 and M27-S4 documents.

Results: The geometric mean MIC against all tested isolates was 0.019 and 0.024 µg/ml for the two novel drugs EFN and LUL, respectively. However, in general, except for nystatin and terbinafine, all other antifungal agents tested showed low MIC values and therefore all the isolates were considered susceptible to these drugs.

Conclusion: As the findings indicated, LUL and EFN showed the lowest GM MIC values against the examined species. Accordingly, these novel imidazoles and triazole antifungal agents can be concluded as proper candidates for the treatment of VVC caused by *Candida dubliniensis* and *Candida africana*.

Keywords: Vulvovaginal candidiasis; Antifungal activity; Luliconazole; Efinaconazole

Increased Incidence of Rhino-orbital Mucormycosis in an Educational Therapeutic Hospital During the COVID-19 Pandemic in Western Iran: An Observational Study

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Background and Aim: COVID-19 patients, especially the patients requiring hospitalization, have a high risk of several complications such as opportunistic bacterial and fungal infections. Mucormycosis is a rare and opportunistic fungal infection that mainly affects diabetic and immunocompromised patients. An increase has been observed in the number of rhino-orbital mucormycosis in patients with COVID-19 admitted to Imam Khomeini Hospital, Kermanshah, Iran, since October 2020. This is a report of the frequency, risk factors, clinical manifestations, treatment and prognosis of COVID-19 associated with mucormycosis infection.

Methods: The medical records of COVID-19 patients with rhino-orbital mucormycosis who were diagnosed in an educational therapeutic hospital in Kermanshah, west of Iran were surveyed. Several parameters were analyzed including demographic, clinical, therapeutic and laboratory characteristics.

Results: Twelve patients with COVID-19-associated rhino-orbital mucormycosis were identified from 12 October to 18 November 2020. All cases reported as proven mucormycosis had a history of hospitalization due to COVID-19. Comorbidities mainly included diabetes mellitus (83.33%) and hypertension (58.33%). Seventy-five per cent of patients received corticosteroids for COVID-19 treatment. The sites of involvement were rhino-sino-orbital (83%) and rhino-sino (17%). Amphotericin B/ liposomal amphotericin B alone or in combination with surgical debridement or orbital exenteration was used as the first-line therapy. The overall mortality rate was 66.7% (8/12).

Conclusion: We found a high incidence of mucormycosis among COVID-19 patients. Diabetes mellitus and corticosteroid use were the dominant predisposing factor of mucormycosis. Mucormycosis is a life-threatening and opportunistic infection; therefore, physicians should know the signs and symptoms of the disease so that a timely diagnosis and therapy can be performed.

Keywords: co-infection; COVID-19; Iran; mucormycosis; SARS-CoV-2

Investigation of the Fungal Contamination of Equipment and Air of Mazandaran Hospitals in 2021

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Background and Aim: Several reports have shown that fungal spores are significant contaminating sources of equipment in hospitals that can potentially cause a wide range of nosocomial infections. Therefore, in this present study we aimed the distribution of fungi in equipment and air samples obtained from different hospitals in Sari, the capital of Mazandaran province.

Methods: During 2021, 81 open plates were used for sampling of air and 50 samples were collected from hospital equipment. All obtained samples were cultured onto Sabouraud's Dextrose Agar media.

Results: In this study, 85 colonies belong to 10 different genera of fungi were isolated. Dominant fungi were, *Cladosporium* (30.6%), *Aspergillus* (22.4%), *Rhizopus* (14.2%), *Penicillium* and *Trichoderma* (8.1%), *Alternaria*, *Geotrichum* and *Neoscytalidium* (4.08%), *Fusarium* and *Paecilomyces* (2.04%), respectively. The highest colony rates were isolated from the ICU (32.9%) and oncology (21.1%) units whereas the lowest rate belonged to the toxicology unit (1.1%).

Conclusion: Isolation of different fungal genera especially *Aspergillus* species from hospital equipment could be considered as a main source of fungal infections in patients at risk. For this reason, improving appropriate air-conditioning and good cleanliness with regular schedules is suggested in different units of the hospital.

Keywords: fungal infection; *Aspergillus*.

Investigation of the Genetic Diversity of *Candida* species Isolated from HIV and non-HIV Individuals in Kerman

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Background and Aim: Although most cases of invasive yeast infection are attributed to *Candida albicans*, there are increasing rates of disease by non-*C. albicans* species in various parts of the world. The rDNA sequences of *Candida* provide suitable targets for PCR as they comprise both conserved and variable regions. Molecular phylogeny is essential for obtaining a better understanding of the evolution of the genus *Candida* and the identification of the relative degree of the *Candida* species. This study aimed to investigate the genetic diversity of *Candida* species isolated from HIV and non-HIV individuals in Kerman.

Methods: Two hundred and thirty-two *Candida* isolates, including 111 *Candida* isolates obtained from 96 HIV/AIDS patients and 121 *Candida* isolates obtained from 98 non-HIV persons, were used in this study. To evaluate genetic diversity and relationships among *Candida* species, PCR products of nine clinical *Candida* isolates were randomly selected for DNA sequence analysis as a representative of each kind of species. The obtained nucleotide sequences were aligned with the sequences registered in NCBI GenBank. All nucleotide sequences were trimmed by Sequence Scanner version 1.0 (Applied Biosystems). Afterward, the similarity percentage was determined by the online BLAST tool and Sequence Alignment software, BioEdit V. 7.2.5.

Results: The phylogenetic analysis showed the presence of two main clades and six separate subclades. Accordingly, about 88.9% of the isolates were located in clade I, and 11.10% of the studied isolates were in clade II. Isolates H41, H60, and H63 isolated from the HIV/ AIDS patients were *Candida kefyr*, *Candida glabrata*, and *Candida tropicalis*, respectively. Isolates H6 and H65 obtained from the HIV/AIDS patients were *C. albicans*. H59 and H40 isolates collected from the HIV/AIDS patients were *Candida parapsilosis*. Isolate S7 that was obtained from non-HIV individuals was *Candida guilliermondii*. Isolate S9 was isolated from non-HIV persons and was *Candida lusitanae*.

Conclusion: Accurate identification of *Candida* species is essential due to antifungal susceptibility patterns that vary among these species. Proper identification helps select antifungal drugs for prevention and treatment. Phylogenetic analysis is recommended to gain a better understanding of *Candida* genus evolution, determine the relative degree of *Candida* species, and deduce the evolutionary pathways of some gene families.

Keywords: *Candida* species; HIV individuals; non-HIV individuals; phylogeny; genetic diversity

Invitro Antifungal Susceptibility to Eight Antifungal Drugs against *Candida* species Isolated from Vulvovaginitis

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Background and Aim: Vulvovaginal candidiasis (VVC) is a significant health issue due to *Candida* species. Although *Candida albicans* is considered a major causative agent of vaginal candidiasis, non-albicans species have increased during previous decades. This research aimed at assessing antifungal susceptibility of VVC isolated *Candida* spp.

Methods: Broth microdilution, according to the M27-A3 and M27-S4 CLSI documents, were employed for determining the antifungal susceptibility tests of caspofungin (CAS), voriconazole (VRC), itraconazole (ITC), fluconazole (FLU), clotrimazole (CLO), ketoconazole (KTO), amphotericin B (AMB), and nystatin (NYS).

Results: The geometric mean values to all *Candida* strains were in increasing order as the following: CAS, 0.075 µg/mL; VRC, 0.091 µg/mL; ITC, 0.15 µg/mL; AMB, 0.22 µg/mL; CLO, 0.23 µg/mL; KTO, 0.28 µg/mL; NYS, 0.88 µg/mL; FLU, 1.48 µg/mL. Further, the MIC ranges of all *Candida* isolates to the tested antifungal agents were in increasing order as follows: CAS: 0.031 - 0.25 µg/mL, KTO and ITC: 0.031 - 2 µg/mL, VRC: 0.031 - 4 µg/mL, CLO and AMB: 0.031 - 8 µg/mL, NYS: 0.06 - 4 µg/mL, and FLU: 0.12 - 128 µg/mL.

Conclusion: We reported 1 (7.2 %) *Candida glabrata* isolate resistance to FLU and 2 (14.3%) *Candida glabrata* isolates susceptible-dose-dependent (SDD) to CAS. We also reported 6 (4.5%), 5 (3.8%), and 2 (1.5%) *C. albicans* resistance to ITC, FLU, and AMB, respectively, but 100% *C. albicans* susceptible to CAS and VRC.

Keywords: Vulvovaginal Candidiasis; Antifungal Agents; *Candida albicans*; *Candida glabrata*

Molecular Characterization of Fungal Colonization on the Provox™ Tracheoesophageal Voice Prosthesis in Post Laryngectomy Patients

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Background and Aim: Successful voice rehabilitation is critical for the patients’ voice communication after surgery. Tracheoesophageal voice prostheses (TVPs) have been the gold standard in rehabilitation, after laryngectomy, producing faster and premier voicing towards esophageal speech. Fungal colonization shortens the device’s lifetime and leads to prosthesis dysfunction, leakage, and subsequent respiratory infection unless replaced with a new one immediately. Therefore, in the current study, we aimed to investigate the fungal colonization patterns and to propose prophylactic measures that shall increase the longevity of voice prostheses to decrease patients’ discomfort and health cost.

Methods: Failed TVPs were removed - due to leakage and/or aspiration - from 66 laryngectomized patients and examined. Fungal colonization patterns were assessed using DNA sequencing techniques. Furthermore, the susceptibility to fluconazole, amphotericin B, nystatin, and white vinegar was evaluated according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.

Results: Resident fungal species from the upper airways colonized all the 66 TVPs (100%). Diabetes (31%) and smoking (98%) were the predominant underlying disease and predisposing factors, respectively. Among the 79 fungal agents isolated from the 66 TVPs, *Candida glabrata* (n=25, 31.7%) was the most common. A significant reduction in minimum inhibitory concentration (MIC) values was observed for white vinegar when used alone.

Conclusion: Conclusion: Our study revealed that white vinegar at a very low concentration could decrease the amount of fungal colonization on TVPs without any adverse effects; its wide accessibility and affordability ensure a decrease in the overall health cost.

Keywords: Voice prostheses; fungal colonization; antifungal susceptibility test; white vinegar; minimum inhibitory concentration; total laryngectomy

Molecular Identification of Aflatoxigenic *Aspergillus* species in Dried Nuts and Grains Collected from Tehran, Iran

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Background and Aim: Agricultural commodities contaminated by molds and mycotoxins can be considered as public health problems in less developed countries, particularly in Iran. Hence the main purpose of this study was to identify mold fungi and molecular analysis of the most important species of aflatoxin-B1-producing *Aspergillus* species in some dried nuts and grains in local markets in Tehran.

Methods: Two hundred fifty samples of wheat, rice, corn, pistachios, and peanuts were collected from the five different locations of Tehran between January 2018 and January 2019. The samples were analyzed by using direct seed inoculation method and grain crushing method. Fungal strains were identified as *Aspergillus* sp. on the basis of morphological characters and further confirmed by using of β -tubulin gene sequencing. To differentiate between aflatoxigenic and non-aflatoxigenic *Aspergillus* sp., the isolates were screened for the presence of aflatoxigenic genes (nor-1, ver-1, omtA, and aflR).

Results: One-handed forty-eight aflatoxigenic *Aspergillus* species (144 *Aspergillus flavus* and 4 *Aspergillus parasiticus*) were identified and aflR gene was the most frequent gene in these species. Five isolates (4 *A. flavus*, 1 *A. parasiticus*) had quadruplet pattern, 64 isolates (63 *A. flavus*, 1 *A. parasiticus*) had more than 1 gene and 39 isolates (38 *A. flavus*, 1 *A. parasiticus*) did not have any genes.

Conclusion: Developing an effective and efficient aflatoxin control program requires identifying and managing key elements that are effective in reducing poison production at farm level or in storage conditions. In this regard, special attention to climate factors, planting systems, before harvesting and after harvest are considered as the most important cases, in order to minimize the chance of producing these toxins in all stages.

Keywords: Grains; Nuts; *Aspergillus*; Aflatoxin

Molecular identification of *Candida albicans*, *Candida dubliniensis*, and *Candida africana* Isolated from Vulvovaginal Candidiasis based on Hyphal Wall Protein 1 Gene

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Background and Aim: Vulvovaginal candidiasis (VVC) is a common and debilitating long-term illness affecting a million women worldwide. This disease is caused mainly by *Candida albicans* (*C. albicans*) and a lesser extent by other species, including the two phylogenetically closely related pathogens *Candida africana* (*C. africana*) and *Candida dubliniensis* (*C. dubliniensis*). In this study, we report detailed molecular epidemiological data about the occurrence of these three pathogenic yeasts in Iranian patients affected by VVC, or its chronic recurrent form (RVVC).

Methods: A total of 133 vaginal yeast isolates, presumptively identified as *C. albicans* by phenotypic and restriction analysis of rDNA, were further analyzed by using a specific molecular method targeting the Hyphal wall protein 1 (HWP1) gene.

Results: Based on the molecular results, among 133 germ-tube positive isolates, we identify 119 *C. albicans* (89.47%), 11 *C. africana* (8.27%), and 3 *C. dubliniensis* (2.26%) isolates.

Conclusion: Although *C. albicans* remains the most common *Candida* species recovered from Iranian VVC/RVVC patients, our data show that its prevalence may be slightly overestimated due to the presence of difficult-to-identify closely related yeast, especially *C. africana*.

Keywords: *Candida africana*; *Candida dubliniensis*; HWP1 gene

Mucormycosis Superinfection during COVID-19 Pandemic: a Review

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Background and Aim: COVID-19 has led the world to a crisis, with the course of disease ranging from mild respiratory illness to severe respiratory failure, making it a global concern. COVID-19 patients are acquiring secondary infections such as mucormycosis also known as black fungus disease in the middle of the pandemic. Mucormycosis (previously called zygomycosis) is a serious but rare fungal infection that mainly affects patients who are immunosuppressed. Most of these patients have diabetes and are treated with steroids for SARS-CoV-2 infection. The combination of these two might have made them more likely to have fungal attack. In this study we investigate the role of mucormycosis and its challenges causing black fungus in patients with COVID-19.

Methods: PubMed, Scopus, Magiran, and Google Scholar were searched using keywords: “COVID-19”, “Mucormycosis”, “Diabetes”, “Immunosuppression”, “Steroids”, and “black fungus”.

Results: Mucormycosis has been associated with many underlying conditions that can cause an individual to have infections. Some of these factors include diabetes, steroidal use, neutropenia, organ or stem cell transplantation, hematological disorders, trauma and burns, metabolic acidosis, intravenous drug usage, renal insufficiency, broad-spectrum antibiotics and etc. apart from taking heavy dose of steroids, COVID-19 patients use oxygen masks and ventilators to fight SARS-CoV-2 infection, which can open a pass for the Mucorales fungus. Black fungus which appears in patients admitted in the Intensive Care Unit (ICU) can be fatal, causing loss of vision and even death. Early symptoms include blockage of the nasal cavity and greyish-black pigmentation in the nose or oral cavity. Ocular swelling is caused by spores of fungus near the eyes, and a few patients might get lesions over the cheeks. At the next stage, this fungus can infect the brain which makes the disease quite severe. As we know, the fatality rate goes up to 50%.

Conclusion: As we continue to investigate more outcomes about the clinical conditions of COVID-19, we should be really careful and watch out for coinfections alongside the way. Mucormycosis, an opportunistic fungus, should be controlled by researchers and healthcare professionals. Understanding its influence and range of severity, especially in COVID-19 patients due to their use of oxygen therapy is really vital. Also, healthcare workers should be educated to change flow meters frequently and to sterilize oxygen tubing. Treatments also include antifungal drugs like Amphotericin-B.

Keywords: COVID-19; Mucormycosis; Diabetes; Immunosuppression; Steroids; black fungus

Oral Candidiasis of ICU Patients with COVID-19: Identification and Antifungal Susceptibility Patterns of Isolates

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Background and Aim: The recent global pandemic of COVID-19 has predisposed the intensive care units (ICUs) patients to candidiasis. Oral candidiasis is a fungal infection caused by *Candida* species, with a high mortality rate. The objective of this study was to identify and evaluate antifungal susceptibility patterns of *Candida* spp. isolated from critically ill COVID-19 patients on long-term ventilation (>7 days) with oral problems.

Methods: Swabs were collected from the oral cavity of 30 COVID-19 patients. Samples confirmed for candidiasis by microscopic and culture examination. *Candida* strains were identified by ITS-PCR. *In vitro* antifungal susceptibility tests against current antifungal agents were performed according to CLSI (M27-A3/S4) guideline.

Results: A total of 30 study participants were enrolled. 18 (60%) males and 12 (40%) females. The mean age was 64 ± 5 years. Among 30 patients, 24 (80%) had diabetes, followed by prior long-term steroid intake (>3 times in the past 6 months) 14(46.66%) and cavitary disease in chest X-ray in 8 (26.66%) patients. The most commonly isolated species among patients were *Candida albicans* (n = 23; 76.66%), *Candida glabrata* (n = 5; 16.66%), and *Candida parapsilosis* (n = 2; 6.66%). All *Candida* spp. isolates from patients were susceptible to fluconazole, amphotericin B and caspofungin.

Conclusion: Local information about the drug susceptibility in *Candida* species is valuable for selecting the empirical therapy of COVID-19 patients.

Keywords: Oral Candidiasis; COVID-19; *Candida* species

Overview on the Prevalence of Fungal Infections, and Microbiome Role in COVID-19 Patients

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Background and Aim: Patients with severe COVID-19, such as individuals in intensive care units (ICU), are exceptionally susceptible to bacterial and fungal infections. The most prevalent fungal infections are aspergillosis and candidemia. Nonetheless, other fungal species (for instance, *Histoplasma* spp., *Rhizopus* spp., *Mucor* spp., *Cryptococcus* spp.) have recently been increasingly linked to opportunistic fungal diseases in COVID-19 patients. These fungal co-infections are described with rising incidence, severe illness, and death that is associated with host immune response. Awareness of the high risks of the occurrence of fungal co-infections is crucial to downgrade any arrear in diagnosis and treatment to support the prevention of severe illness and death directly related to these infections. This review analyses the fungal infections, treatments, outcome, and immune response, considering the possible role of the microbiome in these patients.

Methods: The search was performed in Medline (PubMed), using the words “fungal infections COVID-19”, between 2020–2021.

Results: In general, data show that COVID-19 patients in ICU seem are more susceptible to fungal infections, when compared with patients without ICU admission, due to their immunosuppression status (the same case of HIV patients). Moreover, probably the incidence of aspergillosis is higher in COVID-19 patients, as the virus particularly affects the respiratory system. Correspondingly, in COVID-19 patients, the mortality rate is high in the case of co-infections (both bacterial and fungal species).

Conclusion: The incidence of fungal infections is rising in covid-19 cases due to the increased immunosuppressed population. On the other hand, the diversity of the microbiome in a population creates a different range of severity of infections in each individual. The use of probiotics might open a new insight into the management of fungal pathogens, but there is much to uncover about the probable side effectiveness of clinical application of those in COVID-19 cases.

Keywords: Fungal infections; Covid-19 patients; Microbiota and probiotics

Prevalence of Dermatophytes by Direct Microscopy and Culture Over a Period of 4 Years in Sina Hospital, Tabriz

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Background and Aim: Dermatophytes are a group of fungi which have a tendency towards createine and thus attack the skin, hair and nails thereby causing fungal infections. Dermatophytes are classified into three types comprising *Trichophyton*, *Microsporum*, and *Epidermophyton*, and triggering the dermatophytosis in humans and animals. These fungi are distributed throughout the world, including Iran, and are the most common pathogens involved in skin infections. Dermatophytosis is one of the most important global issues and is largely dependent on the social and economic conditions of any localized geographical region. The aim of this study was to determine the distribution and frequency of fungi in causing the infection based on sex, age and occupation in Sina Hospital.

Methods: A total of 2,151 patients were evaluated for dermatophytes from October 2017 to September 2021. Nail, skin, and scalp scrapings were collected from these patients. Positive cases detected by direct microscopy were confirmed after culture in Sabouraud's Dextrose Agar (SDA).

Results: Among the total patients enrolled in the study, 418 clinical specimens were identified positive for dermatophytes by microscopy. Of these, 87% specimens showed dermatophytic growth on culture media. Majority of these encompassed *Trichophyton* (65%) followed by the *Microsporum* and *Epidermophyton* (31% and 4% respectively). Commonest age group affected was 31- 40 years, with more prevalence in males (n=214) comparative to females (n=203), though not significant statistically. The most common occupation of these patients was housewife, student and self-employments respectively.

Conclusion: Fungi are prone to warm, humid environments, thus infects certain areas of the body, including the groin, scalp, feet, etc. which are more vulnerable for fungus to grow and multiply leading to fungal infections. Dermatophysis is very common in our region, may be due to deprivation of hygienic conditions. Laboratory diagnosis using conventional methods like direct microscopy are easily accomplished but require skill and expertise for identification, thus culture methods are mandatory. Presence of dermatophysis warrants the implementation of intensive epidemiological studies.

Keywords: dermatophytes; Fungi; *Trichophyton*; *Microsporum*; *Epidermophyton*

Prevalence of Fungal Urinary Tract Infections in Children Admitted to the Nephrology Division of Aboozar Children Hospital, Ahvaz, Iran

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Background and Aim: The prevalence of urinary tract infections (UTIs) has been reported in children with nephrotic syndrome. But not much is known about the occurrence of urinary tract fungal infections in children, and previously reported series have reported mostly bacterial causes of UTIs to date. Hence, the present study aimed to describe the epidemiology of urinary tract fungal infections in a tertiary care pediatric hospital.

Methods: A single-center cross-sectional study was performed in the nephrology ward of Abuzar Pediatric Hospital affiliated with Ahvaz Jundishapur University of Medical Sciences between March 21, 2021, to November 21, 2021. Antifungal susceptibility was determined by the disk diffusion method according to the CLSI guideline.

Results: In total, 61 positive fungal samples were identified from 50 hospitalized children in the age range of 1 month to 17 years. Of these, 63% were boys and 37% were girls. The prevalence of urinary tract fungal infections in hospitalized children was estimated at 10%. The causative fungal organism was yeast in all cases. Overall, mild infections were reported in 66% of cases and moderate to severe infections in 34% of cases. 71% of moderate to severe infections occurred in boys and 75% of urinary tract fungal infections in girls were mild. While all patients were sensitive to posaconazole, all patients demonstrated intermediate susceptibility or drug resistance to itraconazole. Among patients, 12.5%, 25%, and 37.5% showed intermediate susceptibility to fluconazole, voriconazole, and amphotericin B, respectively. And 12.5% drug resistance was observed for each of voriconazole and fluconazole drugs.

Conclusion: There is a high prevalence of yeast infections in children with suspected UTI. Also, boys are more likely to get yeast infections and the severity of the infection is higher than girls. The present study demonstrated the importance of diagnosing and selecting the appropriate drug for urinary tract fungal infections in hospitalized children.

Keywords: Urinary tract fungal infections; Children; Yeast; Antifungal susceptibility

Prevalence of Invasive Pulmonary Aspergillosis in the Third and Fourth Waves of the COVID-19 by Using Serum Galactomannan Antigen Test

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Background and Aim: Nowadays, the pandemic of coronavirus (COVID-19) is a major health problem that takes over entire the world. The first cases of this viral disease in Iran were reported in February 2020. Fungal infections, especially invasive Aspergillosis as a co-infection have increased considerably during the outbreak of COVID-19 disease. Invasive pulmonary Aspergillosis is a fatal fungal disease caused by the invasion of *Aspergillus* species to the host's tissue. Detection of the serum galactomannan (GM) antigen by means of immunoassay enzyme might be a useful tool for the diagnosis and treatment of invasive Aspergillosis.

Methods: From Nev. 2020 to the Sep. 2021, a number of 157 patients (87.9% from 6 hospitals and 12.1% of patients were outpatients) were evaluated and followed for invasive Aspergillosis in Ahwaz. All of the collected serum samples were assayed for GM antigen using the *Aspergillus* Galactomannan enzyme-linked immunosorbent assay (ELISA) kite. Serum index ≥ 0.5 is considered to be positive for GM antigen

Results: Overall, GM antigen was assayed in 157 samples from 103 (65.6%) patients who got an infection in COVID-19, (31(56.36%) cases in the third wave of COVID-19 and 72 (70.58%) cases in the fourth wave of COVID-19) and other patients did not infect by the COVID-19. 55 samples of the GM antigens were collected during the third wave of the COVID-19, which the GM antigen test was positive in 12 samples (21.84%) and negative in 43 samples (78.16%) a number of 102 samples collected during the fourth wave, that was positive in 56 samples (54.92%) and negative in 44 samples (40.8%).

Conclusion: Results of this study demonstrated that the level of circulating antigen dependent on COVID-19 disease which showed that the level of circulating antigen is dependent on the strains and prevalence of COVID-19 disease. Because the level of circulating antigen in the fourth wave of COVID-19 (Delta strain) was more than its level in the third wave.

Keywords: Coronavirus (COVID-19); Enzyme-linked immunosorbent assay (ELISA); Invasive Aspergillosis; and Delta strain; galactomannan (GM) antigen

Prevalence of Probable Invasive Pulmonary Aspergillosis in Ahvaz, Southwest Iran During the Third and Fourth Waves of the COVID-19 Pandemic by Using Serum Galactomannan antigen test

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Background and Aim: Nowadays, the pandemic of coronavirus (COVID-19) is a major health problem that takes over entire the world. Fungal infections, especially invasive Aspergillosis as a co-infection have increased considerably during the outbreak of COVID-19 disease. Invasive pulmonary Aspergillosis (IPA) is a fatal fungal disease caused by the invasion of *Aspergillus* species to the host's tissue. The aim of this study was to evaluate probable IPA in patients with complicated acute respiratory distress syndrome (ARDS).

Methods: From Nev. 2020 to the Sep. 2021, a number of 157 patients (87.9% from 6 hospitals and 12.1% of patients were outpatients) were evaluated and followed for probable IPA in Ahwaz. Probable IPA was determined based on clinical findings, chest CT and positive serum galactomannan (GM) antigen results. All of the collected serum samples were assayed for GM antigen using the Dynamiker Aspergillus Galactomannan enzyme-linked immunosorbent assay (ELISA) kite. GM index cutoff ≥ 0.5 is considered to be positive for GM antigen.

Results: Overall, GM antigen was assayed in 157 patients. From 103 (65.6%) patients, 31(56.36%) and 72 (70.58%) cases respectively in the third and fourth wave of COVID-19 pandemic infected with COVID-19 and other patients were not infected by the COVID-19. From the total of 55 samples collected during the third wave of the COVID-19, which the GM antigen test was positive in 12 samples (21.84%) and negative in 43 samples (78.16%) as well as from 102 samples collected during the fourth wave, that was positive in 56 samples (54.92%) and negative in 44 samples (40.8%).

Conclusion: Due to the higher prevalence of probable IPA in the fourth wave of the COVID-19 compared to the third wave, it can be concluded that COVID-19 infection is a major risk factor for IPA. Furthermore, clinical weekly screening of serum GM strongly recommended to perform in COVID-19 patients.



Keywords: Coronavirus (COVID-19); Enzyme-linked immunosorbent assay (ELISA); invasive pulmonary Aspergillosis; Serum Galactomannan antigen



Prevalence of Vulvovaginal Candidiasis and Its Species among Women with Symptoms of Vaginal Infection

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Background and Aim: Vulvovaginal candidiasis (VVC) is a very common vaginal infection that affects 75% of women at least once in their lifetime. Vaginitis caused by non-albicans species is clinically indistinguishable from albicans species and also usually resistant to treatment. Due to the lack of proper treatment and resistance of some *Candida* species to azole drugs, the prevalence of disease has increased and its complications threaten women.

Methods: This study was performed on 120 women referred to Tabriz medical centers with symptoms of vaginal infection. Two sterile swabs were given from the posterior fornix of vagina. The first sample being stretched on a slide and Wiff test was performed. Second sample was cultured in Sabouraud Dextrose Agar (SDA) containing 0.05% chloramphenicol and the growth colonies identified by chromo agar. In addition, a stained slide with methylene blue was used for direct observation of fungal cells, clue cells and other abnormalities. Bacterial and negative samples were excluded from the study.

Results: Prevalence of abnormal vaginal discharge, itching, burning, dyspareunia and dysuria were 87%, 85%, 48%, 31% and 22% respectively. The mean age of patients was 34 ± 0.7 years. From 120 vaginal specimens, 69 specimens were identified as *Candida* including 37 *Candida albicans*, 15 *Candida glabrata*, seven *Candida parapsilosis*, seven *Candida krusei* and three *Candida tropicalis*.

Conclusion: Due to the high prevalence of VVC and its complications; proper diagnosis, treatment and prevention of recurrence are essential, which is done according to the accurate knowledge of the organism, the signs and symptoms of disease.

Keywords: vulvovaginal candidiasis (VVC); *Candida* spp.; Vaginitis

Rapid Identification of *Candida* Isolates Obtained from Healthy Individuals by Real-time PCR-High Resolution Melting Analysis

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Background and Aim: Candidiasis is considered an essential opportunistic fungal disease worldwide and remains a clinical problem, mainly among immunocompromised patients. Due to species-specific antifungal resistance patterns, reliable detection, quantification, and identification methods are required. High-resolution amplicon melting analysis was introduced as a closed-tube method for genotyping and mutation scanning. This study aimed to rapidly identify 121 *Candida* isolates obtained from the oral cavity of healthy individuals with Real-time PCR and HRM.

Methods: In this study, 121 *Candida* isolates from 98 healthy individuals obtained from the oral cavity were used. Real-Time PCR identified these *Candida* isolates and high-resolution melting curve analysis compared to the reference isolates.

Results: In healthy persons, we identified eight species of *Candida* including *C. albicans* (n = 46; 38.33%) followed by *Candida glabrata* and *Candida krusei* (each one, n = 18; 15%), *Candida tropicalis* (n = 13; 10.83%), *Candida lusitanae* (n = 12; 5.17%), *Candida parapsilosis* (n = 10; 4.31%), and *Candida kefyr* and *Candida guilliermondii* (each one, n = 2; 1.66%).

Conclusion: High resolution melting analysis (HRM) technique is a rapid and cost-effective technique for accurate identification of *Candida* species. With the correct diagnosis of *Candida* species, this method allows fast and targeted treatment of patients with invasive candidiasis.

Keywords: *Candida* spp.; High resolution melting; healthy persons

Risk Factors Associated with Oropharyngeal Candidiasis in COVID-19 Patients: A Case Control Study

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Background and Aim: With the emergence and spread of the coronavirus disease-19 (COVID-19) in the world, humans have faced with the biggest challenge in health care systems in recent decades. The aim of the present study is to identify risk factors associated with oropharyngeal candidiasis (OPC) in COVID-19 patients.

Methods: The total number of confirmed COVID-19 patients was 218 (105 participants as cases who experienced OPC and 113 participants as controls without any evidence of OPC). The questionnaire used in this study consists of demography data, treatment strategy, clinical and laboratory data, and underlying diseases to collect information at the time of clinical OPC and follow them until end of hospitalization.

Results: Pseudomembranous candidiasis (77/105, 73.3%) was the most prevalent form of OPC in case patients. The majority of case (58.1%) and control (58.4%) groups were male. Increasing age of COVID-19 patients ($P = 0.03$) and length of hospitalization ($P = 0.016$) were significantly associated with OPC. Diabetes ($P = 0.003$), solid tumor ($P = 0.019$), and hypertension ($P = 0.000$) were the most common underlying conditions. Use of dentures ($P = 0.003$), and poor oral hygiene ($P = 0.000$) were related to OPC in case group. Therapy with chloroquine ($P = 0.012$), IVIG ($P = 0.001$), diuretics ($P = 0.000$), and corticosteroid pulse therapy ($P = 0.000$) were significant associated with the development of OPC in case patients.

Conclusion: It is reasonable to consider that the old age, length of hospitalization, poor oral hygiene, corticosteroids usage, diabetes, solid tumor, and hypertension may predispose to the development of OPC in COVID-19 patients.

Keywords: Oropharyngeal candidiasis; COVID-19; Risk factors

Synthesis and characterization of chitosan–base nanofiber with antifungal activity *in vitro*

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Background and Aim: The incidence of candidiasis has increased dramatically over the past several decades. In recent years, a variety of new antifungal agents with therapeutic characteristic have progressed against fungal infections. This study aimed to synthesize nano fibers with electrospinning method, antifungal activity of nanofiber and cytotoxicity effect of nano fiber in comparison to Fluconazole *in vitro*.

Methods: Nano fibers were prepared by Chitosan with PVA and CNTs-COOH with electrospinning device. The average size of fibers was 70-80 nm. The MTT Assay was done to evaluate the toxicity of nanofibers on sw480 cell line.

Results: As a result the usage of fluconazole alone had higher MIC in comparison of fluconazole in nano fiber with Chitosan polymers. The MTT Assay showed in nanofiber form the toxicity of fluconazole was lower than fluconazole alone (P-value < 0.05).

Conclusion: According the finding the synthesized Nano fibers containing Chitosan improve the drug delivery system with minimum cell toxicity effect. However, therapeutic application of nano fiber against candidiasis needs further investigation *in vitro* and *in vivo*.

Keywords: Antifungal agents; *Candida albicans*; Nano Fiber; Chitosan

The Effects of Ellagic Acid on the Growth and Ergosterol Biosynthesis in Clinical Isolates of *Candida tropicalis*

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Background and Aim: *Candida tropicalis* (*C. tropicalis*) is one of the most important *Candida* species that is considered as second most virulent yeast after *Candida albicans*. In addition, it produces a wide range of virulence factors; moreover, it is one of the causes of oral nosocomial infection in immunocompromised patients. Here, we studied the effects of ellagic acid on the growth and ergosterol biosynthesis of clinical isolates of *C. tropicalis* obtained from oral candidiasis.

Methods: 13 clinical isolates of *C. tropicalis* were treated with serial two-fold concentrations of ellagic acid (9.375-300 µg/ml) for 48 h at 35 °C. The weights of the fungal mass were measured as a sign of the fungal growth and the content of ergosterol was assessed by standard method.

Results: Ellagic acid inhibited *C. tropicalis* growth with MIC in the range of 18.75- 75 µg/ml, in 13 clinical isolates. The ergosterol biosynthesis was also reduced in dose-depending manner after treating with different concentrations of ellagic acid.

Conclusion: Our results revealed that ellagic acid may be considered as a potential agent for establishment of new antifungal drugs.

Keywords: Ellagic acid; *Candida tropicalis*; ergosterol

Using Artificial Neural Network to Predict Predisposing to Vulvovaginal Candidiasis among Vaginitis Cases

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Background and Aim: Mycotic vaginitis is common mucosal infection caused mainly by the yeast *Candida* species. The ANN models can identify patterns between variables with the minimum squared errors and high correlation coefficients without limiting assumptions such as normality. In predicting, modeling, detecting, and classifying relational patterns among variables, artificial neural network (ANN) models, compared to conventional models such as the response surface methodology (RSM), have broad applications. This study attempts to predict vulvovaginal candidiasis (VVC) relative to risk factors and symptoms among Vaginitis Cases using the Artificial Neural Network (ANN) model.

Methods: In this proposal, 250 vaginal swabs were collected from women referred to gynecology Clinics Birjand, Iran. The swabs were microscopically examined and cultured, and participants' demographic information was recorded. A combination of different transfer functions in connection to input-hidden and hidden-output layers was assessed with ANN algorithms. The number of hidden neurons also was changed from 2 to 15. Statistical analyses were performed using SPSS (ver.21) at 0.05 significant level

Results: The prevalence of VVC among 250 patients, with a mean age of 34.6 ± 9.99 years old, was detected 41.0%. To predict predisposing to VVC, the ANN algorithms were optimal when hyperbolic tangent and sigmoid functions were respectively used in hidden (with eight hidden neurons) and output layers. Age (17.02%), miss carriage (11.14%), sexual intercourse (8.55%), intercourse pain (6.94%), education (5.70%), natural vaginal delivery (NVD) (5.61%), and under abdominal pain (5.27%) were important variables.

Conclusion: The result of the ANN model revealed that using factors such as age, abortion history, number of sexual intercourses, dyspareunia, education, NVD, and Lower abdominal, it is possible to predict VVC infection through ANN statistical model.

Keywords: Artificial Neural Network; Vulvovaginal Candidiasis; Risk-factors; Clinical symptoms; Statistical model

Virulence factors and azole-resistant mechanism of *Candida tropicalis* isolated from candidemia

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Background and Aim: Limited knowledge exists on the virulence factors of *Candida tropicalis* (*C. tropicalis*) and the mechanisms of azole resistance that lead to an intensified pathogenicity and treatment failure. We aimed to evaluate the virulence factors and molecular mechanisms of azole resistance among *C. tropicalis* isolated from patients with candidemia.

Methods: Several virulence factors, including extracellular enzymatic activities, cell surface hydrophobicity (CSH), and biofilm formation were evaluated. Antifungal susceptibility pattern and expression level of ERG11, UPC2, MDR1, and CDR1 genes of eight (4 fluconazole resistance and 4 fluconazole susceptible) clinical *C. tropicalis* isolates were assessed. The correlation between the virulence factors and antifungal susceptibility patterns was analyzed.

Results: During a four-year study, forty-five *C. tropicalis* isolates were recovered from candidemia patients. The isolates expressed different frequencies of virulence determinants as follows: coagulase 4 (8.9%), phospholipase 5 (11.1 %), proteinase 31 (68.9 %), esterase 43 (95.6 %), hemolysin 44 (97.8%), biofilm formation 45 (100%) and CSH 45(100%). All the isolates were susceptible to amphotericin B and showed the highest resistance to voriconazole. There was a significant positive correlation between micafungin minimum inhibitory concentrations (MICs) and hemolysin production ($r_s = 0.316$). However, we found a negative correlation between fluconazole MICs and esterase production ($r_s = -0.383$). We observed the high expression of ERG11 and UPC2 genes in fluconazole-resistant *C. tropicalis* isolates.

Conclusion: *C. tropicalis* isolated from candidemia patients extensively displayed capacities for biofilm formation, hemolysis, esterase activity, and hydrophobicity. In addition, the overexpression of ERG11 and UPC2 genes was considered one of the possible mechanisms of azole resistance.

Keywords: *Candida tropicalis*; Virulence factors; Biofilm; Candidemia; Azole resistance

The Comparison of 21-Plex PCR, PCR-RFLP and Vitek 2 System in Identification of Yeasts Isolated from Clinical Samples

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Background and Aim: Non-albicans *Candida* species have been increasing in the last two decades and the pattern of drug resistance in these species are different. Accurate identification of yeasts from colonies as well as clinical specimens is important for management and treatment of the disease. The aim of this study was to evaluate and compare the 21-Plex PCR, PCR-RFLP and Vitek 2 system for yeast identification against rDNA sequencing as a gold standard method.

Methods: One hundred thirty-two yeast isolates were selected from Tehran medical mycology laboratory (TMML) and entered into the study. Identification was performed by 21-Plex PCR, PCR-RFLP, Vitek 2 system and rDNA sequencing of the internal transcribed spacer region (ITS1-5.8S-ITS2).

Results: PCR-RFLP correctly identified 91.7 % of common pathogenic *Candida* species (*Candida albicans*, *Candida glabrata*, *Candida parapsilosis*, *Candida tropicalis*, and *Pneumocystis kudriavzevii* (= *Candida krusei*)) and was able to identify 31.2 % of the uncommon yeast species compared to the ITS rDNA sequencing. Compared with PCR-RFLP, all common *Candida* species and 81.8% of uncommon yeast species were correctly identified by the 21-Plex PCR. Vitek 2 system was also successful in identifying 86.7% of all yeast isolates.

Conclusion: Accurate, rapid, and reliable identification of yeast isolates isolated from the bed can help reduce the costs of the health care system and speed up the treatment process for patients. The application of 21-Plex PCR assays for the identification of common and rare yeasts can reduce turnaround times and costs if applied in developing countries such as Iran.

Keywords: 21Plex PCR; PCR-RFLP; Vitek 2 system; Yeast; Identification



Medical parasitology



A Case Report of Oxyuris Cause of Acute Appendicitis in a Child with ALL

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Background and Aim: *Enterobius vermicularis* is the main worldwide intestinal parasites in children. In ectopic migrations, larvae go out of the intestines and go to urethra, vagina and appendix that can have health risks.

Methods: Eight years old boy referred to the oncology clinic of the Tabriz children's hospital, along with complaint of fever, abdominal pain, cramping, anorexia, weakness, cervical lymphadenopathy, hepatomegaly, armpit mass, abdomen thighs, maculopapular rashes and tiny petechial on the chest and abdomen. In the CT scan of the chest, middle mediastinal and mesenteric lymphadenopathy was observed. The patient was hospitalized with a definite diagnosis of acute lymphocytic leukemia (ALL). On the seventh day of hospitalization, the patient getting into increasing colic pain (contraction and cramping) around the navel and simultaneously developing the pain to the down and right of the abdomen. According to the clinical findings, acute appendicitis was considered for patient.

Results: Pathological studies showed that *Enterobius vermicularis* was the cause of appendicitis. The results of this study, it was found that this parasite can be associated with inflammation of the appendix in acute lymphoblastic leukemia patients.

Conclusion: Screening program remains necessary for immunodeficiency patients or those who use immunosuppressive drugs.

Keywords: *Enterobius vermicularis*; Oxyuris, Appendicitis; Acute Lymphocytic Leukemia (ALL)

A clinical Association between Toll-Like Receptor 2 Arg753Gln Polymorphism with Recurrent Cystic Echinococcosis in Post-surgery Patients; a Case Control Study

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Background and Aim: The recurrence of hydatid cysts in cystectomy patients has dramatically remained a serious concern within the surgical community. Predisposing factors for recurrence of hydatid cysts remained to be identified. Toll-like receptor (TLR) plays a pivotal role in bridging between acquired and innate immunity in cystic echinococcosis (CE) infection.

Methods: 117 CE patients including 66 acute hydatidosis (AH; primary infection) and 51 recurrent hydatidosis (RH; chronic infection), and 117 ethnically matched healthy control (HC) were investigated from endemic regions of Iran in the period of 2015 to 2018. CE patients were definitely confirmed using histopathological and immunological assays. Genotyping of TLR2 Arg753Gln was carried out by restriction fragment length polymorphism and sequencing.

Results: The homozygous mutant-type TLR2 Gln/Gln (A/A) was represented to be associated with the occurrence of RH ($P = 0.04$) and conferred a 9-fold risk for susceptibility, while the heterozygous mutant-type TLR2 Arg/Gln (G/A) indicated a tendency to be associated with the occurrence of RH ($P = 0.07$). There was no discrepancy in the frequency of TLR2 Arg753Gln haplotypes between AH patients and HC individuals ($P = 0.09$). The mutant allele A was observed to be a risk factor for susceptibility to RH patients.

Conclusion: Our results point to a clinical association between TLR2 Arg753Gln haplotypes with RH.

Keywords: Recurrence cystic echinococcosis; Toll-like receptor 2; Genetic polymorphism; Unsuccessful postoperative

A Comparaitive Study on the Effect of Metronidazol with Silver, Curcumin Nanoparticles and Chitosan Nanoparticles against- *Blastocystis* sp. *in vitro*.

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Background and Aim: *Blastocystis* is one of the more prevalent intestinal parasites in human, that it's incidence is between 3-100 % in different countries. One of the obligatory anaerobic protozoans live in colon. The transmission of this parasite is most oral-fecal. So far, 17 subtypes of the parasite have been identified, nine of which are prevalent in human, in most epidemiological studies, st3 the most abundant subtype has been found. Metronidazole as a choise drug is considered nowadays. But consumption of this drug has adverse reactions such as: headache, tongue swell, vomiting, metallic taste in the mouth, tongue dryness. Beside several side effects (carcinogenic, teratogenic, and embryogenic effects, e.g.) and drug resistant to this drug is rising. The purpose of this study is a comparative study on the effect of Metronidazole with Silver, Curcumin Nanoparticles and Chitosan Nanoparticles as against-*Blastocystis* sp. *in vitro*.

Methods: After the parasite was prepared from the group archive. They were cultured in Robinson medium. So, PCR method was used directly of culture medium and st3 was determined. After the medications were taken from the company to determine the toxicity level, the MTT assay was done. For this test was used of Caco2 cells. The effect of silver (10,25,50µg/ml), chitosan (12.5,25,50,75 µg/ml) , curcumin (250,500,1000 µg/ml) nanoparticles in different concentrations after 24 and 48 h were studied. The final numbers of parasite determined with a hemocytometer and Trypan blue were recorded. The metronidazole was used as positive controls.

Results: The IC₅₀ and si were calculated for them. The si for chitosan and curcumin nanoparticles are 13.15 and 2.04 were more effective than metronidazole and silver was 0.143. But the best anti parasite effect between these medications had a nano chitosan.

Conclusion: According to this results chitosan, curcumin nanoparticles have a good anti-*Blastocystis* activities versus metronidazole. Therefore, it is recommended to test in animal models.

Keywords: *Blastocystis*; Nanoparticles; silver; chitosan; curcumin; si

A Comparative Study Effects of Aqueous, Ethanolic Extracts and Essential Oil of *Stachys schtschegleevii* on *Trichomonas vaginalis* in vitro

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Background and Aim: Trichomoniasis is a common worldwide sexually transmitted disease caused by the flagellated protozoan *Trichomonas vaginalis*. Metronidazole is the selective drug in trichomoniasis treatment. However, the reported cases show an increasing trend of drug resistance. This study aimed to evaluate the effect of aqueous, ethanolic extracts and essential oil of *Stachys schtschegleevii* on *Trichomonas vaginalis*.

Methods: In this experimental study, the extracts and essential oil of *Stachys schtschegleevii* were prepared, and to evaluate the effect of them on *Trichomonas vaginalis* parasites, the concentrations of 4, 8, 12, 16 and 24 mg/ml were prepared. Metronidazole and TYM medium were considered as positive and negative controls, respectively. Afterward, 105 live parasites were added to all wells, and all groups were kept at 37°C. Live parasites were counted at 24 and 48h intervals by using Trypan Blue and Neubauer slide. Also, the effect of metronidazole (0.5, 1, 2, 4, 8 and 10 µg/ml) was investigated as the drug control. Then the value of IC₅₀ [Half maximal inhibitory concentration] and the lethal percent were calculated using GraphPad Prism 9. It should be noted that all steps of the experiment were performed in triplicate and the results were considered as average.

Results: The IC₅₀ of aqueous, alcoholic extract and essential oil of *Stachys schtschegleevii* on *Trichomonas vaginalis* were calculated at 11.2, 3.67, 2.72 mg/ml after 24h and 5.52, 2.61 and 1.418 mg/ml after 48h, respectively. The IC₅₀ of metronidazole were calculated 0.28 and 0.1 µg/ml after 24h and 48h.

Conclusion: While the aqueous, alcoholic extracts and essential oil of *Stachys schtschegleevii* have anti-*Trichomonas* effect, the anti-*Trichomonas* properties of essential oil is more than its aqueous and alcoholic extract. Further and more comprehensive studies are suggested to investigate the constituents of this plant and the lethal effect of the parasite in vitro and in vivo conditions.

Keywords: *Trichomonas vaginalis*; Alcoholic extract; Aqueous extract; Essential oil; *Stachys schtschegleevii*; IC₅₀

A Comparative Study on the Effect of Metronidazol and Sophora Alopecoides Alcoholic and Aqueous Extract on *Trichomonas vaginalis* In Vitro

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Background and Aim: Protozoan *Trichomonas vaginalis* is a pathogen of human genital tract and the causes of trichomoniasis, which is as one of the most common non-viral sexually transmitted infections in the world. Metronidazole is a selective treatment for trichomoniasis. Remaining of clinical symptoms of patients after metronidazole treatment, confirm the presence of metronidazole-resistant strains. Therefore, finding an alternative herbal drug with less complications and better effect on resistant strains is essential. Nowadays, herbs are used and specifically *Suffera Alopecoreides* due to alkaloids with a positive effect on the improvement of infections in the body.

Methods: To investigate the inhibitory effect of *Suffera Alopecoreides* extract on growth and inhibition of *Trichomonas vaginalis* the effect of roots and leaves of *Suffera Alopecoreides* and the effect of metronidazole on *Trichomonas vaginalis* and macrophage cells of mice were compared and evaluated in vitro. After extracting the plant composition by soaking, the 0.5, 1, 2, 4 and 5 concentrations were prepared. After exposure to the specific number of parasites and 24 and 48 hours after they were colored by trypan blue, the number of parasites were counted. MTT test was used to determine the toxic effect of the extract on peritoneal cells of the mice. The Prism software was used to determine the IC₅₀, CC₅₀ was used.

Results: All four combinations have a good effect on the parasite, but the treatment index with the aqueous extract of the leaf is lower than that of the other extracts, as well as metronidazole.

Conclusion: All four combinations have a good effect on the parasite.

Keywords: *Trichomoniasis vaginalis*; *Suffera Alopecoreides* Extract; Metronidazole

A Retrospective Study on the Dynamics of the Clinical and Demographical Evolution of Cutaneous Leishmaniasis in the Yazd Province, Iran

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Background and Aim: Leishmaniasis is considered as an important health problem in the world. Cutaneous Leishmaniasis is epidemic in all of places of Iran, this disease begins as small swells and then they gradually grow and turn into wounds. Since the epidemiological study of this problem is effective in preventing and controlling it. The current research was conducted to study the epidemiological study of Cutaneous Leishmaniasis in Yazd province during the past 5 (2014-2018) years.

Methods: This cross-sectional study has been conducted on registered information and documents of the patients having problem in Yazd province remedial and health care centers over the past 5 years. First, the registered demographic and epidemiological data of the patients were extracted, and then they were analyzed by using SPSS 18 software with chi-square test and descriptive statistics.

Results: Patients (897) included 457 males (51.06%) and 439 females (48.94%). The majority of the patients were between 20-29 years old and the minority of them were the range of 5-9 years old. Most lesions were on the hand and leg of the patients. It was also found that there was a significant relationship between age groups and types of Leishmaniasis (P-value less than 0.05).

Conclusion: According to our study, there has been an ascending trend in disease frequency in Yazd province from 2014 to 2018. This increase in diseases due to lack of health and educational status of the people is also a lack of personal hygiene.

Keywords: Frequency; Cutaneous Leishmaniasis; Epidemiology; Yazd

An Epidemiological Survey of Hydatid Cyst Infection in Slaughterhouse of Nahavand City during 2014_2017

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Background and Aim: Hydatid cyst is a common disease in humans and animals that exists all over the world and is also found in our country. Hydatidosis in livestock leads to significant economic losses to the livestock industry and is one of the public health problems in many countries, especially developing countries. The aim of this study was to determine the frequency of hydatid cyst disease in slaughtered animals in Nahavand abattoir in Hamadan province during 2014-2017 that due to the lack of history of investigating this issue in the city, it can help the authorities in making appropriate decisions to plan, control and prevent the disease that is transmitted to humans through livestock.

Methods: This is a descriptive cross-sectional study. The statistical population was cattle, sheep and goats slaughtered in Nahavand slaughterhouse during the years 2014 to 2017. The obtained data were extracted after reviewing the data entered in the slaughterhouse registration file for 4 years and then analyzed by SPSS software version 22.

Results: A total of 145090 cattle were slaughtered, of which 108422 sheep and goats (74.7%) and 36668 cattle and buffaloes (25.3%). Hydatid cyst was observed in 9429 (6.49%) cattle. The prevalence of hydatid cyst in sheep and goats was 6.49% (7044 heads) and in cattle and buffaloes was 6.50% (2385 heads) which showed that it was higher among cows. Also, in the studied samples, most of the infections were related to lungs with 4.10% (5959 heads) and liver infections with 2.39% (3470 heads).

Conclusion: Among the factors of hydatid cyst in the slaughtered animals in Nahavand are unsanitary slaughtering, the presence of stray dogs and herding dogs without health control and supervision.

Keywords: Hydatid cyst; Epidemiological; Nahavand; Slaughterhouse

Anti-protozoal Activity of Magnesium Oxide (MgO) and Calcium Oxide (CaO) Nanoparticles against *Leishmania major* in vitro

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Background and Aim: Leishmaniasis is a parasitic infection that is more common in the tropical and subtropical regions. The pentavalent antimonial compounds are the first drug of choice for *Leishmania* infection, but have several side effects that cause some restriction for use. The current study was aimed to evaluate the effect of magnesium oxide (MgO) and calcium oxide (CaO) *Leishmania major* in vitro.

Methods: To evaluate the antileishmanial activity of nanoparticles (NPs), light microscopic observation was used to determine the number of remaining parasites in each well. In this study, *Leishmania major* promastigotes were exposed to MgO and CaO NPs with different doses ranging from 2.5-40 mg/ml. Then MTT test was used to determine the cytotoxicity effects of MgO and CaO NPs against *Leishmania major*. Moreover, lethal effects of nanoparticles were tested for macrophage cell with MTT assay.

Results: At high concentrations, two nanoparticles had a more lethal effect on parasites. According to the results of MTT assay, the nanoparticles via MgO and CaO NPs led to the death of parasite cells. High concentrations of nanoparticles led to the destruction of macrophages so that less of 20%.

Conclusion: Based on the above assessment, we determined that the MgO NP has a more lethal effect on promastigotes than CaO NP and maybe in the future that be used as a drug candidate.

Keywords: *Leishmania major*; Magnesium oxide; Calcium oxide; In-vitro

Antiparasitic Effect of *Rheum ribes* L. Extract Against the Hydatid Cyst Protoscoleces

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Background and Aim: Cystic echinococcosis is a zoonotic infection in humans and herbivorous animals occurring worldwide, which is caused by the larva stage of *Echinococcus granulosus*. Rhubarb (*Rheum ribes* L.) as an herbal medicine has various therapeutic properties such as antioxidant, anticancer, and antimicrobial. With respect to the potential of the biological activities of this plant in traditional and modern medicine, we aim to examine its protoscolicidal effects against *Echinococcus granulosus* protoscoleces in vitro and ex vivo.

Methods: Collected protoscoleces from liver hydatid cysts of infected sheep were exposed to the different concentrations of the extract (225, 450, 900 mg/mL) for 5-60 min in vitro and ex vivo. Then, by using the eosin exclusion assay, the viability of protoscoleces was studied.

Results: *R. ribes* extract had a potent protoscolicidal activity *in vitro*; at 450 and 900 mg/mL, it killed 56.3 and 100% of protoscoleces, respectively, after 10 min of exposure. In ex vivo assay, the extract needed more time to kill the protoscoleces than the in vitro; so that at the concentration of 900 mg/mL, all protoscoleces were killed after 15 minutes.

Conclusion: The obtained results exhibited the potent protoscolicidal effects of *R. ribes* extract, particularly at the concentration of 900 mg/mL, which completely killed the parasite less than 15 min of exposure. However, more supplementary studies are required to verify these findings by assessing animal models and clinical subjects.

Keywords: *Rheum ribes*; Hydatid Cyst protoscoleces; *Echinococcus granulosus*

Antiparasitic effects of Fennel methanolic extract against hydatid cyst protoscoleces

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Background and Aim: Hydatidosis (hydatid disease) which caused by the larval form of dog tape worm of *Echinococcus granulosus sensu lato* (s.l.) is as one of the most important neglected parasitic diseases globally. Here we determine the effect of Fennel (*Foeniculum vulgare* Mill.) methanolic extract on *Echinococcus granulosus* protoscoleces *in vitro* condition.

Methods: *Echinococcus granulosus* protoscoleces were obtained from the hydatid cysts of domestic livestock. Various concentrations of Fennel extract (25 - 200 mg/ml) and were applied for 10, 20, 30, and 60 minutes. Finally, the viability of hyatid cyst protoscoleces was assessed by Eosin test.

Results: The obtained results revealed that Fennel extract had remarkable protoscolicidal effects on hydatid cysts protoscoleces; so that at the concentration of 200 mg/ml after 10 min of killed completely all protoscoleces. Whereas, after 20 minutes incubation to concentration of 100 mg/ml the mortality mean of protoscoleces was 100%.

Conclusion: The findings showed that Fennel extract had potent protoscolicidal effects against hydatid cyst protoscoleces indicating the potential of Fennel to produce a new scolicial drug to apply in hydatid cyst surgery.

Keywords: Fennel; protoscoleces; Cystic echinococcosis; *in vitro*

Association of Human Leucocyte Antigen (HLA) Class I and II Genes with Cutaneous and Visceral Leishmaniasis in Iranian Patients: A Preliminary Case-Control Study

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Background and Aim: *Leishmania* spp. are a parasite that can be transmitted by sand fly, causing human leishmaniasis infection. The major forms of leishmaniasis are the diffuse cutaneous leishmaniasis, localized cutaneous leishmaniasis, as well as VL, which is the most lethal and the severest type of leishmaniasis that affects internal organs. The human leukocyte antigen (HLA) system is an important constituent of the immune system and is considered as a contributory genetic factor for leishmaniasis. This system is responsible for encoding cell surface molecules specialized to present antigenic peptides to CD4⁺ and CD8⁺ T cells. CD4⁺ T cells have the ability to control *Leishmania* replication at the early stages of infection, while CD8⁺ T cells are involved in IFN- γ production and Th1 differentiation. Therefore, alterations in HLA I & II due to variability of *Leishmania* antigens can affect the susceptibility of humans to leishmaniasis. Identification of a link between particular HLA alleles and susceptibility or resistance to this parasitic infection not only can help us to understand the complex pathogenicity of *Leishmania* and the host immunity molecular pathways but also can help in the development of epitope-based drugs or vaccines. The present study aimed at the identification of distinct HLA-I & II alleles involved in the susceptibility or resistance to VL and CL infections in Iranian populations.

Methods: This study was approved in the Tehran University of Medical Sciences Ethical Committee (IR.TUMS.SPH.REC.1399.210). This case control study was carried out on 31 leishmaniasis patients between March-October 2020. Blood samples were collected from 20 CL patients referring to the leishmaniasis laboratory, the School of Public Health, Tehran University of Medical Sciences and 11 VL patients residing in northwest of Iran. Written consent was taken from all patient. For CL samples, parasitological evaluation was based on the observation of amastigotes indirect smear. CL patients were categorized as healing (those who were showed CL infection for the first time, those who had self-healing CL, and those who recovered from CL after treatment) and non-healing (those with infection recurrence, treatment failure, drug resistance) cases. Direct agglutination testing (DAT) was carried out for the diagnosis of VL infection. Accordingly, VL patients were categorized as symptomatic (DAT titer at 1:3200 and above) and asymptomatic (DAT titer at 1:800 greater than or equal) cases. Control group included those living in the same area as the patients (preferably from

family members or relatives of the patients). Exclusion criteria for the control group included the history of CL infection.

Results: In the present study, a total of 20 CL patients were recorded among which 10 cases showed healing CL and 10 cases had non-healing CL. A total of 11 VL cases were recorded from the endemic region for Kala Azar. Among these cases, 5 patients showed asymptomatic VL infection, while 6 demonstrated symptomatic VL infection. Distribution of HLA alleles: No significant association was observed between age and sex of the patients with infection type (P -value > 0.05). According to the results of multinomial logistic regression, CL susceptibility in patients with A26 alleles was significantly greater than those who did not carry this allele. Statistical analysis indicated a significant association between the infection type and the presence of C1 allele (at CI: 95%). Also, susceptibility to symptomatic VL was significantly higher in those carrying the C1 allele. The results of the Fischer Exact Test indicated the significant association between infection type and the presence of B1.4 alleles. The results of the multinomial logistic regression indicate a strong relationship between the presence of B1.4 alleles and susceptibility to asymptomatic VL.

Conclusion: In this study, HLA-A26, HLA-B38, HLA-C1, and HLA-B1.4 of HLA-I alleles have been suggested to be involved in susceptibility to leishmaniasis, confirming the role of HLA in these infections. These results highlight the importance of patient screening in endemic regions for the identification of host factors involved in susceptibility or resistance to leishmaniasis, which can be employed for the development of effective vaccines for endemic areas. Further studies are required to confirm the association between these alleles and VL infection.

Keywords: Leishmaniasis; Immunology; HLA; genes; Iran.

Can Free-living *Acanthamoeba* Act as a Trojan Horse for SARS-CoV-2 in Viral Survival and Transmission in the Environment?

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Background and Aim: *Acanthamoeba* is a free-living amoeba that has been found on surfaces, air, water and various environmental sources around the world. It enters the human body through the respiratory tract via the nose. Of note, amoebae are well known to act as a potential reservoir for various pathogenic microorganisms including bacteria and viruses such as Adenoviruses and Mimivirus. Given that SARS-CoV-2 of the Coronaviridae family is transmitted through the respiratory tract, and the Trojan horse nature of *Acanthamoeba*, it has been suggested that amoebae act as a vector in the transmission of SARS-CoV-2. The aim of this study was to provide a mini-review of the available literature of coexistence of *Acanthamoeba* with SARS-CoV-2.

Methods: The Preferred Reporting Items for Systematic mini-reviews (PRISMA) guidelines was followed for this systematic mini-review (fig2). To determine the endosymbiont of SARS-CoV-2 with *Acanthamoeba* a literature search was carried out ranging from 1 January 2019 to 3 June 2021. The literature was searched by using a combination of keywords as follows: “Coronavirus” OR “SARS-CoV-2” OR “COVID-19” AND “Acanthamoeba” OR “Free-Living Amoebae” in English language. This was complemented by using the check of references of the published articles were to access to for their related studies. For an article to be included in this study, it had to be peer-reviewed, available in full text, with its primary objective to isolate and identify SARS-CoV-2 with *Acanthamoeba*. However, case reports of *Acanthamoeba* with SARS-CoV-2 were included. A systematic mini-review for all of the selected studies. Articles that were published in languages other than English, institutional protocols, other review papers, or other microorganisms for the analysis of symbiosis and isolation of SARS-CoV-2 from amoeba other than *Acanthamoeba* were excluded from the study.

Results: A total of 197 articles were generated by conducting systematic search within 2019-2021. Taking duplication into account, 178 records were excluded, and 6 articles met the inclusion criteria. Thereafter, 1 record was excluded through an irrelevant in include articles. 2 full-text articles were assessed.

Conclusion: *Acanthamoeba* have been found to phagocytosis fungi, bacteria, and viruses and serve as a repository for these endosymbionts. This mini-review of the literature found five peer-reviewed



studies discussing the interactions of *Acanthamoeba* and SARS-CoV-2 of No studies have documented SARS-CoV-2 as an endosymbiont, but the literature suggests it could be possible due to the size of the virus, similar modes of infection, ubiquitous environmental presence of both organisms, and documented infections of hosts via *Acanthamoeba* with other viruses. There is a significant amount of research still needed to understand the interactions between *Acanthamoeba* and SARS-CoV-2.

Keywords: SARS-CoV-2; *Acanthamoeba*; mini-review



Contamination of Fresh Vegetables with Pathogenic *Acanthamoeba* Genotypes (T4, T5 and T9) in Iran

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Background and Aim: *Acanthamoeba* spp. can cause amoebic keratitis (AK) and encephalitis and may act as a proper carrier of foodborne pathogens. The aim of this study is to estimate the presence of *Acanthamoeba* and its genotypes in commonly consumed fresh vegetables.

Methods: A total of 70 samples including garden cress (22), chives (12), mint (13), parsley (10), and basil (13) were collected from grocery store in Tehran province. Vegetables were washed, the water was filtered through 0.45 µm nitrocellulose filter, and *Acanthamoeba* cultured onto 2% non-nutrient agar (NNA) medium. Cultures were analyzed using morphological examination and for the molecular identification the ASA1 region of the 18S rRNA gene (stem 29-1 of 18S rRNA gene) was targeted to characterize genotypes of the harvested specimens. Pathogenic potential of the isolated strains was also analyzed using biological assays.

Results: In total, 18 (25.7%) out of 70 tested vegetable samples in NNA were positive for *Acanthamoeba* spp. based on the morphology criteria. Samples including garden cress 10/22 (45.45%), chives 3/12 (25%), mint 2/13 (15.38%), basil 2/13 (15.38%) and parsley 1/10 (10%) were positive. The diagnostic fragment 3 was successfully sequenced in 15 samples. The genotype T4 was characterized among 11 (73.3%) samples. Three strains belonged to the T5 genotype corresponding to *A. lenticulata* and one isolate showed high identity and query coverage with T9 genotype.

Conclusion: Pathogenic bioassays showed three, two, and one strains belonging to the genotypes T4, T5, and T9 which are ranked highly pathogenic. The present study is the first to show contamination of the most commonly consumed fresh vegetables with pathogenic *Acanthamoeba* genotypes.

Keywords: *Acanthamoeba* spp.; Genotypes; Raw vegetables; Iran

Covid-19 Infection and Cystic Echinococcosis

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Background and Aim: The occurrence of Covid-19 infection in patients with hydatid cyst and its complications is a matter for consideration that has occupied the minds of clinicians, virologists, and parasitologists. Some evidence suggests that the two diseases do not occur together. So, the question is, is it possible for hydatidosis to prevent Covid-19 infection?

Methods: Cases of co-infection of Covid-19 and malaria, as well as Covid-19 with different types of respiratory pathogens such as different types of bacteria, fungi, and viruses, have also been reported in some scientific journals. There are two possibilities: either there is an interaction between hydatid cyst antigens and Coronavirus proteins. Or the use of albendazole by patients with hydatid cysts has protected them against Covid-19 infection.

Results: In the case of co-infection of worms and Covid-19, some reports suggest that the resulting immune responses protect the host against Covid-19 infection. Studies have shown that co-infection of mice with *Schistosoma mansoni* and Coronavirus reduces the severity of Covid-19 infection.

Conclusion: This article discusses the occurrence of Covid-19 infection in patients with cystic echinococcosis.

Keywords: Covid-19 infection; cystic echinococcosis

COVID-19-associated Opportunistic Infections: a Snapshot

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Background and Aim: Immunosuppressive therapy has shown promising results for control of the cytokine storm syndrome (CSS) in severe cases of COVID-19. However, immunosuppressive agents could increase the risk of opportunistic infections by mitigating the immune responses. A number of opportunistic infections have been reported among COVID-19 patients, which most of them received immunosuppressive therapies. This study is aimed to summarize the main opportunistic pathogens that reported among COVID-19 patients.

Methods: A systematic review was performed on the case reports, case series, or cross-sectional studies regarding the reported opportunistic pathogens among COVID-19 patients.

Results: The most reported opportunistic pathogens were *Aspergillus* spp., *Candida* spp., *Cryptococcus neoformans*, *Pneumocystis jiroveci* (carinii), mucormycosis, Cytomegalovirus (CMV), Herpes simplex virus (HSV), *Strongyloides stercoralis*, *Mycobacterium tuberculosis*, and *Toxoplasma gondii*. Most patients received immunosuppressive agents (e.g., corticosteroids and cytokine blockers).

Conclusion: The COVID-19 patients with underlying diseases or who received immunosuppressive therapy have an increased risk of getting opportunistic pathogens. Strategies for screening and early diagnosis, treatment, and prophylaxis could mitigate the risk of these infections among COVID-19 patients. As such, standard prophylactic strategies are needed in high-risk COVID-19 patients, such as those who received immunosuppressive therapy, patients with underlying diseases, patients who admitted to the ICU, and those who received invasive mechanical ventilation.

Keywords: COVID-19; immunosuppressive therapy; opportunistic pathogens

Detection of *Toxoplasma gondii* in Solid Cancer Patients Using the loop-mediated isothermal amplification (LAMP) and Nested PCR Assay

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Background and Aim: *Toxoplasma gondii* (*T. gondii*) is an obligate intracellular protozoan parasite that exists worldwide. Various techniques have been developed for *T. gondii* detection. The aim of current study was determination of the prevalence of *T. gondii* infection in patient with solid cancer using ELISA and two different molecular tests. This study was carried out on patient with cancer in Ali Ibn Abitaleb hospital in Zahedan cities, Southeast of Iran.

Methods: Totally, 154 serum samples from patient with cancer were test for detection of IgG and IgM antibodies against *T. gondii* by ELISA test. All of the sample were also test by Loop amplification (LAMP) and Nested-PCR with RE gene. The obtained results were analyzed using Kappa statistical test and SPSS 22 software.

Results: Totally, 25.3% of the samples from the patient with cancer were positive for IgG against *T. gondii*. After using Nested-PCR technique, 31.1% samples were positively based on RE (Repetitive Element) gene. After using LAMP technique, 33.7% of the cancer samples were found to be positive. The sensitivity and specificity of LAMP method compared to Nested-PCR for the diagnosis of toxoplasmosis in patient with cancer was calculated as a 97.9% (99.9% - 88.9%: 95% CI) and 95.2% (98.2% - 99.2%: 95% CI), respectively.

Conclusion: Based on our result, the RE-LAMP technique was more specific and sensitive than RE-nested-PCR method in the detection of *T. gondii* DNA. In comparison with the RE-nested PCR, RE-LAMP could detect more *T. gondii* DNA in seronegative blood sample.

Keywords: *Toxoplasma gondii*; LAMP assay; Nested PCR; RE gene

Determination of *Enterobius vermicularis* Infection in Children 2-12 years old in Sabzevar During 2020

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Background and Aim: Enterobiasis is a global disease that usually causes itching around the anus and neurological complications in children. The prevalence of this infection has been reported differently in different parts of Iran. The aim of this study was to determine the frequency of disease in children aged 2 to 12 years in Sabzevar.

Methods: In order to carry out this project, 250 children were sampled in clusters in coordination with the children's parents. The samples were tested in the parasitology laboratory of the medical school using the specific Scotch test method and at the end, the data and results were analyzed by statistical tests.

Results: In this study, 250 children were included in the study. The mean age of the subjects was 6.82 ± 2.87 years (2 to 12 years) and also out of 250 children included in the study, 149 (59.6%) were boys and 101 (40.4%) They were girls. Of the 250 children surveyed, 22 (8.8%) had a positive result. The findings of our study showed that *Enterobius vermicularis* infection was not associated with age, gender, mother's education, mother's job, place of residence and hand washing, but was significantly associated with anal itching, gritted of teeth, mood swings, sleep disturbance and nail biting.

Conclusion: Improving the level of living standards from an economic and cultural point of view as well as increasing public health awareness is probably one of the important and effective factors in reducing the prevalence of infection in children in this region. In children with symptoms, it is better to perform laboratory tests regardless of the social and economic situation of the family, and if positive, appropriate treatment for patients as soon as possible to increase the quality of life of children, especially their sleep

Keywords: *Enterobius vermicularis*; Prevalence; Children

Determination of the Immune Status against Toxoplasmosis and Its Related Factors in Newly Married Women in Joveyn City in 1400

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Background and Aim: Toxoplasmosis is one of the most common causes of parasitic infections in humans and other warm-blooded vertebrates that has a worldwide spread. The aim of this study was the determination of immune status against toxoplasmosis and its related factors in newly married women in Joveyn city (Razavi Khorasan) in 1400.

Methods: In this descriptive cross-sectional study, 85 women were randomly selected and after completing a questionnaire, they were examined for the presence of IgG antibodies against *Toxoplasma gondii* by ELISA. The results were analyzed using SPSS statistical test.

Results: According to the kit protocol, 81.18% of the subjects were negative for anti-*Toxoplasma* IgG antibodies and 18.82% were positive. The highest prevalence of positive individuals was in women aged 30 to 35 years and the lowest prevalence was in women under 20 years ($P=0.03$). There was a statistically significant relationship between different age groups, contact with garden soil or agricultural land, consumption of undercooked (grilled) meat and slicing of meat by injured hand and contact with cats and *Toxoplasma* infection.

Conclusion: Due to the high percentage of IgG negative individuals, *Toxoplasma* screening test is recommended before pregnancy. The health authorities of the province, while formulating educational programs and including this serious issue, should take urgent action to raise the awareness of women.

Keywords: Toxoplasmosis; women; Anti *Toxoplasma* IgG

Development of HRM Real-Time PCR for Assemblage Characterization of *Giardia lamblia*

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Background and Aim: A total of 90 stool samples were collected from dogs, referred to a dog shelter and a veterinary clinic. In addition, 395 stool samples obtained from pet dog owners and shelter keepers, as well as individuals referred to a medical laboratory as controls, were collected in Shahryar district, Tehran, Iran. Stool samples were parasitologically examined and the positive *G. lamblia* isolates were tested with Nested-PCR/sequencing for the *tpi*, *gdh*, and *bg* genes, and HRM real-time PCR. Microscopical examination revealed 20 (22.2%) and 34 (8.6%) *Giardia*-positive samples from dogs and humans, respectively. Regarding HRM real-time PCR, the prevalence of assemblages A and B in humans was 55.8% and 14.7%, respectively. In addition, 14.7% of samples were mix assemblages. HRM real-time PCR detected most of microscopically-positive samples in comparison to PCR/sequencing in both humans and dogs. The high prevalence of assemblages A and B in dogs signified the importance of a same source for infection between dogs and humans.

Methods: A Total of 90 fecal samples were acquired from a dog's shelter and a veterinary clinic in Shahriar County, Tehran, Iran. Totally 395 stools samples were also isolated from two medical laboratories. All fecal samples were examined microscopically by using wet mount slide, formalin ether concentration, trichrome staining and Modified Ziehl-Neelsen methods. Genomic DNA (gDNA) was extracted directly from preserved *Giardia*-positive feces using DNA stool Mini Kit (Yekta Tajhiz, Iran) according to the manufacturer's instructions. The extracted DNAs were stored at -20 °C until use. To determine the species and genotype of *G. lamblia*, the extracted DNA was subjected to a Nested-PCR protocol that targets 532 bp in triose phosphate isomerase (*tpi*), 530 bp in glutamate dehydrogenase (*gdh*), and 511 bp in beta giardin (*bg*) and finally some of the *Giardia*-positive PCR isolates were sequenced. Specific HRM primers were designed to amplify *gdh* gene fragment with different melting temperatures for four assemblage of *G. lamblia* (A, B, C, D). To evaluate sensitivity of the test, serial dilutions of a parasite *Giardia* was prepared from ~ 10⁶ to 1 parasite per mL of stool sample and all positive samples were investigated by HRM real-time PCR.

Results: A total of 90 stool samples were collected from dogs, referred to a dog shelter and a veterinary clinic. In addition, 395 stool samples obtained from pet dog owners and shelter keepers, as well as individuals referred to a medical laboratory as controls, were collected in Shahryar district, Tehran, Iran. Stool samples were parasitologically examined and the positive *G. lamblia* isolates were tested with Nested-PCR/sequencing for the *tpi*, *gdh*, and *bg* genes, and HRM real-time PCR.

Microscopical examination revealed 20 (22.2%) and 34 (8.6%) Giardia-positive samples from dogs and humans, respectively. Regarding HRM real-time PCR, the prevalence of assemblages A and B in humans was 55.8% and 14.7%, respectively. In addition, 14.7% of samples were mix assemblages. HRM real-time PCR detected most of microscopically-positive samples in comparison to PCR/sequencing in both humans and dogs. The high prevalence of assemblages A and B in dogs signified the importance of a same source for infection between dogs and humans.

Conclusion: Our results showed the presence of *G. lamblia* in both humans and dogs with higher prevalence in dogs. Furthermore, assemblages A and B were prevalent in both humans and dogs. The presence of assemblages A and B in both humans and dogs suggests a probable transmission cycle between humans and animals, particularly dogs. Our study also showed that real-time PCR coupled with HRM was not only detected more microscopically positive samples in comparison to conventional PCR/ sequencing, but also characterized assemblages A and B in dogs, and mix assemblages in both humans and dogs. Therefore, considering the higher sensitivity of HRM real-time PCR due to the small size of the targeted fragment, it seems that this technique could be a suitable technique to simultaneously detect and genotype Giardia in clinical and epidemiological practices.

Keywords: Giardia, Assemblage; Humans; Dog; HRM real-time PCR; PCR/sequencing

Diagnostic accuracy of parasitological method compared to molecular method (PCR) in detection of microsporidia in HIV+/ AIDS in Kurdistan province

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Background and Aim: Microsporidiosis is one of the parasitic diseases that also is an opportunistic infection. In people with a normal immune system, intestinal microsporidia are usually seen as a transient infection, but in individuals with immune system defects, these microorganisms can cause severe digestive infections and, in some cases, cause even published infections, which in These conditions may even contaminate the whole body of the patient. The aim of this study was to investigate the diagnostic value of the parasitological method in comparison with the molecular method (PCR) in the diagnosis of intestinal microsporidiosis in HIV + / AIDS patients in Kurdistan province in 1399.

Methods: This study was performed on stool samples collected from 161 patients with HIV + / AIDS, and the results of the samples of patients were recorded in tables and information forms and analyzed using SPSS software Statistical analysis.

Results: One hundred sixty-one patients were enrolled in the study. The mean age of the patients was about 41 years in the age range (12-66 years). It was 93.79% male. In the parasitology method, 8 cases (4.97%) were positive in the PCR method (6.21%) were positive for intestinal *Microsporidia*. Sensitivity 60% and 98.9% characteristics were obtained for the parasitological method.

Conclusion: The results of this study showed that the contamination rate of intestinal *Microsporidia*, unlike previous studies in Iran, has significantly decreased, which can be due to the different populations and the geographical area that has been studied. This was the first study to look at infections caused by Intestinal *Microsporidia* in people with HIV / AIDS and Using cellular and molecular methods (PCR) and sequencing in Kurdistan province and showed gave the presence of intestinal *Microsporidia* in this group of people.

Keywords: Parasitological method; PCR; *Microsporidia*; HIV + / AIDS

Effect of Tellurium Oxide Nanoparticle on *Toxoplasma gondii* invitro Condition

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Background and Aim: *Toxoplasma gondii* is an obligate intracellular parasite in humans and many warm-blooded animals in the world. Objective: In this study, we will investigate the different antiparasitic effects of tellurium oxide nanoparticles against *Toxoplasma gondii* (TeO₂).

Methods: In this study, we used tachyzoite assay (ocular counting test) to determine the lethal effect of tellurium nanoparticles on parasites. Toxic effects of nanoparticles on macrophage cells were also evaluated using MTT assay. The type of parasitic cell death (necrosis or apoptosis) was determined by flow cytometric analysis and its cause was measured by that's the device.

Results: According to the examination of the parasite by light microscopy, it was found that tellurium nanoparticles have destructive and lethal effects on *Toxoplasma*. The results of the MTT test showed that at high concentrations (such as 400 µg / ml) the toxic effects on macrophage cells were greater. Flow cytometric analysis also estimated the percentage of apoptosis (primary and secondary) of parasites at 56.97%.

Conclusion: Based on the above out comes, it is determined that tellurium oxide nanoparticles have a toxic effect on *Toxoplasma gondii* and led to apoptosis in this parasite. This research boosts different tests in another parasite.

Keywords: Tachyzoite; *Toxoplasma gondii*; Tellurium oxide Nanoparticles; Flow Cytometry; MTT assay

Effects of Negative Air Ions (NAIs) on the Lesions of Cutaneous Leishmaniasis in *in vitro* and in BALB/c Mice

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Background and Aim: Despite the remarkable advancement in the cutaneous leishmaniasis (CL) treatment, available compounds and tools could not overcome distinct obstacles including parasite/host complicated interactions, and also could not decrease the serious side effects. Negative air ions (NAIs) generators are widely available for home or industrial uses. Studies have been reported that NAIs had various positive effects on human health and reduced distinct harmful microorganism's load; but no studies have examined their effects against *Leishmania* parasite. The aims of this study were to investigate the effect of NAIs therapy on the *Leishmania* major (L. major) the causative agent of CL in in-vitro and in mice model.

Methods: In-vitro anti-leishmanial effects of NAIs therapy were measured applying direct parasitological methods. For the in-vivo study, the footpad lesion size and parasite load in L. major infected BALB/c mice were quantified by using the metric caliper tool and qPCR, respectively. For BALB/c mice immune responses evaluation, the levels of IFN- γ , IL-4, NO level, and arginase activity were measured.

Results: NAIs therapy significantly decreased the viability of *Leishmania* parasite promastigotes and amastigotes form of L. major without any negative toxicity to the host cells. The footpad lesion size, *Leishmania* parasite load, IL-4, and arginase activity were decreased in the presence of NAIs; whilst IFN- γ and NO were significantly raised. NAIs therapy significantly could influence infection progression in contrast to control groups but is less effective than the amphotericin B treatment group.

Conclusion: We established satisfactory findings in L. major parasite clearing both in-vitro and BALB/c mice. NAIs therapy effectively increases host cellular immunity and induced T-helper 1 (TH1) responses and restricted *Leishmania* parasite burden. Therefore, NAIs therapy can be considered as a useful tool for clearing the L. major without any negative toxicity to the host cells. The applications and mechanisms of NAIs therapy warrant further investigation.

Keywords: Negative air ions (NAIs); *Leishmania major*; Leishmaniasis; BALB/c; T-helper 1 (TH1); Immune responses

Effects of Silver Nanoparticles via Ginger Rhizome Extract on *Leishmania infantum*: An *invitro* study

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Background and Aim: Leishmaniasis is a parasitic infection that is more common in the tropical and subtropical regions. According to the World Health Organization report, leishmaniasis is considered as a major global health issue in more continents. The current study was aimed to evaluate the effect of silver nanoparticles via Ginger rhizome extract on *Leishmania infantum* *in vitro*.

Methods: Different concentrations of silver nanoparticles via Ginger rhizome extract promastigotes were investigated. We used promastigote assay and MTT methods to evaluate the toxicity and lethal effects of nanoparticles on *Leishmania infantum* promastigotes. Also, lethal effects of nanoparticles were tested for macrophage cell with MTT assay.

Results: At high concentrations, nanoparticles had a more lethal effect on parasites. According to the results of MTT assay, the nanoparticles via Ginger rhizome extract led to the death of parasite cells. The IC₅₀ of nanoparticles via Ginger rhizome extract were determined as 16.8 ppm according to MTT assay. High concentrations of nanoparticles led to the destruction of macrophages so that 7.5 % of the cells killed at 40 ppm concentration of nanoparticles.

Conclusion: Regarding the above results, it can be concluded that silver nanoparticles via Ginger rhizome extract has a fatality effect on *Leishmania infantum* promastigotes *in vitro*.

Keywords: *Leishmania infantum*; Ginger rhizome; *in vitro*

Effects of Silver Nanoparticles via *Thymus vulgaris* Extract on *Leishmania infantum* *in vitro*

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Background and Aim: Leishmaniasis is a parasitic infection that is more common in the tropical and subtropical regions. According to the World Health Organization report, leishmaniasis is considered as a major global health issue in more continents. The current study was aimed to evaluate the effect of silver nanoparticles via *Thymus vulgaris* extract on *Leishmania infantum* *in vitro*.

Methods: Different concentrations of silver nanoparticles via *Thymus vulgaris* extract promastigotes were investigated. We used promastigote assay and MTT methods to evaluate the toxicity and lethal effects of nanoparticles on *Leishmania infantum* promastigotes. Also, lethal effects of nanoparticles were tested for macrophage cell with MTT assay.

Results: At high concentrations, nanoparticles had a more lethal effect on parasites. According to the results of MTT assay, the nanoparticles via *Thymus vulgaris* extract led to the death of parasite cells. The IC₅₀ of nanoparticles via *Thymus vulgaris* extract were determined as 26.6 ppm according to MTT assay. High concentrations of nanoparticles led to the destruction of macrophages so that less of 10% of the cells survived at 50 ppm concentration of nanoparticles.

Conclusion: Regarding the above results, it can be concluded that silver nanoparticles via *Thymus vulgaris* extract has a fatality effect on *Leishmania infantum* promastigotes *in vitro*.

Keywords: *Leishmania infantum*; *Thymus vulgaris*; *in vitro*



Effects of Thymoquinone on *Leishmania infantum* in vitro

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Background and Aim: Leishmaniasis is a parasitic infection that is more common in the tropical and subtropical regions. According to the World Health Organization report, leishmaniasis is considered as a major global health issue in more continents. The current study was aimed to evaluate the effect of Thymoquinone on *Leishmania infantum* in vitro.

Methods: With different concentrations of Thymoquinone, promastigotes were investigated. We used promastigote assay and MTT methods to evaluate the toxicity and lethal effects of Thymoquinone on *Leishmania infantum* promastigotes.

Results: At high concentrations, Thymoquinone had more lethal effect on parasites. According to the results of MTT assay, the Thymoquinone led to the death of parasite cells. The IC₅₀ of Thymoquinone were determined as 9.46 µM according to promastigote assay.

Conclusion: Regarding the above results, it can be concluded that Thymoquinone has a fatality effect on *Leishmania infantum* promastigotes in vitro.

Keywords: *Leishmania infantum*; Thymoquinone; in vitro

Archives of Medical Laboratory Sciences

Evaluation of Effects of Hydro-alcoholic Extracts of *Colchicum autumnale*, *Chamomile* and *Tanacetum parthenium* on *Leishmania Major*

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Background and Aim: Leishmaniasis is a complex disease with a broad spectrum of clinical features, usually divided into Cutaneous Leishmaniasis (CL), Muco-cutaneous Leishmaniasis (MCL), and Visceral Leishmaniasis (VL). Plant extracts or plant-derived compounds are likely to provide a valuable source of new medicinal agents. In endemic countries, a number of traditional plants are commonly used to treat infectious conditions. Advances in the research of natural products for the treatment of leishmaniasis have been recently reviewed. The aim of this study is to evaluate the leishmanicidal activity of three plants hydro-alcoholic extracts of *Colchicum Autumnale*, *Chamomile* and *Tanacetum Parthenium* on *Leishmania Major* (MRHO/IR/75/ER) promastigotes with compared to Glucantime, in vitro.

Methods: According to the aim of study, extracts of each plant were experimentally provided in concentrations of 800, 400, 200, 100, 50, 25, 12/5, 6/25 and 3/12 mg per ml. Then, these concentrations were inoculated in containing parasites and in terms of the number of parasites at 24, 48 and 72 hours were analyzed.

Results: These results showed that all three extracts only in high concentrations makes inactivity of all of *Leishmania* parasite, and decreasing the concentration of the extracts increased the number of parasites alive. Furthermore, Glucantime was more effective in reducing the number of *Leishmania* than *Colchicum Autumnale* and the difference was significant. Also more likely *Chamomile* and *Tanacetum parthenium* had similar impact and the differences were not significant.

Conclusion: These three plants extracts significantly are effective on activity of *Leishmania*, although suggested that more invitro and *in vivo* studies to demonstrate the effectiveness of these extracts are required.

Keywords: *Leishmania major*; *Colchicum autumnale*; *Chamomile*; *Tanacetum parthenium*

Evaluation of Anti-leishmanial Effects of Hydro-alcoholic Matthiola Plant Extract *in vitro*

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Background and Aim: Studies have shown that the hydroalcoholic extract of Matthiola plant has antibacterial and antifungal effects such as gram-positive and gram-negative bacteria also *Candida albicans*. On the other hand, the problems of treatment of cutaneous leishmaniasis, including their injection use such as glucantime and their side effects, led to the design of this study.

Methods: First, after preparation and approval of the plant by Herbarium, Medicinal Plants Research Center, hydroalcoholic extract was prepared by maceration method. Then the standard species of *Leishmania major* (MRHO / IR / 75 / ER) was cultured in NNN and RPMI enriched media. After that, concentrations of 6.5, 12.5, 50, 100, 400, 800 mg / ml of the extract were prepared and the *Leishmania* parasite was affected. Survival rate of parasites was assessed by MTT test. The effectiveness of the extract has been reported as IC50 and F90 Indicators.

Results: The mean light absorption in all times 0, 6, 24, 48 and 72 showed a significant difference. Concentration of 800 mg had the lowest light absorption and concentration 6.5 had the highest light absorption. IC50 and F90 indices for *Leishmania* parasite show that the mortality rate increases with increasing the effect time of the extract on the parasite. The IC50 of the extract was 316.14, 272.76 and 153.27 mg / ml at the time of zero, six and 24 respectively. Also, F90 index in times 0, 6, 24, 48 and 72 were 1318/18, 1286/22, 1149/65, 766/60 and 693/48 mg / ml respectively.

Conclusion: According to previous studies on the antibacterial and antifungal effects of hydro alcoholic extract of this plant and on the other hand due to its compounds such as retinol, tetrapentacontan, dihydrotacystrol, 9-octadesnoic acid and 1,2,3-propanthryl ester and based on our results it seems this plant also has anti-leishmaniasis effects.

Keywords: Matthiola; *Leishmania*; hydroalcoholic extract; plant; antileishmania

Evaluation of Anti-*Toxoplasma* Activity of Morphine on Tachyzoites of *Toxoplasma gondii* *in vitro*

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Background and Aim: Common medicines for the treatment of toxoplasmosis have limited efficacy and unwanted side effects. Opiates can affect both innate and cell-mediated immunity and stimulate the immune responses in different parasitic infections. In this work, anti-*Toxoplasma* effects of morphine were evaluated on the tachyzoites of *Toxoplasma gondii* (*T. gondii*) *in vitro*.

Methods: In order to evaluate the parasite survival, tachyzoites of *T. gondii* were seeded in 96-well plates containing RPMI 1640 medium, in the presence of various concentrations (100, 10, 1, 0.1 and 0.01 µg/ml) of morphine in triplicate and were kept at a temperature of 37 °C for 6 and 24 hours separately. Also, wells without drug and sulfadiazine plus pyrimethamine were used as negative and positive controls, respectively. After incubation, the number of treated and untreated parasites at the different drug doses was estimated by direct counting in a hemocytometer using a phase contrast microscope.

Results: The results demonstrated that tachyzoite proliferation decreased significantly in the presence of all concentrations of morphine compared to those exposed to no drugs after 24 h ($P < 0.05$). Furthermore, it was shown that exposure to different concentrations of morphine for 3 h was not effective and toxic when compared to the negative control. The growth inhibitory effects of the drugs were dose-dependent, such that 0.01 µg/ml and 100 µg/ml concentrations showed the most and the least efficacies, respectively, on inhibiting the proliferation of tachyzoites of *T. gondii*.

Conclusion: Morphine inhibited the proliferation of tachyzoites of *T. gondii* in a concentration and time-dependent manner and provided the best result after 24 hours for all concentrations. Notably, a morphine concentration of 0.01 µg/mL was most effective in inhibiting tachyzoites growth after 24 h incubation.

Keywords: *Toxoplasma gondii*; morphine; *in vivo*; tachyzoites

Evaluation of Epidemiological Status of Hydatid Cyst in Patients Admitted to Hospitals in Khuzestan Province During the Years 1390 to 1400.

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Background and Aim: Hydatid cyst or echinococcosis is a disease that is found worldwide and in many Mediterranean regions. The most polluted areas of Iran include the Alborz and Zagros Mountain ranges, where animal husbandry is common. Khuzestan province is one of the regions where hydatid cyst disease has the highest level. In this study, we study the epidemiology in hospitals of Khuzestan province.

Methods: Demographic characteristics, contact with dogs, raw medicinal plants, clinical signs, diagnosis, treatment method, number and location of cysts were collected by examining the records of patients with hydatid cyst in the province hospitals during 1390 to 1400.

Results: Of the 183 patients identified, 112 were female. Mean age (female 37.7 ± 19 , male 36.7 ± 17), housewives accounted for 49.2% of patients. 65% of the patients lived in urban areas and 42% had a history of contact with dogs. The liver was reported to be the most involved organ. The most common clinical signs were abdominal pain and hepatomegaly. 59% of victims had only one cyst.

Conclusion: Surgery was the most common treatment and radiology was the most common diagnostic method. The prevalence of human hydatidosis has been overlooked as the most important disease that should be considered by health policy makers. In addition, training programs are needed to better understand the symptoms of the disease and identify sources of infection in the high-risk group.

Keywords: *Echinococcus granulosus*; epidemiological; Khuzestan; Iran

Evaluation of the Occurrence of Covid-19 Disease in Patients with Hydatid Cyst in Ardabil Province

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Background and Aim: In order to evaluate the occurrence of Covid-19 disease in patients with hydatid cyst, this study was performed from April to September 2020 in Imam Khomeini Hospital in Ardabil (Corona center of the province).

Methods: For this purpose, 38 patients with a history of surgery and treatment of hydatid cysts were evaluated for the Covid-19 infection test. Before sampling, the code of ethics was obtained from the National Ethics Committee of the university. Among these patients, 8 patients were in close contact with a patient with Covid-19 infection (close relatives). On the other hand, the anti-hydatid antibody was performed in 150 patients with Covid-19 infection. ELISA test was used to evaluate hydatid cyst antibody and PCR test was used to evaluate Covid-19 infection.

Results: The results showed that all patients with hydatid cyst or treated with anti-hydatid cyst drugs were negative for the Covid-19 PCR test. In addition, the serum of all patients with Covid-19 infection was negative for anti-hydatid antibodies. While 3-10% of the population of Ardabil province were positive for anti-hydatid antibodies.

Conclusion: There are two possibilities for these results: either there is an antagonistic relationship between hydatid cyst antigens and Coronavirus proteins or the use of albendazole has led to protection against Covid-19.

Keywords: Covid-19; hydatid cyst; clinical signs; Ardabil

Evaluation of the Therapeutic Effect of Topical Tamoxifen on Experimental Cutaneous Leishmaniasis Caused by *Leishmania major* in Balb/c Mice

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Background and Aim: Iran is one of the endemic countries of cutaneous Leishmaniasis (CL). Not only is there no effective vaccine for the disease, but there is no definitive therapy for it. Using topical drugs can be a good treatment option. Tamoxifen has cell induction apoptosis and anti-viral properties which can be used in the control of eukaryotic parasites such as *Leishmania*.

Methods: Forty Balb/c female mice aged 4 to 6 weeks were used along with *Leishmania major* (MRHO/IR/75/ER). Mice were infected with 1X10⁶ metacyclics and divided into 5 groups of 8. The drug was prepared in two forms, ethanolic tamoxifen 1% (ET) and tamoxifen citrate ointment 0.1% (TCO), and test and control groups were treated for 28 days. the ulcer size average and mean parasite burden was clinical recovery indications.

Results: The ulcers' mean diameter in both experimental groups were significantly different from control groups without treatment and placebos. The largest ulcer's mean size was related to the untreated control group. The group treated with TCO had the smallest mean lesion diameter but was not significantly different from the group receiving ET. The parasite burden was equal in both groups. And the difference in decreasing the mean parasite burden compared to the negative control and placebo groups was significant. Clinical and parasitological follow-up of the groups up to 2 weeks after the end of the therapy course continued to confirm the lasting efficacy of topical tamoxifen in both formulations.

Conclusion: The results clearly showed the effect of topical tamoxifen on CL lesions in the Balb/c model. We suggest tamoxifen as an effective option for the CL treatment.

Keywords: Therapeutic; Topical Tamoxifen; Cutaneous Leishmaniasis; *Leishmania major*; Balb/c mice

Evaluation of Toxoplasmosis Infection in Patient with Lymphadenopathy in the West of Mazandaran Province, 1399

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Background and Aim: *Toxoplasma gondii* is an obligatory intracellular protozoan parasite that is very common in Iran and has clinical symptoms including: Congenital toxoplasmosis causes chronic ocular toxoplasmosis, and acquired toxoplasmosis is usually asymptomatic, with congenital toxoplasmosis sometimes having musculoskeletal complication. The aim of this article was to evaluate toxoplasmosis infection in patients with lymph node disease in western Mazandaran province in 1399.

Methods: In this descriptive and analytical study, 457 patients with lymphadenopathy were studied in 1399. Blood samples were isolated from patients and tested for IgG and IgM antibodies using ELISA test and analyzed the results using T_test and ANOVA statistical tests and SPSS- ver 19 software.

Results: Out of 457 samples, 372 (81.4%) were female and 85 (18.5%) were male. ELISA test results showed that 314 people were positive for IgG antibody and 7 people were positive for IgM antibody.

Conclusion: The results showed that most of the patients with lymphadenopathy from this study were in the chronic stage of toxoplasmosis infection and only 7 people were in the active stage of the disease, which needs additional tests.

Keywords: *Toxoplasma gondii*; lymphadenopathy; toxoplasmosis; ELISA; chronic

Familial Infection by *Fasciola* Parasite in Kermanshah Province, Iran

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Background and Aim: *Fasciola* is an important zoonotic parasite that causes fasciolosis and commonly affects livestock and humans, are accidental hosts. Here, we report familial infection by *Fasciola* during the acute stage of disease in 2018.

Methods: A 33-year-old woman was referred to the Gastroenterology and Hepatology Clinic of the Imam Reza Therapeutic Educational Hospital (Kermanshah, Iran), Kermanshah city due to having a 2-week history of severe illness, up to 39 degrees Celsius, jaundice, right upper quadrant abdominal pain and loss of appetite. There was a blood eosinophilia of 25% and computed tomography revealed hypodense areas at 4th and 8th segments of liver, suggesting a parasitic infection with liver involvement.

Results: Antibodies specific to *Fasciola* ES antigens were detected in the serum sample by ELISA test. A history of ingesting local watercress was reminded by patient. Fasciolosis was also proven in her 81-year-old father who for some time had complained of right upper quadrant abdominal pain and loss of appetite, her 40-year-old brother with similar symptoms. The patients were received triclabendazole (Egaten) at a dose of 10 mg/Kg, followed by a subsequent resolution of clinical symptoms and a decrease in blood eosinophil counts.

Conclusion: Consumption of wild freshwater plants such as watercress is a risk factor for fascioliasis.

Keywords: *Fasciola*; ELISA; Diagnosis; Kermanshah; Iran

Fe₃O₄@Bio-MOF Nanoparticles Combined with Artemisinin, Glucantime®, or Shark Cartilage Extract on Iranian Strain of *Leishmania major* (MRHO/IR/75/ER): An *in vitro* and *in vivo* Study

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Background and Aim: In the present study, we examined the effects of Fe₃O₄@bio-MOF nanoparticle (Nano-FO) plus artemisinin (Art) and glucantime (Glu) or shark cartilage extract (ShCE) on *Leishmania major* *in vitro* and *in vivo*.

Methods: This experimental study was conducted at the laboratory of Department of Parasitology, Tarbiat Modares University, Tehran, Iran during 2016-2017. The promastigote and amastigote assays were performed were conducted at the presence of 3.12- 400 µg/mL of the drug combinations. According to *in vitro* IC₅₀ results, the combinations of 12.5µg/mL NanoFO with 25 µg/mL Art as well as 200 µg/mL Glu and 0.5 mL of 20 mg/kg of ShCE were used to treat BALB/c mice. During and at the end of the treatment, the lesion sizes were measured. Parasite loads, cytokine levels were evaluated at the end of the treatment.

Results: The IC₅₀ of Fe₃O₄@bio-MOF-Artemisinin (Nano-FO/Art), Fe₃O₄@bio-MOFGlucantime (Nano-FO/Glu), and Fe₃O₄@bio-MOF-Shark cartilage extract (Nano-FO/ShCE) on promastigotes were 12.58 ± 0.12, 235 ± 0.17, and 18.54 ± 0.15, respectively. These results on amastigotes were 10.32 ± 0.01, 187 ± 0.03, and 338 ± 0.07 µg/mL, respectively. The apoptosis percentage of these combinations were 32.54%, 20.59%, and 15.68% in promastigotes and 15.68%, 12.84%, and 3.51% in infected macrophages, respectively with no toxicity on uninfected macrophages. *In vivo* results showed that the size of lesions significantly decreased against all drugs combinations, but Nano-FO/Art combination with Selectivity Index of 23.62 value was safe, and more effective on healing of lesions than other drugs combinations (P = 0.003).

Conclusion: This study suggested that Nano-FO/Art combination can be considered as an anti-leishmania combination therapy in CL induced by *Leishmania major*.

Keywords: Artemisinin; Glucantime; Shark cartilage extract; *Leishmania major*

Frequency Distribution of Hydatid Cyst in Patients Admitted to Hospitals in Ilam City During 2014 to 2018

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Background and Aim: Hydatid cyst, which is caused by the larval stage of the parasite *Echinococcus granulosus*, is one of the most important parasitic diseases of humans and animals. Hydatid cyst disease in humans is important because it affects sensitive and vital organs of the body, especially the liver and lungs. This study investigated the frequency distribution of hydatid cyst in patients admitted to hospitals in Ilam city during 2014 to 2018.

Methods: This is a descriptive and retrospective study with respect to demographic information (age, sex, Job, level of education and place of residence) and clinical information (involvement of various organs and length of hospital stay) from the records of patients admitted to hospitals in Ilam city during 2014 to 2018.

Results: 49 participants in this study, 27 (55.1%) were female and 22 (44.8%) were male. The mean age of the subjects was 44.19 with a standard deviation of 16.62 and the age group of 21 to 30 years (36.7%) had the highest frequency. 59.1% of patients were from urban areas and most of them were housewives (34.6%) and farmers (24.4%). 36 patients (73.4%) had hepatic hydatid cyst and 8 patients (16.3%) had alveolar hydatid cyst. 44 patients (89.7%) had a single-organ involved and 5 patients (10.2%) had a two-organ involved. The duration of hospitalization after surgery varied from 2 to 68 days. The duration of hospitalization was 1-10 days.

Conclusion: The results of this study showed that hydatid cyst disease can be surprisingly common in seemingly low-risk populations, such as those who live in urban areas or live without direct contact with dogs and farm animals. Occupationally, housewives were more likely to have hydatid cysts than other occupational groups, with more contact with vegetables contaminated with parasite eggs being the most important cause of infection.

Keywords: Frequency; hydatid cyst; hospitalized patients; Ilam

Genotyping of *Acanthamoeba* Isolates from Keratitis Patient Referred to Farabi Hospital

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Background and Aim: Amoebic keratitis is caused by free-living *Acanthamoeba* spp., which is abundant in the environment and can also be found in contact lens and eyewash solutions. This study aimed to evaluate amoebic keratitis in patients with keratitis symptoms.

Methods: In this study, 32 clinical samples were collected from keratitis patients referred to Farabi hospital. All samples were examined based on morphological and molecular techniques. The genotypes were determined by sequencing the partial of the 18S rRNA gene.

Results: The obtained results revealed that most of these *Acanthamoeba* strains belonged to genotype T4 in clinical samples. The presence of T3 genotype in clinical samples was also shown after the genotyping analysis and to our knowledge, this is the first report of T3 genotype *Acanthamoeba* keratitis patients in Iran.

Conclusion: The widespread distribution of pathogenic *Acanthamoeba* across and the increasing number of contact lens wearers in Iran requests more awareness within the public and health professionals as a risk for human health in Iran and worldwide.

Keywords: Genotype; *Acanthamoeba* Keratitis; T4

Geo-climatic Factors and Prevalence of Chronic Toxoplasmosis in Pregnant Women

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Background and Aim: In this study, we evaluated the effects of geo-climatic parameters and other potential risk factors on the prevalence of chronic toxoplasmosis (CT) in pregnant women.

Methods: We searched PubMed/MEDLINE, Web of Science, EMBASE, Scopus, and SciELO databases for sero-epidemiological studies published between January 1988, and February 2021. We performed meta-analysis and meta-regression by using a random effect model to synthesize data. A total of 360 eligible datasets, including 1,289,605 pregnant women from 94 countries, were included in this study.

Results: The highest and lowest prevalence rates were estimated for latitudes of 0-10° (49.4%) and $\geq 50^\circ$ (26.8%); and for the longitude of 80-90° (44.2%) and 110-120° (7.8%), respectively. Concerning climatic parameters, the highest and lowest prevalence rates were estimated in regions with the mean relative humidities of $> 80\%$ (46.6%).

Conclusion: Therefore, different regions in the world may benefit from different types of interventions, and novel preventive measures in a region should be developed according to local climate, agricultural activities and people culture.

Keywords: *Toxoplasma* infection; pregnant women; geo-climatic parameters; risk factors; meta-analysis; meta-regression

High Frequency of *Cystoisospora belli* and *Cyclospora cayetanensis* Infection among Iranian Patients with Colorectal Cancer

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Background and Aim: From among intestinal parasites, coccidian intestinal parasites such as *Cystoisospora belli* (*C. belli*, previously known as *Isospora belli*), and *Cyclospora cayetanensis* (*C. cayetanensis*) are well-known as opportunist parasites, particularly in patients with cancer. This study assessed the prevalence of *C. belli* and *C. cayetanensis* in patients with cancer in Lorestan Province, Southwest of Iran.

Methods: This cross-sectional descriptive study was conducted on 87 patients with colorectal cancers, referred to the general hospitals of Lorestan from October 2017 to August 2018. A fresh stool specimen was collected from each subject in a sterile labeled container. The collected stool samples were concentrated through sucrose flotation method and then prepared for Ziehl-Neelsen staining for microscopic examination. Demographic and possible risk factors such as age, sex, education, residence, and unwashed vegetable/fruit consumption were collected by an applied questionnaire.

Results: Out of the 87 patients with colorectal cancer, eight (9.2%) were found positive for *C. belli* and *C. cayetanensis* infections, with five (5.74%) and three (3.44%) patients positive for *C. belli* and *C. cayetanensis* infections, respectively. Results also showed that sex and unwashed vegetable/fruit consumption were significantly associated with the prevalence of these parasites ($p < 0.01$).

Conclusion: The findings revealed the considerable prevalence of *C. belli* and *C. cayetanensis* in patients with colorectal cancers. It is therefore essential for gastrointestinal specialists to pay special attention to the prevalence of coccidian parasites in patients with colorectal cancer.

Keywords: Cancer; Coccidian parasite; Iran; Ziehl–Neelsen staining.

Induction of Apoptosis by Morphine on Tachyzoites of *Toxoplasma gondii* in vitro

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Background and Aim: Despite of great advances, establishing an effective therapeutic scheme for prevention and control of toxoplasmosis in both humans and animals still remains a great challenge for researchers to select new anti- *Toxoplasma* drugs with specific activity against the parasite. In this work, apoptotic effects of morphine were evaluated on the tachyzoites of *Toxoplasma gondii* (*T. gondii*) in vitro.

Methods: Tachyzoites of *T. gondii* were cultured in the presence of various concentrations of morphine and incubated for 6 h at a temperature of 37 °C in 5 % CO₂ and 95 % humidity. In addition, wells without drug and sulfadiazine plus pyrimethamine were used as negative and positive controls, respectively. The parasites were transferred to 1.5 ml microtubes and centrifuged at 3000 rpm for 5 min. The supernatant was then drained off and replaced with 500 µL binding buffer. Afterwards, 5 µL of annexin V and 5 µL of propidium iodide (PI) were added to cell pellets, and then incubated in the dark at a temperature of 24 ± 2 °C for 5 minutes. Using FACSCaliber (BD Biosciences), absorption of annexin-v was estimated and finally analyzed by CellQuest software.

Results: It was observed that the survival rates among five concentrations of morphine (100, 10, 1, 0.1 and 0.01 µg/ml) were 95.62 %, 95.28 %, 95.28 %, 94.85 %, and 94.40 %, respectively, after 6 h of treatment whereas the control group (tachyzoites) without treatment had 99.86 % viable parasites. In groups treated with sulfadiazine, pyrimethamine and sulfadiazine plus pyrimethamine demonstrated more lethal effects on tachyzoites of *T. gondii* rate than morphine group.

Conclusion: Apoptosis occurred in tachyzoites of *T. gondii* after exposure with different morphine concentrations, although these apoptotic effects were not significant. Morphine showed lower toxicity upon parasite compared with sulfadiazine, pyrimethamine and sulfadiazine plus pyrimethamine group

Keywords: *Toxoplasma gondii*; morphine; in vitro; apoptosis

Investigate of *Blastocystis* Subtypes in HIV+ patients in Karaj.

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Background and Aim: *Blastocystis* is a cosmopolitan intestinal protozoan parasite which is mainly found in humans and animals. So far, 17 subtypes of the parasite have been identified based on SSU-rRNA analysis, nine of which are prevalent in human beings. Since morphological discrimination is less reliable, molecular methods are mostly used for diagnosis. *Blastocystis* is more prevalent among mentally-retarded individuals, immunocompromised patients and graft recipients. The aim of this study was to investigate the *Blastocystis* subtypes in HIV+ patients in Karaj.

Methods: A number of 60 stool sample were collected from HIV+ patients. After microscopic examination, samples were preserved in 5% potassium dichromate. DNA extraction of each sample was done using CTAB method, followed by RNA elimination. Subsequently, after DNA barcoding of all extracted sample by PCR, positive sample were probed by two subtype-specific primers. Also, one sample of each positive subtype was sequenced for confirmation.

Results: Following DNA barcoding, 20(33.3%) sample were positive for *Blastocystis*. Of these, 12(54.5%) cases were ST3, 8 (45.5%) cases were found to be ST1.

Conclusion: *Blastocystis* is the most prevailing intestinal protozoan parasite of humans on which there are many equivocations yet. The dominance of human ST3 have been reported in many studies. Being considered as the most prevalent *Blastocystis* subtype worldwide. For the first time in Iran ST9 has been reported. Moreover, the unknown subtype possesses inter-subtype variation. Further studies are recommended to shed light on the epidemiology of *Blastocystis* in country.

Keywords: *Blastocystis*; Subtypes; PCR bar-coding; HIV+ Patients

Investigation of Antiparasitic Activity of Silver Nanoparticles on *Leishmania major*

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Background and Aim: Cutaneous leishmaniasis (CL) is the most common form of skin disease caused by a single-celled parasite that is transmitted by the bite of a phlebotomine sandfly. Symptoms usually appear three months to a year after the bite. Nanotechnology is now recognized as a key activating technology (KET) for its various applications in science and medicine. In recent years, the scientific value of NPs in the treatment of leishmaniasis has increased due to the improvement of anti-leishmaniasis drugs. In this study, silver biogenic nanoparticles (AgNPs) and amphotericin-bound silver biogenic nanoparticles (AmB-AgNPs) are used.

Methods: In this study, experimental infection in several Balb/c mice by injection of *Leishmania major* promastigotes was created. After the appearance of wounds in the tails of mice, they were divided into four groups of 10 and divided into two groups of treatment with (AgNPs) and (AmB-AgNPs). once a day in the wound at three points in the group receiving glucantime they had done it muscularly for 5 weeks was injected. In the negative control group, the mice did not receive any medication. Then, the weight of the mice and the diameter of the *Leishmania major* wound was daily for 5 weeks. They were measured and recorded and compared with each other.

Results: Wound diameters in the nanoparticle recipient treatment group (AgNPs) and (AmB-AgNPs) were significantly less from the untreated control group, but the wound diameters were larger than those treated with glucantime.

Conclusion: silver biogenic nanoparticles (AgNPs) and amphotericin-bound silver biogenic nanoparticles (AmB-AgNPs) nanoparticles show anti-leishmaniasis effects in the in vitro environment. But this effect is lower than glucantime and does not completely heal the lesions.

Keywords: antiparasitic; silver nanoparticles; *Leishmania major*

Investigation of the Effect of Chloroformic Extract of *Allium Paradoxum* on *Trichomonas vaginalis* Parasite *in vitro*

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Background and Aim: Trichomoniasis is one of the most important human parasitic diseases that can cause genital disorders in both men and women. Infertility is the crucial clinical manifestation of *Trichomonas vaginalis* (*T. vaginalis*). There are several treatments for this type of parasite, metranidizole is the first choice and golden standard treatment for this complication that has a wide range of side effects, so finding newer treatments is critically essential. The aim of this study was to evaluate the effect of chloroformic extract of *Allium paradoxum* (*A. paradoxum*) on *T. vaginalis* *in vitro*.

Methods: In this experimental study, *T. vaginalis* parasites were collected from patients with trichomoniasis; who had been referred to gynecology clinics and cultured in Dorset medium. *A. paradoxum* was collected from the mountainous areas of Mazandaran province in spring and dried at laboratory temperature and away from direct sunlight, and finally powdered using an electric shredder. Using maceration method, chloroform extract of this plant was prepared at concentrations of 50, 100, 200, 400 and 800 µg / ml. Different concentrations of this plant were tested at different times at 37 ° C on *T. vaginalis* parasite. The survival rate of this parasite was also studied using 0.1% eosin as a vital staining.

Results: *A. paradoxum* at concentrations of 50 and 100 µg/ml showed low effectiveness on *T. vaginalis* trophozoites and the concentrations of 200, 400 and 800 µg/ml, after 24 hours has a very strong anti-*Trichomonas* activity, similar to metranidizole.

Conclusion: According to the results of this study *A. paradoxum* could be an alternative anti-*Trichomonas* treatment of course after additional studies. Future investigations id needed to assess the toxicity and other side effects of this plant.

Keywords: *Allium paradoxum*; *Trichomonas vaginalis*; Vaginitis; Trichomoniasis

Investigation of the Function of Monoclonal Antibody Production Techniques in the Diagnosis of Amastigotes and Promastigotes of *Leishmania infantum*

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Background and Aim: Leishmaniasis is widely distributed around the world and is highly important for human as a serious infectious disease. Hybridoma technologies can be used for the reliable development of monoclonal antibodies (mAbs) and their subsequent production in different cells. In principle, the method has been widely applied in various fields such as diagnosis applications, disease monitoring and identifying prognostic markers. The purpose of this study Investigation of the role of mAbs production techniques in the diagnosis of amastigotes and promastigotes of *Leishmania infantum* (*L. infantum*).

Methods: Amastigotes and promastigotes of *L. infantum* were cultured in specific medium Amastigotes and promastigotes antigens together with complete Freund's adjuvant and incomplete Freund's adjuvant were injected into 6-week-old female mice. There after spleen lymphocytes were fused with myeloma SP2/0 in ratio of 10:1 by polyethylene glycol. Positive hybrids in HAT and HT medium were identified with ELISA and subsequently subcloned. limiting dilution method was applied for monoclonal determining.

Results: In this study we produced several Hybridoma cells and mAbs against amastigotes and promastigotes of *L. infantum* antigens. These antibodies recognized specific antigens with different techniques.

Conclusion: This study produced mAbs against amastigotes and promastigotes of *L. infantum* antigens. These antibodies have reactivity against *L. infantum* and could be used in diagnosis of leishmaniosis. Since mAbs recognize only their target antigens and no other unrelated proteins, pinpoint medical treatment is possible. Global demand is dramatically expanding.

Keywords: monoclonal antibody; diagnosis; amastigotes; promastigotes; *L. infantum*

Investigation of the Prevalence of Head Lice and Factors Affecting Them in Infected People Referring to Gerash County Health Center

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Background and Aim: Pediculosis is one of the most common parasitic diseases that affect a wide range of age groups. The prevention of head lice infections promotes the physical and mental health of people in the community. Thus, the aim of this study was to investigate the prevalence of head lice infections and the factors affecting them in those who referred to health centers in Gerash County from 2011 to 2018.

Methods: This descriptive cross-sectional study was conducted on patients suspected with head lice infections in health centers in Gerash. These infections were diagnosed by observing adult lice, nymphs, or nits on the head with the help of a magnifying glass. The collected data were then analyzed using SPSS software, version 22 and p-value less than 0.05 was considered as the significance level.

Results: In general, 66,410 patients suspected with head lice were examined, of whom 2,547 cases (3.83%) were reported with confirmed infections including 2395 female (94.03%) and 152 male (5.97%) cases. The highest levels of infections were reported in the age group of 6-10 years while the lowest levels of infections were found in children less than 6 years of age. A significant statistical relationship was observed between head lice and gender, age, season, and the place of residence, and year of infection ($p < 0.05$).

Conclusion: Due to the increasing trend of head lice infections in recent years, pediculosis is still considered a health problem. Therefore, training on personal hygiene, adequate access to health services, and early diagnosis and treatment can play an important role in the prevention and elimination of head lice.

Keywords: Head lice; Pediculosis; Epidemiology; Gerash

Isolation and Identification of *Cryptosporidium* spp. in the Inlet, Sludge and Outlet Sections of Wastewater Treatment Plants of Chaharmahal and Bakhtiary Province (2018-19)

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Background and Aim: *Cryptosporidium* is an obligate, intracellular – extracytoplasmic coccidian protozoan parasite belonging to the phylum Apicomplexa that infects human and a wide range of vertebrate by ingestion of contaminated food or water with oocyst. The contamination of drinking water sources to oocysts of different species of *Cryptosporidium* through enter the feces of various domestic and wild animals or through exposure to human sewage can cause human epidemics. The purpose of this study was to compare the amount of *Cryptosporidium* contamination in the wastewater treatment plant inlet, sludge and outlet sections in the Chaharmahal and Bakhtiari province.

Methods: In this study, 56 samples of six wastewater treatment plants (Saman, Joungan, Borujen, Hafshjan, Shahrekord and Farsan) were studied during four seasons from 2018-19. The samples of wastewater treatment plant including inlet, sludge and outlet were study by Ziehl–Neelsen staining method for microscopic examination. Then, to confirm the diagnosis, the amplification of 18S rRNA gene was performed by PCR and Nested PCR methods, also RFLP method was used in order to identify the species and genotypes of *Cryptosporidium* by SspI and VspI restriction enzymes.

Results: Microscopically, 10 of 56 samples were positive with oocysts of *Cryptosporidium* by Ziehl–Neelsen staining method and one sample was positive for this parasite using the molecular assay, and the findings of RFLP method showed that it was *Cryptosporidium parvum* isotype bovis.

Conclusion: The findings of this study showed, although presence of *Cryptosporidium* in wastewater is low but due to the presence of *Cryptosporidium parvum* in wastewater effluents of treatment plants and use of wastewater in agricultural fields can be considered as a source of animal contamination.

Keywords: *Cryptosporidium*, Urban wastewater, Chaharmahal and Bakhtiari, PCR, PCR-RFLP.

MicroRNA-365 Promotes Apoptosis in Human Melanoma Cell A375 Treated with Hydatid Cyst Fluid of *Echinococcus granulosus sensu stricto*

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Background and Aim: Hydatid cyst fluid (HCF)-based therapeutics has experimentally targeted approaches for treating human cancer cell lines. MicroRNA-365 (miR-365) has been reported to be an important tumor suppressor miRNA in cancers. However, it remains unknown, how miR-365 plays a pivotal role in inducing apoptosis in HCF-treated cancer cells *in vitro*.

Methods: The fertile/infertile HCF was aspirated from liver of infected sheep and in terms of molecular taxonomy was identified as G1 genotype of *Echinococcus granulosus sensu stricto*. A375 human melanoma cancer cells were cultured into two groups: fertile and infertile HCF-treated A375 cells. To assess the cytotoxicity of various concentrations of HCF on melanoma cells, cell viability was determined by using MTT assay. The IC₅₀ value of HCF on A375 cells was determined 85 µg/mL. Caspase-3 enzymatic activity was evaluated by fluorometric assay in the HCF-treated melanoma cells. In addition, the mRNA expression of Bax, Bcl-2, Caspase-9 and miR-365 were determined by qRT-PCR.

Results: Findings of MTT assay showed that concentrations 85 µg/mL to 100 µg/mL of fertile HCF have the highest mortality (50%–52%) on A375 cells during 24 h. The fold change of Bax/Bcl-2 ratio, Caspase-9, miR-365 and Caspase-3 activity was higher in the fertile HCF-treated melanoma cells compared to infertile fluid treated A375 cells and human normal epithelial cell (as control cell). In conclusion, we over-expressed the miR-365 in melanoma A375 cells, via treatment of fertile HCF. Our findings suggested that inducing high expression of miR-365 might be a negative regulator of melanoma growth through activation of pro-apoptotic Bax, Caspase-9 and Caspase-3 that are essential to intrinsic apoptotic pathway.

Conclusion: These findings provide new insights into the use of *Echinococcus* HCF-derived metabolites in the design of drug therapies and *in vivo* tumor cell vaccine to combat melanoma progression.



Keywords: Hydatid cyst fluid; Melanoma cell line A375; MicroRNA-365; Apoptosis



Misdiagnosis and mistreatment of demodicosis

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Background and Aim: Demodicosis in humans caused by two species of *Demodex* called *folliculorum* and *brevis*, almost in face. Although, there is no clinical symptoms in individuals with normal immunity system, but in many cases, dermatitis, rough, dry and scaly skin rosacea, particularly asymmetrical papulopustular or granulomatous variants and in some cases, perioral dermatitis, blepharitis are observed. Clinical manifestations of demodicosis are varied. The infestation may be free of symptoms or the lesions might be as rose- red pustules and the most typical features of rosacea is characterized by the presence of an erythematous papule- pustule rash, mainly in the face and Inflammation in acute and chronic forms may occur. The majority of cases are misdiagnosed with dermatophytosis and rosacea.

Methods: One hundred and ninety cases with face lesions referred to the laboratory for fungal examination. Scraping from the skin of lesions and preparation of smears with 20 % KOH was carried out and examined microscopically.

Results: *Demodex folliculorum* was observed, although in few cases co-infection of dermatophytosis and cutaneous leishmaniasis were demonstrated. The duration of lesions in most of the cases was more than 2 years. Before any examination, all patients were treated for rosacea or fungal infection and no healing occurred and even some cases became worse.

Conclusion: It is necessary that before the administration of any medicine, the cause of infection must be identified.

Keywords: Demodicosis; dermatophyte; rosacea; misdiagnosis

Molecular Diagnosis of *Strongyloides stercoralis* in Faecal Samples

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Background and Aim: *Strongyloides stercoralis* (*S. stercoralis*), is a parasitic nematode in humans, lives inside the mucosa of the small intestine and causes strongyloidiasis. This study was conducted for microscopic diagnosis and molecular characterization of *S. stercoralis* in Mazandaran; the endemic region of the parasite in Iran.

Methods: From April 2017 to September 2017, a number of 2195 human stool samples were collected from various regions of Mazandaran province, North of Iran. All stool samples were analyzed using the formalin ether method. The positive fecal samples of *S. stercoralis* and 300 random samples were selected for DNA extraction. A set of primer pairs were used for conventional PCR to activate amplification of 509 bp target in a PCR reaction in the mitochondrial cytochrome c oxidase subunit 1 (Cox1) gene.

Results: In the fecal examination, 21 (0.95%) stool samples were found to be positive for *S. stercoralis*; and 162 (7.38%) samples were positive for other parasites. All 21 positive samples were confirmed by PCR method as *S. stercoralis*. The sequence of the samples had 99% overlap with *S. stercoralis* in GenBank.

Conclusion: The results showed that conventional PCR could detect all specimens that microscopically were positive.

Keywords: *Strongyloides stercoralis*; molecular characterization; Cox1 gene; Iran

Molecular Identification of *Leishmania* species in Patients with Cutaneous Leishmaniasis Referred to Health Centers in Kurdistan Province in 2020

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Background and Aim: Cutaneous leishmaniasis is one of the health problems of tropical and subtropical countries of the world, including Iran. In Iran, the prevalence of cutaneous leishmaniasis is increasing so that in recent years, new endemic areas of this disease have been identified. According to several cases of leishmaniasis in Hamadan province, this study was conducted to identify the species of human cutaneous leishmaniasis in Hamadan province.

Methods: In this descriptive cross-sectional study, all available slides (59) that were positive for cutaneous leishmaniasis and kept in the archives of Kurdistan health centers were collected. First, all slides were examined for the presence of amastigote, and then samples were examined from the slide surface for DNA extraction and PCR by specific primers. Finally, positive molecular cases were sequenced for confirmation.

Results: In this study, out of 59 patients under study, 55 were detected positive by microscopy and 56 by molecular method. 38 cases (67.86%) were diagnosed with *Leishmania major* and 18 cases (32.14%) were diagnosed with *Leishmania tropica*. 53 cases were related to male patients and 3 cases were female cases. 73.2% had dry lesions and 62.7% had less than two active skin lesions. All patients were treated with glucantime and no treatment failure was reported. 38 patients (67.86%) improved with topical injection and 18 patients (32.14%) improved with systemic injection. The highest frequency of infection (44.64%) was observed in the age group of 21 to 30 years. 35 patients (62.5%) were residents of the city and most (22 patients) were infected (39.28%) employees. All patients had a history of traveling to other provinces, the most of which was 12 cases (21.42%) to the city of Mehran and then (17.85%) ie 10 cases to different parts of Ilam province. Statistical analysis showed that there was no significant relationship between the frequency of cutaneous leishmaniasis in the subjects and the epidemiological variables studied.

Conclusion: Cutaneous leishmaniasis is not considered as a health problem in Kurdistan province, but due to travel and proximity to border and endemic areas, there is always a potential risk of the disease. So, planning to control the disease and continuous monitoring of new cases should be always considered. The study of the relationship between demographic information and individuals with cutaneous leishmaniasis-causing species was not statistically significant, which could be due to the limited population.

Keywords: Cutaneous leishmaniasis; *Leishmania major*; *Leishmania tropica*; PCR

New Prevalence Surveillance of *Toxoplasma gondii* Among Rodents and Stray Cats by ELISA Avidity and nested-PCR Methods, Northeast of Iran

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Background and Aim: Rodents and stray cats are the sources of many parasitic infections including *Toxoplasma gondii* (*T. gondii*), for other animals and human. Toxoplasmosis has a wide range of laboratory factors in its intermediate and definite hosts. Regarding the importance of rodents and stray cats as the hosts that spread the *T. gondii*, it is necessary to obtain comprehensive information about these animals in the life cycle of *T. gondii*. The objective was to investigate the new prevalence of toxoplasmosis among target animals in Iran, using GRA6 gene in combination with ELISA avidity.

Methods: In this study, 286 rodents and 210 stray cats were collected and their heart tissues extracted to obtain DNA, blood samples and IgG Ab of *T. gondii* parasite. We detected the positive tissue samples in our study by the nested-PCR method. Then, we examined *T. gondii* IgG ELISA avidity for assessment of toxoplasmosis among rodents and stray cats. This study, was conducted in January to March 2017, based on the prevalence study.

Results: The findings revealed that 246/286 (86.01%) of rodents and 180/210 (85.71%) of stray cats were positive by IgG ELISA avidity methods. moreover, 68 rodents' samples and 38 stray cats' samples were positive concerning the GRA6 *Toxoplasma* gene; and these positive samples were at intermediate levels for IgG avidity.

Conclusion: We concluded that the new prevalence of toxoplasmosis among rodents and stray cats was at high levels, using the serologic method in Northeast of Iran and the results of quantitative ELISA avidity were as the same as those of the nested-PCR for detecting recent toxoplasmosis in these hosts.

Keywords: Iran; Molecular detection; New prevalence; Serology study; Toxoplasmosis.

Occurrence of Intestinal Microsporidiosis among Referred Individual to Medical Centers Laboratories in Eastern Iran

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Background and Aim: Microsporidia are obligate intracellular pathogens. The parasite can infect a wide range of vertebrate and invertebrate hosts among all over the world. The current study was performed to evaluate the prevalence of intestinal microsporidiosis by staining and molecular methods in South Khorasan province of Iran.

Methods: From December 2019 to December 2020, 720 stool samples were stained by specific Weber Modified Trichrome to detect *Microsporidia* spores. The DNA extraction was done by Kit and to amplify the SSU rRNA gene of *Microsporidia* Spp. the Nested-PCR was used with genus specific primers. Finally, data analyzing was performed by chi-square (X²) test, using SPSS. 21 software.

Results: From 720 stool samples, 25 (3.47%) samples were microscopically positive for intestinal *Microsporidia* infection. Out of 25 positive samples of *Microsporidia* in microscopic method, 22 samples were positive in PCR method. In this study, most of the molecularly approved patients were referred to the hospital (68.00%) and were in the age range of 30 to 50 years (40.80%). 92.00% of patients also had gastrointestinal symptoms and were infected in the summer (40.00%), ($P < 0.05$). Using specific primers and sequencing genes, it showed that all of these samples were infected with *Enterocytozoon bienersi*.

Conclusion: This study was the first evaluation of the prevalence of *Microsporidia* in South Khorasan province. *Microsporidia* infection is prevalent in the public population of eastern Iran. Statistical analysis of the results showed that factors such as education, age, gastrointestinal symptoms and season can be risk factors associated with parasitic infections.

Keywords: *Microsporidia*; Staining, Nested-PCR; South Khorasan

Phylogenetic Analysis of *Giardia* Genotypes in Iran based on Triose Phosphate Isomerase (TPI) Genes

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Background and Aim: Giardiasis is a zoonotic disease that is a forgotten disease and a cause of diarrhea. Giardiasis has a worldwide prevalence that is estimated to affect approximately 280 million people annually. *Giardia lamblia* is genetically composed of 8 different assemblages (A, B, C, D, E, F, G, H). Various studies show that *Giardia* assemblages are different in pathogenicity. The aim of this study was to investigate the nucleotide sequences related to different types of *Giardia* assemblages in Iran and Kurdistan province and compare these nucleotide sequences with the samples in the World Gene Bank and other parts of Iran and the world and the relationship between infectious species in humans and livestock as well as possible ways of transmission to humans using molecular bioinformatics analysis. Examined. Investigating this issue is important in terms of applying appropriate control and prevention strategies and improving the health and well-being of society.

Methods: This study was performed cross-sectionally. PCR products of *Giardia* samples based on triose phosphate isomerase gene in Kurdistan province and various human and animal groups from other parts of Iran and the world have been available in the World Gene Bank. After reviewing various studies and searches conducted in the World Gene Bank, 32 nucleotide sequences registered from different parts of the world were selected for this gene. Bioinformatics analysis was performed using Chromas programs for editing, Blast for gene comparison and homology determination, ClustalW2 for group alignment, MEGA11 for phylogenetic tree drawing, GenBank for receiving similar genes. Finally, using all the mentioned software, comparisons of similarities and differences and genetic analysis of *Giardia* species in Kurdistan province in comparison with Iran and other parts of the world were performed.

Results: The alignment of 32 selected samples in this study showed that less than 10 nucleotides of 500 nucleotides related to this gene, were different. Blasting of samples based on the triosephosphate isomerase gene from the Kurdistan province showed that these sequences have more than 95% overlap and homology with other registered sequences of *Giardia* parasite in other parts of Iran for the same gene. Also, by selecting some samples from Iran and comparing them with other registered sequences from other parts of the world, it showed that this similarity also exists for Iranian samples in comparison with other registered sequences from different parts of the world. This homology is

more than 95% between human and animal samples and samples isolated from the environment such as drinking water sources.

Conclusion: All registered sequences related to Iran are almost completely similar, which may indicate the existence of a type of *Giardia* for each of these assemblages in all parts of Iran. Since *Giardia* specimens in humans, animals, and the environment are similar we conclude that *Giardia* is a zoonosis. In this study, three phylogenetic trees have been drawn. in the first phylogenetic tree for 32 selected *Giardia* samples from different regions of Iran and the world has been drawn we consider Sequences that are placed together in a complex are a type of assemblage. Assemblages A and B were placed in one root and assemblage E was placed in another root. The second phylogenetic tree was drawn for all *Giardia lamblia* assemblages (except assemblage H) and one sample of *Giardia ardeae* (out-group) from the World Gene Bank. According to this tree, assemblages E, F, and A are more similar to each other. Also, assemblages B, C, and D are more similar to each other. these 6 assemblages are more different from assemblage G. The third comparative phylogenetic tree for sub assemblages A and B of the World Gene Bank were performed based on the triosephosphate isomerase gene. Sub-assemblages BIII and BIV

Keywords: Phylogenetic; *Giardia*; TPI

Phylogenetic Analysis of *Toxoplasma gondii* Type II and Type III by PCR RFLP Plus Sequencing on Wild-Rats of Golestan Forest, Iran

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Background and Aim: *Toxoplasma* is a genus of intracellular parasites, which was observed in the many types of mammalians. *Toxoplasma* protozoa that were detected from many types of rats have the equal phenotypic characteristics but have different clinical symptoms and genotypes. Mostly, domestic animals and rodents are the hosts in the reservoirs in the environment. The objective of this article was to phylogenetic identification of toxoplasmosis in wild-rats of Golestan forest using secretory antigens type 1 gene.

Methods: In this surveillance, the researchers gathered 286 wild rodents from Golestan forest and earned DNA from brain and heart tissues to prove SAG1 gene from these hosts. They split these rodents into four groups and then analyzed the positive samples by the Polymerase chain reaction basis.

Results: In this surveillance, they discovered 24% of tissues of the wild-rats were infected for these protozoa. Over 55.8% of tissues belonged to *Rattus*, 17.6% tissues belonged to *Rattus norvegicus*, 14.7% of tissues belonged to *Mus musculus*, 11.7% of tissues belonged to *Rombumys opimus*, also with attention of phylogenetic analysis, 50 infected rats belonged to genotype III and 18 infected rats belonged to genotype II.

Conclusion: Conclusion showed that different types of wild rats disseminated this type of infection; also, SAG1 molecule was a very important molecule to detect toxoplasmosis in rats of Golestan area.

Keywords: Toxoplasmosis; PCR-RFLP; SAG1; Golestan forest; Rats

Prevalence of Blood Fillers in Stray Dogs in Shirvan City of North Khorasan Province

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Background and Aim: Heartworm (*Dirofilaria immitis*) is mosquito-borne filarial nematode capable of causing serious cardiopulmonary disease in canines and felines, and pulmonary dirofilariasis in man. This research was conducted with the objectives of determining the incidence and assessing possible risk factors of canine heartworm in the Shirvan city of North Khorasan province.

Methods: In the present study, from December 2020 to July 2021 blood samples were collected from 109 guard dogs and 91 sheepdogs. And the presence of microfilaria was checked by modified Knott test.

Results: In total, only 3 of the 200 dogs were infected with microfilariae. All 3 samples were infected with *Dipetalonema reconditum* microfilaria and no specimen was infected with *Dirofilaria immitis* microfilaria. Infection rate was 2 samples in Sheepdogs (1%) and 1 sample in guard dogs (0.5%). Although the infection rate was higher in sheepdogs, there was no statistically significant difference ($p < 0.05$). Although the rate of infection was higher in dogs over 3 years old (1%) compared to those under 3 years old (0.5%) and it was higher in male dogs (1%) compared to female dogs (0.5%), but the differences were not statistically significant.

Conclusion: the results from this study showed that the only cause of infection, in the investigated area was *Dipetalonema reconditum*.

Keywords: Dog blood fillers; *Dirofilaria immitis*; *Dipetalonema reconditum*

Prevalence of *Toxocara* Infection in Patients Referred to a Medical Diagnostic Laboratory in Rasht, North of Iran

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Background and Aim: Toxocariasis is a zoonotic disease caused by the accidental ingestion of the eggs of animal nematodes, *Toxocara canis* and *Toxocara cati*, and the invasion of the viscera by the larvae which results in visceral and ocular larva migrans (VLM & OLM). The infection is characterized by various complications such as hepatomegaly, eosinophilia, and is associated with pulmonary and ocular symptoms.

Methods: In the present retrospective study, the test results of 120 (100 males [%83.3] and 15 females [%16.7]) patients referred to a medical diagnostic clinical laboratory in Rasht, North of Iran, having anti-*Toxocara* antibody (IgG) test from December 2017 to December 2019 were recorded and studied. Serological tests had been performed by indirect immunofluorescence assay (IFA). The results of the patients' complete blood count (CBC) test were also recorded.

Results: The elevated of anti-*Toxocara* IgG were seen in five (%4.1) out of 120 studied patients. Considering the sex distribution, the anti-*Toxocara* IgG test of the patients demonstrated that Two (%40) and three (%60) out of five positive patients were females and males, respectively. Three (%60) of the positive cases had eosinophilia above %10. Interestingly, all five patients with *Toxocara* infection were farmers.

Conclusion: According to the results of the present study, toxocariasis in North of Iran could be a public health problem, especially among farmers or those who are in contact with soil and animal reservoirs.

Keywords: Toxocariasis; Visceral Larva Migrans; Ocular Larva Migrans; Iran

Report of *Cryptosporidium* and *Balantidium coli* Simultaneous Occurrence in Domestic Dogs in Mazandaran

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Background and Aim: Nowadays, interaction with some animals called pets and keeping them with humans has caused the recognition and prevention of some common diseases to be more important than before. *Balantidium coli* is One of the most common pathogenic parasites. *Balantidium coli* is a type of intestinal ciliated protozoan that can cause clinical manifestations in a wide range of vertebrates such as diarrhea, anorexia, and urinary tract infections. *Cryptosporidium* is another intracellular parasite of the api-complex and sporozoa genus, which has been recognized in recent years as one of the most common intestinal pathogens and the cause of cryptosporidiosis. Clinical symptoms of this parasite include weakness and disability, diarrhea, severe abdominal spasm and significant weight loss, and fever and dehydration.

Methods: After observing some clinical signs, fecal samples were taken from patients for diagnosis we used floating methods, isolation and purification of *Cryptosporidium* and *Balantidium coli* oocytes. Flotation method was used and then the samples were stained with Ziehl–Neelsen and trichrome, respectively.

Results: After isolation and staining, the samples were examined under an optic microscope and both parasites were positive.

Conclusion: Such complications can be prevented by observing animal hygiene protocols and conducting periodic checkup, as well as the availability of sanitary water resources.

Keywords: *Cryptosporidium*; *Balantidium coli*; domestic dogs

Report of Simultaneous Occurrence of *Entamoeba histolytica* in Humans and Animals

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Background and Aim: Today, Parasitic diseases are among the most important infectious diseases and pose health problems in many countries, most especially in developing countries, due to their wide prevalence. The *Entamoeba histolytica* parasite is a protozoan of the amoeba branch and the *Entamoeba* genus invade and destroy the colonic epithelium leading. The parasite can be transmitted to animals from contaminated water and food and from eating cysts in human feces.

Methods: Diarrhea, abdominal pain, bloating, and nausea are obvious symptoms for this parasite. After observing the symptoms of the disease in the accompanying animal and taking a stool sample and following similar symptoms in the owner, a stool sample was taken from the owner and Flotation method was used to isolate the parasite trophozoite and stained with hematoxylin.

Results: After the isolation and staining process, the *Entamoeba histolytica* parasite were positive in the domestic dog. Also, the parasite was positive in the owner.

Conclusion: Such complications can be prevented by observing environmental hygiene and hand washing before consuming food and using healthy water and food protection.

Keywords: *Entamoeba histolytica*; Domestic dog; Parasitic

Risk of Recurrence of Malaria in the Shadow of Covid-19

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Background and Aim: Nowadays, COVID-19 has spread rapidly around the world and has changed to pandemic disease and it has attracted the scientists and researchers' attention from different fields of medical science as a hot research topic. There is an urgent need to tackle this invisible enemy, but other deadly diseases such as malaria should not be overlooked, otherwise, the world would be confronted with re-emergence and the dreadful impact of neglected tropical diseases. Unfortunately, similar symptoms between the two diseases. Thus, it is possible that in emergency time, a malaria patient who has symptoms common with the COVID-19 may be misclassified as COVID-19 and vice versa. To prevent a global re-epidemic and in particular the rise in disease and death rate in Malaria-prone countries, the attention and efforts of all Health organizations and medical researchers are needed.

Results: To prevent a global re-epidemic and in particular the rise in disease and mortality in Malaria-prone countries, the attention and efforts of all Health organizations and medical researchers are needed.

Conclusion: this time when the burden of malaria is high, disorders caused by the coronavirus pandemic can increase the death rate by up to 36% in the next 5 years compared to when the virus was not present (15). To eradicate malaria, treat this invasive virus, covid-19, especially in countries where two diseases coexist, the need to upgrade health systems guidelines seems essential until together, overcome these diseases.

Keywords: malaria; covid-19; pandemic disease; re-incidence

Secondary Infection of Hydatid Cysts in Mice, *in vitro*

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Background and Aim: Cystic echinococcosis (CE) is a chronic zoonosis which presents with variable clinical manifestations. For developmental biology, drug efficacy, immunological and other analytical and descriptive studies on cysts of *Echinococcus granulosus*, two alternative tools have been developed. One is in vitro culture and the other is laboratory animal models. the aim of this study was survey secondary infection of hydatid cysts in mice.

Methods: Infected tissues with hydatid cysts were collected from local slaughterhouses of Mazandaran province. Protoscoliecs were examined for viability and washed 3 times by normal saline. In this study, mice divided in two groups. Healthy mice (n=5) and immunosuppressed mice (n=13) that received 0.01 ml Betamethasone LA. Each mouse received approximately 2000 Protoscoliecs via intraperitoneal. After 3 and 5 months, mice were dissected and visceral were subjected to microscopic analysis.

Results: Three months later in three immunosuppressed mice, 9 cysts of about 2×1 mm in the outer part of the intestine, around the kidneys and ureter were observed. In the following, after five months in two immunosuppressed mice, three cysts of about 4×2 mm around the kidneys observed. No cysts were found in healthy mice.

Conclusion: This study showed the intraperitoneal method is a suitable method for formation of secondary hydatid cyst. white mice are appropriate model for growth of secondary hydatid cysts. The secondary hydatid cyst in mice is visible from the third month.

Keywords: *Echinococcus granulosus*; hydatid cyst; Intraperitoneal; Mice

Serological Survey of *Neospora caninum* and *Toxoplasma gondii* Co-Infection in Rodents in Northwestern Iran

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Background and Aim: Our knowledge of the epidemiology of rodents' parasitic agents in Iran is scarce, although some of these pathogens play an important role in human and veterinary medicine, such as *Toxoplasma gondii* (*T. gondii*) and *Neospora caninum* (*N. caninum*). The purpose of this study was to determine the seroprevalence of *Toxoplasma gondii* and *N. caninum* in rodents of northwestern Iran between Mar and Dec 2015.

Methods: Overall, 157 serum samples from rodents (101 *Meriones persicus*, 41 *Mus musculus*, and 15 *Cricetulus migratorius*) were assayed by the indirect fluorescence antibody test (IFAT) for antibodies to *T. gondii* and *N. caninum*.

Results: We found a prevalence of 20.38% (32/157) for *N. caninum*, 35% (55/157) for *T. gondii*. Co-presence of antibodies to *N. caninum* and *T. gondii* was found in 10 (6.36%) rodents. A significant association was found between the rodents' species and seropositivity to *N. caninum*.

Conclusion: The high seroprevalence of toxoplasmosis and neosporosis in rodents in the study area has implications for translocation of these infections across wider geographical regions since these rodents are mostly preyed on by cats or dogs; hence, which can transfer the parasite to other hosts.

Keywords: Indirect fluorescence antibody test (IFAT); Iran; *Neospora caninum*; Rodents; *Toxoplasma gondii*

Serology Detection and Clinical Manifestations of Ocular Toxoplasmosis in Patients in Northwest Iran

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Background and Aim: Our goal was to use Serology Methods to verify and characterize clinical diagnoses of ocular toxoplasmosis. Clinical cases were evaluated against IgM and IgG *Toxoplasma* antibodies, and IgG avidity was tested.

Methods: A cross-sectional study was conducted in 33 patients with suspected active ocular toxoplasmosis. Patients were examined by an ophthalmologist and clinical manifestations were recorded. *Toxoplasma gondii* IgG and IgM from serum samples were analyzed by chemiluminescence immunoassay and ELISA. Acute vs chronic infection was evaluated by IgG avidity testing

Results: Twenty-eight patients (84.8%) were seropositive, two (6%) were both IgG and IgM positive, while one (3%) showed IgG avidity

Conclusion: Clinical manifestations and serological detection of *Toxoplasma* were highly correlated in the diagnosis of ocular toxoplasmosis.

Keywords: Serology; IgM; IgG; chorioretinal scarring; northwest Iran; ocular toxoplasmosis

Seroprevalence of Human Hydatidosis in Nahavand, Hamadan, Iran, 2021.

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Background and Aim: Hydatid cyst is an infectious disease with a worldwide distribution which is caused by the larval stage of *Echinococcus granulosus*. This study was aimed to investigate the seroprevalence of hydatidosis in Nahavand, Hamadan, Iran.

Methods: This cross-sectional study was carried out on 400 individuals referring to Shahid Ghodoosi and Shahid Alimoradi hospitals of Nahavand city, Hamadan, Iran. IgG antibodies against hydatid cyst, B2 antigen were evaluated by ELISA after signing the informed consent. Demographic information was recorded by using a questionnaire. Data were statistically analyzed by SPSS software.

Results: Out of 400 participants, 5 (2 females and 3 males, 1.25%) were positive for anti-*Echinococcus* antibody. One positive case (20%) was from urban, and 4 (80%) from rural population. Ninety-three percent of participants had any knowledge about hydatidosis and all 5 positive cases belonged to this group. Three of 5 seropositive subjects had the history of prolonged contact with dogs.

Conclusion: According to the result of the present study, although the seroprevalence of hydatidosis in Nahavand is lower than the other regions in the country, prevention monitoring programs of hydatidosis should be noted by health authorities due to the serious economical and medical consequences.

Keywords: *Echinococcus granulosus*; Hydatid Cyst; Seroepidemiology

Study of the Frequency of Gastrointestinal Helminth in the Feces of Sheepdogs and Watchdogs in some Rural Areas of Shirvan City, North Khorasan Province

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Background and Aim: Intestinal helminths in dogs provide a potential source of infection in humans due to the close contact between humans and dogs. Because of the limited information about helminth infection in dogs in Shirvan city, a cross-sectional study was conducted to determine the frequency of gastrointestinal helminth in the feces of sheepdogs and watchdogs in this area.

Methods: In this study, 140 fecal samples were collected from December 2020 to July 2021, and tested for helminth eggs using sedimentation technique. Characteristics of each dog including age, sex, stool consistency and history of consumption of infected viscera were recorded in the relevant registration form with a specific identification code.

Results: Overall, 39 of 140 fecal samples (27.85%) were found infected with helminths. The observed helminth eggs of fecal samples were as follows: *Taenia* (12.85%), *Dipylidium caninum* (9.28%), *Trichuris* (8.08%), *Toxocara canis* (2.94%) and Hook worm eggs (1.47%). There were no significant differences for the prevalence of gastrointestinal helminths between female (7.85%) and male (20%) and between young (2.14 %) and adult (25.71%) animals.

Conclusion: The present study showed dogs of the suburban areas of Shirvan city were infected with high prevalence of helminths, some of which can be a serious threat to public health. Therefore, it is recommended to increase the awareness of people and treat dogs with appropriate anthelmintic to reduce the risk of transmission of zoonotic parasites from dogs to humans and other animals such as livestock.

Keywords: Gastrointestinal helminth; sheepdogs; watchdogs; Shirvan

Synergistic Anti-Leishmanial Activities of Morphine and Imiquimod on *Leishmania infantum* (MCAN/ES/98/LIM-877)

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Background and Aim: This study was performed to evaluate in vitro and in vivo Leishmanicidal potential of morphine (Mph), imiquimod (IQ), and their combination

Methods: *Leishmania infantum* promastigote and amastigote assays were performed at the presence of 0.015–150μM Mph, 0.04–416μM IQ, and their combination. The inhibition effects of these drugs on promastigotes were evaluated after 24, 48, and 72h. The cytotoxic effects of the drugs were evaluated by MTT as well as flow cytometry after 72h. We explored the therapeutic effects of Mph and IQ in BALB/c mice at the end of the treatment using parasite load determination and cytokine assay. One group of mice received Mph for three weeks before infection.

Results: The results of promastigote and amastigote assays showed the cytotoxic effects of the drugs at low concentrations. The cytotoxic effects were higher on promastigotes than amastigotes ($p < 0.05$). There was a negative correlation between drug concentration and amastigote/promastigote viability. Imiquimod alone or combined with Mph showed remarkable cytotoxic effects at all concentrations ($p < 0.05$). Flowcytometry results revealed apoptosis in the parasite following exposure to the drug combinations. Accordingly, the reduction of parasite loads in the spleen and liver was observed ($p < 0.05$) with simultaneous increases in IFN- γ and IL-4. We believe that the in vivo leishmanicidal effect was mediated by Mph through IL-4 and by IQ through both IL-4 and IFN- γ .

Conclusion: Results pointed out the promising effects of Mph and IQ at low concentrations, especially when combined.

Keywords: Morphine; Imiquimod; Synergism effect; *Leishmania infantum*

The Therapeutic Effect of Ozonated Olive Oil Plus Glucantime on Human Cutaneous Leishmaniasis

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Background and Aim: Leishmaniasis is one of the main health problems in developing countries, caused by intracellular protozoan parasites of the *Leishmania* genus. Although research has been successful in discovering vaccines and anti-parasitic drugs like antimony compounds, their side effects like high toxicity, prolonged regeneration, etc., have raised the replacement importance of natural products with antioxidant and antibacterial properties. It can be said that an appropriate alternative to this is the ozonated olive oil. Ozone by introducing O₂ in involved tissues and bloodstream could degrade parasite amastigotes and lead to cleared leishmaniasis infections. So, the present study aimed to evaluate the effect of ozonated olive oil in Iranian leishmaniasis patients compared to glucantime, a choice drug for the treatment of leishmaniasis.

Methods: Thirty patients with confirmed leishmaniasis lesions were included and divided into two groups, 15 cases as control and 15 cases as test with lesions of 30-50 mm² in diameter. The control group received glucantime intralesionally and the test group ozonated olive oil plus glucantime, 2 times daily.

Results: The mean of lesion size was 50.94 Plus or minus 33.20 before and 15 Plus or minus 14.34 after treatment in control ($p < 0.00$) and 50.88 Plus or minus 31.74 before and 9.93 Plus or minus 14.18 after treatment in the test group ($p < 0.00$). Moreover, the mean course of therapy was 10.4 Plus or minus 1.84 weeks and 8.93 Plus or minus 2.15 weeks in control and test groups, respectively ($p = 0.636$). Significant differences were reported in lesion size after treatment between the two groups ($p < 0.00$).

Conclusion: Data suggested ozonated olive oil can have synergistic effects with glucantime in the treatment of cutaneous leishmaniasis.

Keywords: Amastigote; Antimony compounds; Cutaneous leishmaniasis; Glucantime; Ozonated olive oil

Therapeutic Efficacy of Nanocompounds in the Treatment of Cystic and Alveolar *Echinococcoses*: Challenges and Future Prospects

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Background and Aim: *Echinococcus granulosus* sensu lato and *Echinococcus multilocularis* are the causative agents of life-threatening cystic and alveolar *Echinococcoses* (CE and AE), respectively, which lead to serious public health concerns across the globe. Benzimidazoles (BMZs) are the drugs of choice for the treatment of human CE and AE. Presently, the chemotherapeutic failures of BMZs against CE and AE are caused by their low aqueous solubility, poor absorption, and consequently their erratic bioavailability. Among the BMZ compounds used for CE/AE treatment, albendazole (ABZ) and mebendazole (MBZ) are the only drugs licensed for human use. Nevertheless, the administration of these BMZs for a long period of time leads to undesirable adverse effects. Therefore, there is an urgent need for designing new formulations of BMZs with increased bioavailability. To bridge these therapeutic gaps, nanoparticle enantiomers of ABZ and drug delivery systems based on nanostructured entities currently provide an interesting new formulation of already existing drugs to improve the pharmacokinetic effects of BMZs. This study provides an overview of the tested nanocompounds against *Echinococcus granulosus* and *Echinococcus multilocularis*, including their effective dose, type of nanoparticles (NPs), assay setting, and therapeutic outcomes.

Methods: In order to select the papers reporting nanocompounds and their therapeutic efficacy for the treatment of CE and AE, 40 papers were addressed based on six bibliographic databases including Science Direct, Medline, Scopus, PubMed, Google Scholar, and ProQuest between 1993 and May 2019. Finally, 27 articles met our eligibility criteria (various types of NPs and nanodrugs tested against CE and AE *in vitro* and *in vivo* conditions) and were thus included in the current study.

Results: This review suggests that BMZ derivatives loaded in NPs can significantly improve the scolicidal and cysticidal activities compared with single BMZ. Moreover, BMZ-loaded polymeric NPs show a tendency to increase mortality rate against protoscoleces and microcysts compared with metallic formulations, nanoemulsions, lipid nanocapsules, solid lipid NPs, liposomes, and nanocrystals.



Conclusion: In the future, the use of the newly structured entities, attained by bridging ligands to the modified surface of NPs, as well as the electromagnetically produced nanodrugs could be helpful for developing fine-tuned formulations as an alternative to the already existing drugs against these neglected parasitic infections.

Keywords: Nanocompounds; Scolicidal agents; Cystic echinococcosis; Alveolar echinococcosis; Benzimidazoles



***Toxoplasma gondii* and *Neospora caninum* In Brain Tissue of Rodents in North-West Iran**

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Background and Aim: Rodents live in proximity to humans and domestic animals. These creatures can serve as reservoir hosts for many zoonotic parasites; therefore, they increase the risk of human infections. The aim of this study was to investigate *Toxoplasma gondii* (*T. gondii*) and *Neospora caninum* (*N. caninum*) in rodents caught in Meshgin-Shahr district, Iran.

Methods: In a cross-sectional study, brain samples were collected from 70 rodents caught in Meshgin-Shahr district during March and December 2015. The specimens were examined for exposure to *T. gondii* and *N. caninum* with molecular methods.

Results: Seventy rodents were caught, including 50 *Meriones persicus*, 11 *Mus musculus* and 9 *Cricetulus migratorius*. Thirty rodents were female and 40 were males. Using PCR (B1 gene), *T. gondii* was detected in 7.1% (5/70) of the rodents while *N. caninum* was not detected. The prevalence of *Toxoplasma* infection was higher in female rodents (4.28%) compared to male rodents (2.86%), but the difference was not significant.

Conclusion: The results showed a low risk of *T. gondii* and *N. caninum* among rodents. Finally, further research is needed to understand the role of these rodent species in the transmission of the above protozoan pathogens to humans and livestock in this area.

Keywords: rodents; *Neospora*; *Toxoplasma*; prevalence; PCR; Iran

Toxoplasmosis infection in Chronic Liver Disease Patients

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Background and Aim: *Toxoplasma gondii* can affect the human liver and cause pathological changes such as hepatomegaly, granuloma, hepatitis, necrosis, and liver cirrhosis. Epidemiological studies on the association between toxoplasmosis infection and chronic liver disease are scarce. The aim of this study was to determine the rate of toxoplasmosis infection in chronic liver disease patients with HCV-positive disease and in fatty liver patients.

Methods: This cross-sectional study was performed on 150 samples prepared from three groups, including individuals with hepatitis C, persons with grade 2 and higher fatty liver disease, and patients without liver complications as a control group during 1397 and 1398. Serum and whole blood were taken from each subject for ELISA and PCR study. Toxoplasma IgG kit was used for ELISA test and two pairs of specific primers were used to amplify the *Toxoplasma gondii* GRA6 gene in Nested-PCR.

Results: Out of 50 controls, only 3 (6%) showed positive chronic toxoplasmosis. Of 50 patients with HCV +, 21 (42%) and of 50 patients with the fatty liver disease showed 17 (34%) infections with chronic toxoplasmosis. This difference in infection ratio was statistically significant.

Conclusion: The results of this study indicate an association between chronic toxoplasmosis and patients with hepatitis C and fatty liver.

Keywords: Toxoplasmosis; Hepatitis C; Fatty Liver

Useful Parasite or Harmful Parasite for Coronavirus

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Background and Aim: Intestinal parasites infection and severe acute respiratory syndrome coronavirus diseases COVID-19 are infectious diseases and can activate the immune system. Therefore, the correct diagnosis of immune activation pathways by these two can be very effective in increasing or decreasing performance on the severity of the covid-19 disease.

Methods: We searched various websites such as Scopus, PubMed, Science Direct, Web of Science, and Google Scholar databases for related research that were published between 1 January 2019 and 30 November 2021.

Results: The results showed that many factors can be effective in increasing or decreasing the relationship between parasites and coronavirus, and parasites can play a different role in this process. These factors include an efficient immune system (which varies according to the physiological conditions of the host body, age, sex, and other factors), the type of parasite, and the strain of the virus that varies in different regions according to the viral strain in the patient's body. The relationship between the parasite and the virus can be effective.

Conclusion: Due to the fact that various factors can be effective in this relationship, more research is needed to better understand this relationship.

Keywords: Intestinal parasites; Immune system; COVID-19



Medical bacteriology



Frequency of β -lactamase Genes among Uropathogenic *Escherichia coli* in Tabriz

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Background and Aim: Extended-spectrum β -lactamase (ESBL) producing bacteria have become prevalent worldwide and they are frequently associated with multidrug-resistant uropathogenic *Escherichia coli* (UPEC). The current study aimed to evaluate the presence of β -lactamase genes among Multidrug-resistance UPEC in urine culture of outpatients and hospitalized patients in Tabriz

Methods: One hundred and fifty *Escherichia coli* isolates were collected from urine culture. Fifty percent of the specimens were related to outpatients and 50% of them were related to inpatients. DNA extraction was done by boiling method. PCR was utilized to detect presence of *bla*_{CTX-M}, *bla*_{TEM} and *bla*_{SHV} genes.

Results: Out of 150 isolates, 121 (80.7%) were positive for ESBLs. Seventy (46%) of isolates contained *bla*_{CTX-M} gene, 68 (45.3%) contained *bla*_{TEM} gene and 30 (20%) contained *bla*_{SHV} gene.

Conclusion: The high frequency of β -lactamase genes among *E. coli* isolates is alarming. So, it is important to monitor ESBL-producing *E. coli* in order to avoid spread of resistant isolates in the community.

Keywords: ESBLs; UPEC; *bla*_{CTX-M}; *bla*_{TEM}; *bla*_{SHV}

An Overview of Nanoparticle Production from Plant Gums and Their Action as Antimicrobial Agents

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Background and Aim: There have been various chemicals and routines for the treatment of infections. The extensive use of antibiotics has led to serious issues including antibiotic resistance and serious side effects. The use of plant gum nanoparticles is one of the several ways that can be employed greatly for the treatment of infections and have gained so much popularity by the scientists recently due to their several advantages over chemicals including being biodegradable, biocompatible, nontoxic and having fewer side effects. Several studies have been performed recently, stressing the undeniable advantages of these substances in the treatment of illnesses compared with their chemical counterparts.

Methods: In this article, we review several studies mentioning the pros of using plant gum NPs compared to their chemical counterparts and also their creation.

Results: Out of all of the plant gums, Gum Arabia, gum Karaya, Kondagogu gum, and gum Tragacanth, Guar gum, and gum Ghatti have gathered more interest as anti-inflammatory subjects for studies by scientists because of their several pros including having more tissue bio-availability, being easy to use, etc. The use of plant gums can be limited due to a series of disadvantages but this can be untangled by using natural nanoparticles which can be synthesized via several ways including Microwave (MW)-assisted technique, ultrasonic irradiation, etc. Among various metallic or non-metallic NPs, the most frequent of them in these studies are AgNPs (silver nanoparticles) and AuNPs (Gold nanoparticles). According to these studies, AgNPs have a more bactericidal effect than AuNPs which is due to them being more bioavailable and being more of an antioxidant.

Conclusion: There are studies suggesting that these NPs have great potential in the treatment of multi-drug-resistant bacteria. Also, studies have indicated that these substances have great anti-cancer effects due to their anti-inflammatory roles.

Keywords: Gum; Nanoparticles; Antimicrobial Activity; Treatment Agents

Antibiotic Resistance Pattern and Carbapenemase Genotype of *Pseudomonas aeruginosa* Clinical Strains Behnaz Deihim^{1 *}, Parisa Masoudipour²

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Background and Aim: *Pseudomonas aeruginosa* is an opportunistic non-fermenter bacterium that causes a variety of infections. Today, the emergence of antibiotic-resistant strains is one of the problems of the international community that can make it difficult for patients to receive specific treatment. In this study, the pattern of antibiotic resistance and carbapenemase genotype of *Pseudomonas* strains of clinical samples of patients referred to Ganjavian Hospital in Dezful were investigated.

Methods: In the present cross-sectional study, 49 strains of *Pseudomonas aeruginosa* were isolated from different clinical specimens. Strains were confirmed using 16sRNA gene and microbiological susceptibility testing to aminoglycoside antibiotics, fluoroquinolones and third generation cephalosporins was performed using Kirby Bauer disk diffusion test. Sensitivity to meropenem and colistin was determined by E-test. Carbapenemase genes (IMP, VIM, NDM) were detected.

Results: The highest frequency of strains was in urine and endotracheal tubes and blood cultures, which were isolated from ICUs. Strains were most resistant to piperacillin and third-generation cephalosporins and fluoroquinolones. The most effective antibiotic was piperacillin tazobactam. Resistance to meropenem and colistin were 71% and 22%, respectively. IMP, VIM and NDM genes were observed in 6, 7 and 7 strains of carbapenemase-producing strains, respectively. Co-expressing of three genes were detected in 3 strains (2 tracheal and 1 blood cultures).

Conclusion: Due to the prevalence of resistant strains to the third generation of cephalosporins and carbapenem and the simultaneous expression of 3 carbapenemase genes in this study, continuous monitoring of hospital strains and rational prescription of antibiotics is essential.

Keywords: *Pseudomonas aeruginosa*; Antibiotic resistance; Carbapenemase Genes

Antibiotic-resistance Patterns and Toxin Profiles among *Clostridioides difficile* Strains Isolated from Hospitalized Patients with Diarrhea in Hamadan Hospitals

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Background and Aim: Today, concerns over *Clostridioides difficile* (*C. difficile*) infection (CDI) have significantly increased due to reduced susceptibility to antibiotics used for CDI treatment. Toxins produced by *C. difficile* strains are associated with disease severity and outcome. This study was designed to investigate the prevalence of *C. difficile*, its toxin-producing genes, and antibiotic resistance patterns in diarrheal samples from hospitalized patients in Hamadan, Iran.

Methods: In this cross-sectional study, a total of 130 diarrheal samples of patients admitted to different wards of three hospitals in Hamadan from November 2018 to September 2019 were collected. *C. difficile* isolates were identified by culture on CCFA and PCR (Polymerase chain reaction). The presence of toxin-encoding genes (*tcdA* and *tcdB*) and binary toxin genes (*cdtA* and *cdtB*) was analyzed by PCR. Resistance of the isolates to metronidazole, vancomycin and clindamycin antibiotics was determined using the agar dilution method

Results: Out of 130 diarrheal samples from hospitalized patients, 16 (12.3%) *C. difficile* isolates were obtained. PCR results were positive for two toxin-producing genes, *tcdA* and *tcdB*, in all (100%) *C. difficile* isolates, and the binary toxin genes *cdtA* and *cdtB* were detected in 6 (37.5%) and 8 (50%) isolates, respectively. The results of antibiotic susceptibility testing showed resistance to metronidazole, vancomycin, and clindamycin in 3 (18.7%), 3 (18.7%), and 2 (12.5%) isolates, respectively, and all isolates were resistant to rifampicin.

Conclusion: The results of this study showed toxigenic *C. difficile* with *tcdA*+/*tcdB*+ profile is a major cause of nosocomial diarrhea in Hamadan, and clinical laboratories should routinely perform *C. difficile* diagnostic testing on diarrheal specimens of hospitalized patients. Resistance to conventional antibiotic therapy against *C. difficile* should be considered as a warning to prevent irrational administration of antibiotics.

Keywords: Antibiotic resistance; Binary toxin; *Clostridioides difficile*; TcdA; TcdB

Antimicrobial Resistance Profile of Gram-negative Bacteria Isolated from Blood Samples of Children with Bacteremia During Covid-19 Pandemic in Mofid Children Hospital, Iran

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Background and Aim: Emerging of antimicrobial resistance (AMR) isolates is known as a challenge for public health systems, because they are able to increase the prevalence of resistance genes through other bacteria and limit therapeutically options. In this study we focused on antimicrobial resistance patterns of gram-negative bacteria (GNB) isolated from blood samples of children with bacteremia during covid-19 pandemic in Iran.

Methods: We prepared blood samples of children with bacteremia by Becton Dickinson Diagnostic Instrument Systems, Sparks, Md (BACTEC) system. We used selective culture MacConkey and biochemical tests, fermentation of sugars, IMViC and urease activity as the standard protocols. Resistance isolates were determined against imipenem, meropenem, gentamycin, ceftazidime, levofloxacin, cefepime, aztreonam and tigecycline using disc diffusion method according to the latest updates of Clinical and laboratory standard institute (CLSI) 2021.

Results: A total 255 GNB strains collected from children with bacteremia who were hospitalized during covid-19 pandemic. Out of 255 GNB strains, 43.52% (111/255) of them were Enterobacteriaceae spp., 33.72% (86/255) of them were Pseudomonas spp. and 22.74% (58/255) of them were Acinetobacter spp. The antimicrobial resistance frequency of Enterobacteriaceae spp., Pseudomonas spp. and Acinetobacter spp. were like: imipenem 33.33%, 66.27%, 84.21% (37/111, 57/86, 48/58), meropenem 39.63%, 86.6%, 87.93% (44/111, 59/86, 51/58), gentamycin 39.63%, 55.81%, 63.79% (44/111, 48/86, 37/58), ceftazidime 40.54%, 77.9%, 91.37% (45/111, 67/86, 53/58), levofloxacin 38.73%, 24.41%, 37.93% (43/111, 21/86, 22/58), cefepime 38.73%, 65.11%, 86.2% (43/111, 56/86, 50/58), and aztreonam 34.23%, 63.95%, 87.93% (38/111, 55/86, 51/58), respectively.

Conclusion: The most frequent GNB isolated from blood samples of children with bacteremia during covid-19 pandemic belonged to Enterobacteriaceae spp., and these strains showed the most resistance activity against gentamycin. The high frequency of GNB bacteremia which are resistance to multiple antibiotics in children could put them in life threatening risks.

Keywords: GNB; antimicrobial resistance; bacteremia; BACTEC

Bacterial Co-infection and Detection of the Predominant Beta-lactamase Genes and AmpCs in Patients with COVID-19

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Background and Aim: Bacterial co-infection in patients hospitalized, with COVID-19 are an important and challenging complication. However, biological findings in patients with COVID-19, confirms an associated bacterial co-infection, but antibiotic therapy and role of multidrug-resistant bacterial, in COVID-19 is not very transparent.

Methods: Two hundred and seventy patients enrolled from May 22 to June 27, 2021, included all of 270 patients in intensive care unit (ICU). Nasopharyngeal and oropharyngeal swab samples were collected by trained personnel and processed at Reference Molecular Laboratory Yasuj (Iran) using real-time polymerase chain reaction (RT-PCR) kit. For detection of bacterial infection all of 270 samples were cultured on Blood Agar, Eosin Methylene Blue (EMB) and MacConkey Agar. Antimicrobial susceptibility assays were performed according to the CLSI guidelines. Also, genetic identification of bacteria was performed by analyzing the 16S rRNA gene.

Results: fifty-seven (21.5%) of 270 samples showed bacterial infection, 44 (16.5%) gram negative bacteria and 13 (5%) gram positive bacteria. The most abundant bacteria that were isolated from the samples was *Acinetobacter baumannii* (n=18, 31%,). Also, the analysis of antimicrobial resistance for all isolates shows that the multidrug-resistant phenotype (MDR). They showed the highest resistance to imipenem (86.4%).

Conclusion: The bacterial co-infection in patients with COVID-19 is serious and is associated with the progression of COVID-19 disease and induction of resistance to the most of antibiotics.

Keywords: Bacterial co-infection; COVID-19; *Acinetobacter baumannii*; antimicrobial resistance

Bacterial Co-Infections in Coronavirus Disease: a Review

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Background and Aim: COVID-19 is an emerging viral disease caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). SARS-CoV-2 infects more people, especially the elder population around the world than other coronavirus, such as SARS-CoV and MERS-CoV which is considered to be a challenge for the health system. SARS-CoV-2 has led to more than 100 million cases and 2 million deaths globally in 12 months. Beside the pathogenesis of SARS-CoV-2, bacterial co-infection plays a significant role in the occurrence and development of SARS-CoV-2 infection by raising the difficulties of diagnosis, treatment, prognosis of COVID-19, and also it is possible that some patients might die from bacterial co-infection rather than the virus itself. In this research we studied the bacterial co-infection in COVID-19.

Methods: This research was based on articles which were published from 2019 to 2021. PubMed, Scopus, and google scholar were searched using keywords: "COVID-19", "SARS-CoV-2", and "bacterial co-infection".

Results: When bacterial coinfections did occur, *Streptococcus pneumoniae*, *Haemophilus influenza*, *Legionella pneumophila*, and *Staphylococcus aureus* were the most common pathogens. Atypical bacteria were very rare. *Mycobacterium tuberculosis*-COVID-19 coinfections are also reported. Additionally, patients admitted in the intensive care units have increased rates of bacterial coinfection, and those diseases can be due to super-infections by nosocomial antibiotic-resistant bacteria.

Conclusion: Co-infections are rare in patients with Covid-19, and diagnostic tests are not necessary for most patients admitted with Covid-19, but physicians should be wary of bacterial infections because co-infection in Covid-19 patients may worsen the outcome of the disease, and timely identification simultaneous infections can help improve the health of Covid-19 patients during epidemics.

Keywords: COVID-19; SARS-CoV-2; bacterial co-infection

Biocides' Resistance Mechanisms Intervening in the Dissemination of *Acinetobacter baumannii*: Another Grave Apprehension in Health-care Setting

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Background and Aim: In healthcare setting, *Acinetobacter baumannii* (*A. baumannii*) is a well-known bacterial species to cause multidrug-resistant nosocomial infections. The organism survives on dry surfaces and can be found on various inanimate objects. Biocides are presumed to have multiple antibacterial targets reducing the likelihood of bacteria developing resistance to these compounds however, outbreaks of nosocomial MDR *A. baumannii* infections are not uncommon. *A. baumannii* has been found to be resistant to even hand sanitizers, which is a serious matter of consideration in COVID-19 period. This review attempted to discuss resistance mechanisms developed by *A. baumannii* towards some of the biocides.

Methods: Google scholar, PubMed, Science direct and Scopus research engines were selected to search the best literature research. 'Name of the biocide', 'reduced susceptibility', 'resistance', 'tolerance', '*Acinetobacter baumannii*' and 'minimum inhibitory concentration (MIC)' were used for each search. The search constraint from 2013 to till date.

Results: Biocides are grouped into several categories, depending on the functional group they possess, the mode of action or their target. Mechanisms related to resistance comprise of cellular changes on biocide accumulation, cell envelope changes, limited uptake, and efflux mechanisms expression. According to the reports, typical disinfectants including 70% ethanol, chlorhexidine, sodium hypochlorite and quaternary ammonium compounds (QACs) are effective against *A. baumannii* isolates if used at the manufacturers' recommended concentrations. Presence of efflux pumps and genetic causes of disinfectant resistance is amongst the most common cause of disinfectant resistance especially the *qac* genes (*qacE* and *qacEΔ1*) in *A. baumannii*. Biofilm formation on many medical devices is another mechanism used by *A. baumannii* strains to prevent the efficacy of disinfectants.

Conclusion: Number of biocides have been explored nevertheless, precise information about the MIC breakpoints has not been recommended. All environmental isolates must be monitored regularly, and their sensitivity to disinfectants must be assessed.

Keywords: *Acinetobacter baumannii*; Biocide; Resistance

***Chlamydia trachomatis* and *Mycoplasma genitalium* Infections on IVF Outcomes among Women Referred to Razavi Hospital and Milad Infertility Center, Mashhad, Iran.**

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Background and Aim: *Chlamydia trachomatis* (*C. trachomatis*) and *Mycoplasma genitalium* (*M. genitalium*) are the leading causes of women's genital tract. These two bacteria, cause urethritis, cervicitis, and pelvic inflammatory disease, leading to infertility in women. This study was performed to determine the prevalence of these two infections in infertile women and their effect on IVF treatment failure.

Methods: In this study, cervical swab sampling was performed from 100 infertile patients referred to the infertility treatment center of Razavi hospital and Milad infertility center in Mashhad. The swabs were immediately placed in Screw Cap Test Tubes containing thioglycollate medium and transferred to the laboratory. The incubation was performed at 37 °C for 18 h, and the tubes were centrifuged. The resulting residue was transferred into the microtubule, and after three washes, DNA extraction was performed by boiling. Finally, the supernatant was used separately for PCR (Uniplex PCR) with specific primers for each bacterium.

Results: The results showed that 23 samples were positive for *C. trachomatis*, and most cases of IVF failure were related to this group (17 points). Only one specimen was positive for *M. genitalium*, associated with IVF failure patients.

Conclusion: Considering the prevalence of *C. trachomatis* and *M. genitalium*, screening is essential in infertile women suffering from relatively severe conditions and high infertility treatment costs, including IVF. So, it is suggested that before any infertility treatment, the patients be monitored for these bacteria and receive antibiotic therapy if the result is positive.

Keywords: *Chlamydia trachomatis*; *Mycoplasma genitalium*; Infertility; IVF; PCR

Comparison of Prevalence and Antimicrobial Resistance Patterns of Gram-positive Cocci in the Nostrils of Healthcare Workers in a Pre- and Post- Covid-19 Era in North Khorasan, Iran

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Background and Aim: In the COVID-19 era, co-infections can lead to an increase in morbidity and mortality. Normal flora bacteria can transfer to the pulmonary tract and create bacterial co-infections. The nasal cavity is one of the main areas with normal flora in the human body. In this study, we evaluated the prevalence and antibiotic resistance of gram-positive Cocci in the pre- and post-COVID-19 era in healthcare workers. We evaluated 376 nasal swabs of pre-and 376 swabs of the post-COVID-19 era. The most common gram-positive Cocci in the first samples were *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Staphylococcus capitis*, *Staphylococcus hominis*, *Staphylococcus haemolyticus*, *Streptococcus pneumoniae*, and *Enterococcus faecalis*, and in the second samples were *S. aureus* and *S. epidermidis*, *S. warneri*, *S. hominis* and *E. faecalis*, respectively. We found resistance to linezolid and vancomycin in *S. aureus*, *S. epidermidis*, and *S. hominis*. We also found a high resistance rate to antiseptics in our isolates.

Methods: We evaluated 376 nasal swabs of pre-and 376 swabs of the post-COVID-19 era.

Results: We found resistance to linezolid and vancomycin in *S. aureus*, *S. epidermidis*, and *S. hominis*. We also found a high resistance rate to antiseptics in our isolates.

Conclusion: The increasing linezolid resistance in bacterial strains becomes a real threat for patients, and monitoring such infections combined with surveillance and infection prevention programs is very important to decrease the number of linezolid-resistant staphylococcal strains.

Keywords: COVID-19; Co-infection; Gram-positive Cocci

Concomitant or Secondary Bacterial Infections in Hospitalized COVID-19 Patients

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Background and Aim: Secondary bacterial pneumonia or concomitant bacterial infection, though rare, but can occur in hospitalized patients with COVID-19 following the initial phase of a viral respiratory infection or in the recovery phase. The management approach in COVID-19 should ideally consider the overall manifestations and course of the disease. This review attempted to discuss the prevalence of concomitant bacterial infections and the type of etiological agent/s reported at various places in hospitalized patients so as to prevent, diagnose and treat the cases appropriately.

Methods: Four search engines were selected to search the best literature research- Google scholar, PubMed, Science direct and Scopus. “COVID-19”, “infections”, “bacterial”, “pneumonia”, “secondary infections”, and “concomitant infections” were used for each search.

Results: Reports shows that 1% hospitalized patients with Covid-19 were afflicted with concomitant or secondary infection. Most likely infection developed in these cases was bloodstream infection. The most common organisms were *Escherichia coli*, *Staphylococcus aureus* and *Klebsiella pneumoniae*. Cases of concurrent / secondary infection were older (mean 70 years) vs. 55 years, and a higher proportion of cases included black or Asians. Late infections (more than 48 hours after admission) with gram-positive bacteria were more common in COVID-19 patients. Similarly, cytokine storms and secondary bacterial infections are thought to be responsible for the development of acute kidney injury in COVID-19 patients. Some infections by drug-resistant organisms have also been discussed in patients with COVID-19, such as *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, multidrug-resistant *Escherichia coli*, *Enterococcus* spp. and *Acinetobacter baumannii*. Cephalosporins, carbapenems, and quinolones were the good early choices for COVID-19 patients, whether in the form of single treatment or combination along with antiviral agents.

Conclusion: Concomitant or secondary bacterial infection is a significant complication which can have worse outcomes in COVID-19. Careful monitoring and prompt antibiotic treatment may be helpful for selected patients.

Keywords: Secondary infections; Bacterial; Concomitant; COVID-19

Current Trends in Colistin Susceptibility Testing Methods in Enteric Gram-negative Bacilli and Other Non-fermentative Pathogens

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Background and Aim: Polymyxins, an age-old sort of cationic polypeptide antibiotic, are regarded as final-resort drugs in handling infections caused by multidrug-resistant Gram-negative bacteria. An exact assessment of colistin susceptibility is crucial due to increasing colistin resistance and absence of other antibiotics in curing multidrug resistance pathogens. Both Clinical and Laboratory Standards Institute and European Committee on Antimicrobial Susceptibility Testing have investigated colistin broth microdilution and offer particular recommendations, but this method has a challenging perform in routine daily practice. It is time consuming and requires ≥ 48 h. In this issue, several alternative rapid tests are developed such as ResaPolymyxin Acinetobacter/Pseudomonas NP. The sensitivity and specificity of 100% for the Rapid ResaPolymyxin Acinetobacter/Pseudomonas NP test to Enterobacterales have been reported. Colistin broth disk elution test and colistin agar test are another novel potential methods that were validated by the Clinical laboratory standard institutes subcommittee for colistin screening in the Enterobacterales and *Pseudomonas aeruginosa*. In addition, recently, Rapid flow cytometry (FASTinov) method was introduced which requires only 2 h compared with conventional methods. This review will take a look at current trends on methods for colistin resistance measurements.

Results: Nowadays, both EUCAST and CLSI subcommittee awarded suggestion verifying that broth microdilution (BMD) has been until now the only applicable method. Disk diffusion does not work because of the poor diffusion of the large colistin molecule. E-test is unreliable by giving warning from EUCAST and other studies.

Conclusion: The review of the included articles has revealed that the emergence of plasmid-mediated (*mcr* genes) and chromosomally encoded colistin resistance is considered a serious problem due to a lack of alternative antibiotics and reliable methods for detecting colistin resistance and susceptibility in routine laboratories are needed, this is not only critical for optimum susceptibility testing, but also helps to guide decision-making for patient treatment.

Keywords: Gram-negative bacteria; antimicrobial susceptibility testing; colistin; *in vitro* activity; method

Detection of *GES* and *VEB* Extended Spectrum β -lactamase Genes in *Klebsiella oxytoca* in Khorramabad, Iran

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Background and Aim: *Klebsiella oxytoca* (*K. oxytoca*) is an immobilized gram-negative bacillus, aerobic, voluntary anaerobic, oxidase negative, belongs to the Enterobacteriaceae family and is considered as an opportunistic pathogen. *K. oxytoca* like other members of the Enterobacteriaceae family, may acquire genes encoding resistance, including extended spectrum beta-lactamases (ESBL), metallo- β -lactamases, and carbapenemases. The aim of this study was detection of *VEB* and *GES* genes in *K. oxytoca* isolated from clinical specimens in Khorramabad, Iran.

Methods: *K. oxytoca* collected from hospitals of Khorramabad, Iran from January 2019 to January 2020. Then ESBL-producing strains were isolated by disc diffusion method according to CLSI protocol. After DNA extraction from all *K. oxytoca* clinical strains *VEB* and *GES* genes detected by PCR method.

Results: The present study was a cross-sectional study, 32 *K. oxytoca* were collected randomly. Of these, 21 strains (65.6%) were isolated from women and 11 strains (34.4%) were isolated from men. The majority of people were sampled in the age group (41-60). The mean age of the subjects was 22.18 ± 46.16 years. Most strains were isolated from urine samples with frequency of 87.5% and then from wounds and sputum with a frequency of 6.3% each. Among 32 isolates, 56.6% (N = 21) of isolates were ESBL positive. Also, the results showed that none of isolates were carried *VEB* and *GES* genes.

Conclusion: The results of the present study indicate the spread of ESBLs and the absence of *VEB* and *GES* genes, which requires more extensive studies to identify other ESBL genes.

Keywords: Extended Spectrum B-Lactamase (ESBL); *GES*; *VEB*; PCR

Detection of *H. pylori* in Tissues with Benign Prostatic Hyperplasia Isolates from Hospitalized Patients in Qom, Iran

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Background and Aim: *Helicobacter pylori* (*H. Plyori*) is an important cause of human infections. There is accumulating evidence relating to *H. pylori* infection to urological diseases. The aim of this study was to investigate the presence of *H. pylori* infection in prostate tissues of patients with benign prostatic hyperplasia (BPH).

Methods: In this descriptive cross-sectional study the number of 160 patients with benign prostatic hyperplasia, referred to Beheshti and Kamkar hospital between October 2015 and March 2016 were selected using the connivance sampling method. Open surgery with TURP method (Transurethral resection of the prostate) was performed on them and their prostate sample was delivered to lab for testing. DNA extraction of all collected tissues was carried out using PCR. Data were analyzed using descriptive and central statistics with spss 23 software.

Results: The mean age of patients was 67.4 ± 6.28 (range 50-85) years. After DNA extraction of 160 prostate tissue samples, *H. pylori* was detected only in three specimens (1.83%).

Conclusion: According to the result of the study, *H. pylori* infection cannot be reported as an effective cause of BPH.

Keywords: *Helicobacter pylori*; Polymerase Chain Reaction; Prostatic Hyperplasia

Doxycycline-encapsulated Solid Lipid Nanoparticles for the Enhanced Antibacterial Potential to Treat the Chronic Brucellosis and Preventing Its Relapse: *in vivo* Study

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Background and Aim: Brucellosis is one of the most important infection of diseases. Due to its large period of treatment and survival ability of bacteria inside the macrophages, relapse of this disease is the main challenge, especially, after the treatment. The current study was carried out to evaluate the antibacterial effect of solid lipid nanoparticles loaded with doxycycline on the *Brucella melitensis* in *in vivo* conditions.

Methods: The double emulsion synthesized doxycycline-encapsulated solid lipid nanoparticles (DOX-SLN) was characterized using DLS and FE-SEM. The efficacy of the DOX-SLN on the acute and chronic Wistar rat infected brucellosis was investigated. The pathological assessments were made on the spleen and liver in the treated rates.

Results: The *in vivo* experimental results demonstrated that the treated rats with DOX-SLN had significantly decreased the *B. melitensis* CFUs in their spleen and liver compared to that of the treated rates with free doxycycline and untreated ones. The pathologic results indicate that the improvement trend of spleen and liver tissues in rats treated by DOX-SLN was satisfactory.

Conclusion: According to *in vivo* results, the DOX-SLN has better effects on the treatment of chronic brucellosis. Therefore, DOX-SLN is recommended to treat the brucellosis and avoid its relapse.

Keywords: *Brucella melitensis*; Doxycycline-encapsulated solid lipid nanoparticles; *in vivo*; Spleen; Liver; Relapse

Establishing Artificial Intelligence and Reducing Health-care System Costs Respond to COVID-19

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Background and Aim: The application of artificial intelligence (AI) and machine learning (ML) digital software has become part of our everyday lives, and the related algorithms are becoming part of our everyday laboratory procedures. The scientists present AI and ML to find better approaches to progress clinical microbiology practice.

Methods: This study was done in order to integrating these practices into local, national, and international healthcare systems, which will save lives, and suggest specific methods by which implementation can happen swiftly and efficiently. For accessing to relevant articles and studies, a search was done in main databases and studied in this field.

Results: ML algorithms are focused on general predictions based on previous examples. The hypothetical advantages of the AI approach in Real time PCR to identify closely related serotypes are twofold increased speed and lower cost, when compared to alternative existing methods. Healthcare settings are in an urgent need for decision-making technologies to handle SARS-CoV-2 and help them receive the right proposals in real-time to avoid its spread. The ongoing development in AI and ML try to works skillfully to mimic human intelligence. It may also play an important role in understanding and proposing the development of the medication, prediction, screening, contact tracing, forecasting and drug/vaccine for the Covid-19 pandemic and reduce the human intervention. Training in image-based infectious disease diagnostics relies on image data sets curated with great effort through manual interpretation of images by skilled clinical microbiologists. After training, accuracy and robustness must be extensively verified under real-world conditions in comparison to diagnostic interpretation by trained microbiologists. However, after this resource-intensive training, validation and implementation, AI-based diagnosis is essentially free, and image analysis can be performed on lower-end computers that already exist in most microbiology laboratories.

Conclusion: Therefore, AI-based image analysis in infectious disease diagnostics will likely find its niche by providing answers in healthcare systems both large and small where other techniques are not cost-effective, immediate or comprehensive. It is also evident that AI-assisted image analysis can be performed at a distance, only requiring that the remote site have a microscope and a way to transmit images to the AI using the internet or cellular service

Keywords: Artificial intelligence; COVID-19

Evaluation of Antibacterial Potential of *Rosmarinus officinalis* Essential Oil Green Nanogel against Clinical and Standard Bacterial Strains

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Background and Aim: *Staphylococcus aureus* and *Pseudomonas aeruginosa* are important pathogens that cause many infections such as skin diseases in humans. Today, antibiotic resistance is an important challenge for health care systems worldwide; Because of this, production of new antibacterial agents have become crucial.

Methods: In our study, antibacterial potential of *Rosmarinus officinalis* essential oil was investigated; ingredients of *Rosmarinus officinalis* essential oil were identified as three major ingredients: α -pinene, β -pinene and 1,8-cineole. A nanogel-based component of *Rosmarinus officinalis* was prepared and physically impregnated on the surface of the paper mats. Antibacterial effects of the prepared prototype at three concentrations (2.5, 5, and 10 mg/mL) with exposure times of 24 h were investigated against clinical and standard strains of *Pseudomonas aeruginosa* and *Staphylococcus aureus*.

Results: At 2.5, 5, 10 mg/mL concentration with 24 h exposure, the growth of a standard strain of *S. aureus* 100, ~ 95, ~ 90 % was decreased, respectively. growth of other strains shown no significant reduction in the growth rate.

Conclusion: The prepared nanogel could thus be used as a green potent antibacterial agent candidate as cleaning pad, wound dressing and others.

Keywords: *Rosmarinus officinalis*; essential oil; antibacterial

Evaluation of Carbapenem Resistance and Class D β -lactamase Genes in *Acinetobacter baumannii* Isolates from Hospital Environments

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Background and Aim: In current study the antimicrobial susceptibility patterns and the prevalence of class D β -lactamase genes were determined in *Acinetobacter baumannii* (*A. baumannii*) isolates from hospital environment.

Methods: Fifty *A. baumannii* isolates were collected from different surfaces and equipment of hospital and confirmed by PCR -sequencing of *rpoB* gene. The susceptibility to different antibiotics were measured by disk diffusion and broth microdilution methods. Class D β -lactamase -encoding genes were characterized by PCR.

Results: *A. baumannii* was detected in 22.1% of environmental surfaces and equipment. Antibiotic susceptibility results indicated 100% of isolates were imipenem resistant and colistin and tigecycline susceptible. All (100%) of *A. baumannii* isolates were recognized as MDR and XDR. The frequency of *bla*_{OXA-51-like}, *bla*_{OXA-23-like}, *bla*_{OXA-24/40-like} was 100%, 94%, and 92% respectively. None of the isolates possessed the *bla*_{OXA-58-like} or *bla*_{OXA-143-like} genes.

Conclusion: Our results indicated that the most frequent OXA-type carbapenemases identified in the studied environmental *A. baumannii* isolates was the OXA-23-like.

Keywords: *Acinetobacter baumannii*; Hospital environments; Class D β -lactamase; Carbapenems

Evaluation of *Clostridium difficile* Colonization in Hospitalized Children with Malignancy in Hematology/Oncology Ward

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Background and Aim: Children who are exposed to chemotherapy can be constantly hospitalized for various infectious agents such as *Clostridium difficile* (*C. difficile*). This study was performed on sick children referred to one of the pediatric hospitals in Khorasan Razavi province who referred to the hematology / oncology department for chemotherapy during 2016 and 2017.

Methods: The questions in the checklist and consent form were prepared from the patients' parents. To evaluate the possibility of colonization of *C. difficile* bacteria, fecal samples were taken from the patient twice: the first sample on the first day of hospitalization and the second sample before the patient was discharged. The minimum length of stay was considered to be at least 3 days. Children's stool samples in cases of diarrhea were transferred to the laboratory of the study site by stool swab method and in non-diarrheal cases, complete stools were taken for up to one hour. Using GDH kit, the possibility of bacterial colonization was examined within 20 minutes and the test result was obtained in two negative or positive answers. A total of 100 patients in different age groups who were hospitalized in the children's hospital for at least 3 days were examined. With *C. difficile* agent, history of surgery, history of use of gastric acid inhibitors, type of ward admitted and had some clinical symptoms such as fever, diarrhea and vomiting.

Results: The results showed that 30% of children were colonized with *C. difficile* after admission. While 10% of children were carriers of *C. difficile* in their stool samples at the beginning. 60% of the studied children also survived the disease without colonization. This study showed that the rate of *C. difficile* colonization in hospitals with children with malignancy is another nosocomial infection in pediatric wards

Conclusion: The researchers of this project recommend washing hands and observing hospital hygiene.

Keywords: *Clostridium difficile*; colonization; hospitalized children; malignancy

Evaluation of Frequency of Mobile Genetic Elements (Transposons & Insertion sequences) in Class D carbapenemase Producing *Acinetobacter* spp. Isolated from Clinical Specimens among Kerman Hospitals

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Background and Aim: *Acinetobacter* is one of the most common hospital pathogens that is widespread in the intensive care units (ICU). According to previous studies presence of carbapenem-hydrolyzing Ambler class D β -lactamase is a common mechanism of antibiotic resistance in this bacterium. The spread of OXA carbapenemase in *Acinetobacter* has become a growing concern all over the world. The aim of this study is evaluation of frequency of mobile genetic elements (Transposons & Insertion sequences) in class D carbapenemase-producing *Acinetobacter* spp. isolated from clinical specimens among Kerman hospitals and to detect frequency isolates that were containing ISAb₁ in upstream of *bla*_{OXA} gene.

Methods: One hundred and eight clinical *A. baumannii* isolates were collected from clinical specimens of Kerman hospitals, Iran, from May 2018 to October 2019, Antimicrobial susceptibility testing was determined using a disc diffusion method; minimum inhibitory concentration (MIC) was measured by micro broth dilution test. PCR were conducted for the identification of *bla*_{OXA} (51,58,23,24) and ISAb₁ (1,2,3,4), IS18 and Tn (2006, 2007, 2008, 2009) genes and detect the presence of ISAb₁/*bla*_{OXA} 51, ISAb₁/*bla*_{OXA23}, ISAb₄/*bla*_{OXA23} genes.

Results: The results showed that 100% of the isolates were resistant, to carbapenems. In these isolates, the genes of OXA51, ISAb₁, Tn2008 had most frequently with 96.5%, 96.5%, and 86.7%, respectively, additionally ISAb₄, IS18, Tn2007, Tn2009, ISAb₄, and OXA23 genes were not found in any isolates. The frequency of clusters in ISAb₁/*bla*_{OXA-51} and ISAb₁/*bla*_{OXA-23} genes was identification in 26.5% and 88/5% of isolates, respectively.

Conclusion: The results of this study showed the importance of transposable elements in the spread of resistance to carbapenems and the development of resistant genes.

Keywords: OXA Carbapenemase; Insertion sequence; Transposon; Extensively drug Resistance; *Acinetobacter baumannii*;

Evaluation of Procalcitonin Serum Level with Clinical Pulmonary Infection Score Index in Patient with Possitive Culture of Pulmonary Secretion in Intensive Care Units (ICU) in Beheshti Hospital 2020-2021

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Background and Aim: Ventilation Associated Pneumonia (VAP) due to bacterial infection is the most common cause of a new infiltration in patients undergoing mechanical ventilation, which is associated with the clinical condition of new infiltration or progression of previous infiltration with fever, leukocytosis or leukopenia and purulent tracheobronchial discharge. The aim of this study was to Evaluate the procalcitonin serum level with Clinical Pulmonary Infection Score Index in patient with positive culture of pulmonary secretion.

Methods: In a descriptive cross-sectional study, 92 patients admitted to the intensive care unit of Shahid Beheshti Hospital in Hamadan during the years of 2020-2021 with suspicion of VAP in terms of mean serum level of procalcitonin with clinical criteria Pulmonary infection, age, sex, length of hospital stay, mean arterial pressure, and time interval between intubation and pneumonia were assessed. After registering in the checklist, patient data were analyzed with SPSS software version 22 at 95% confidence interval.

Results: The mean age of patients was 66.62 ± 16.93 years and 56.5% were male and 43.5% female and the mean serum procalcitonin in total was 4.29 ng/l. in patients with CPIS higher than 6 and 6 and les, it was 4.79 ± 1.21 and 0.21 ± 0.14 respectively ($P < 0.001$). There was a significant correlation between serum procalcitonin level with duration of hospitalization ($P = 0.048$), duration of intubation ($P = 0.039$) and WBC count ($P = 0.004$); But there was no significant relationship between serum procalcitonin level with CRP, hospital outcome, endotracheal culture and vasopressor uptake.

Conclusion: In patients with positive culture of intubated pulmonary secretions in ICU, serum level of procalcitonin has a significant correlation with the clinical criteria of lung infection. Therefore, serum procalcitonin can be used as a reliable method for the diagnosis of VAP.

Keywords: Procalcitonin; Intubation; Clinical criteria; lung infection

Extended Spectrum β -lactamase Production in Uropathogenic *Escherichia coli*

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Background and Aim: The presence of extended-spectrum β -lactamase (ESBL) producing *Escherichia coli* have become prevalent worldwide and are frequently associated with multidrug resistance. The current study aimed to evaluate antimicrobial patterns of Uropathogenic *Escherichia coli* isolates in Tabriz.

Methods: One hundred and fifty *Escherichia coli* isolates were collected from urine cultures. Bacterial identification and antimicrobial susceptibility testing was performed using established methods. Antimicrobial susceptibility tests were performed with disc diffusion tests. ESBL production was phenotypically confirmed by double disk synergy (DDS) test.

Results: The majority of ESBL producing isolates were resistant to co-trimoxazole, amoxicillin, ciprofloxacin. Sixty percent of the isolates were MDR. The phenotypic ESBL rate was 45.3%.

Conclusion: This study demonstrated that multidrug-resistant *Escherichia coli* isolates in Tabriz is high. The isolates showed the highest susceptibility to imipenem, phosphomycin, nitrofurantoin. Therefore, these antibiotics can be useful for treating *Escherichia coli* infections.

Keywords: ESBL; imipenem; UPEC; MDR

Fecal Colonization, Phenotypic and Genotypic Characterization of ESBL-producing *Escherichia coli* Isolates in Transplant Patients in Shiraz Nemazee and Abu Ali Sina Hospitals

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Background and Aim: The current work aimed at investigating the intestinal carriage of Extended-spectrum β -lactamases (ESBLs)-producing *Escherichia coli* (ESBL-EC), determining the clonal relatedness, phylogroups and adhesion factors among ESBL-EC strains obtained from rectal swabs from transplant patients in a tertiary care teaching hospital.

Methods: This cross-sectional study was conducted during February to August of 2020 in Shiraz, Iran. Rectal swabs (RS) were randomly collected from transplant patients. The combination disk was used as a phenotypic method to detect ESBL-producing strains. PCR was performed for the genotypic detection of ESBL and adhesion factors, classify the phylogenetic group of each ESBL-EC strain and major sequence types (STs).

Results: A total of 118 *Escherichia coli* isolates were collected from rectal swab from hospitalized transplant patients. Among them, 37.9% (49/118) patients were colonized with ESBL isolates. The majority of the isolates were from kidney transplant patients (29/49, 59.2%). Overall, of the 49 isolates identified as ESBL-producers, 42 (85.7%) isolates harbored the *bla*_{CTX-M} gene. The detection of the different pathotypes from 49 ESBL-producer isolates showed that the most prevalent pathotype was STEC (32.7%). According to the multiplex PCR results, most (42.8%) of the ESBL-producer isolates belonged to phylogroup B2. The most frequent sequence type (ST) among *Escherichia coli* isolates was ST131 (26.5%).

Conclusion: Our study revealed high production of ESBL-encoding genes with *bla*_{CTX-M} as the most predominant gene, moreover, it revealed high intestinal colonization with ST131 ESBL-EC. Therefore, to reduce and prevent the spread of ESBL-EC, early detection of fecal carriage at the beginning of admission in clinical settings especially in immunocompromised patients is recommended.

Keywords: *Escherichia coli*; Fecal colonization; ESBL; Transplant patient

Frequency of *vacA* and *oipA* Genes in *Helicobacter pylori* Samples Isolated from Patients with Gastrointestinal Disorders.

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Background and Aim: *Helicobacter pylori* (*H. pylori*) is the main etiology of peptic ulcers and chronic gastritis. *H. pylori*, with its great diversity in pathogenic genes, is directly and indirectly involved in many disorders of the gastrointestinal tract and even outside the gastrointestinal tract. The aim of this study was to evaluate the frequency of this infection in patients referred to endoscopic centers and the presence of some important virulence genes such as *vacA* and *oipA*.

Methods: In this study, from 100 patients who referred to an endoscopy center, two pieces of tissue were prepared from the antrum or duodenum by a physician. One of the two pieces of tissue isolated from the patient's stomach was allocated for urease test and the other piece was used for DNA-extraction by phenol-chloroform method. First, the frequency of *H. pylori* was examined in all samples by PCR and glmM primer, and then for glmM positive samples, multiplex-PCR test was performed with primer of *vacA* and *oipA* genes. Finally, the results of PCR were analyzed by agarose gel electrophoresis.

Results: In the reviews of this study 67% of biopsies were urea-positive. Bacterial frequency with glmM gene was 53%. The frequency of *vacA* and *oipA* pathogenic genes among positive *H. pylori* samples was 73% and 48% by PCR, respectively.

Conclusion: The importance of pathogenic genes and its high frequency in patient samples indicate the need to use rapid and accurate identification methods such as PCR in clinical diagnoses. Also, due to the fact that in Multiplex-PCR test, several genes are examined simultaneously, this method is preferred to better identify the dominant strains in each region, by examining the number of more pathogenic genes to provide more effective treatments. Finally, considering the importance and consequences of *H. pylori* infection, the obtained epidemiological information and data analysis and gene analysis should be evaluated more comprehensively.

Keywords: pathogenic genes; glmM; endoscopy; *Helicobacter pylori*

Frequency of Bacteria Causing Bacteremia in Covid -19 Positive Children Using BACTEC System in Mofid Children Hospital, Tehran, Iran

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Background and Aim: Bacteremia is one of the main reasons of mortality in children through the world. Children with bacteremia could be very vulnerable to any infection. The rise of coronavirus disease 2019 (Covid-19) among children particularly who are hospitalized and got bacteremia could lead to death. In this study, we investigated the frequency of different bacterial strains isolated from blood samples of children with bacteremia and confirmed covid-19 test.

Methods: Blood samples of children with bacteremia and confirmed covid-19 collected in BACTEC (Becton Dickinson Diagnostic Instrument Systems, Sparks, Md) bottles. The bacterial isolates detection was done by this system, characterization of the strains was done by selective media including MacConkey agar and biochemical tests.

Results: A total 96 blood samples collected from 96 children with bacteremia and positive covid-19. Out of 96 pathogens isolated from blood samples, 88 (91.6%, 88/96) of them were bacteria. The frequency of isolated bacterial strains isolated from blood samples of children with bacteremia and Covid-19 were like: *Klebsiella* spp. (36.36%, 32/88), *Staphylococcus* spp. (19.3%, 17/88), *Pseudomonas* spp. (17%, 15/88), *Acinetobacter* spp. (6%, 6/88), *Enterococcus* spp. (5.6%, 5/88), *Escherichia* spp. (4.5%, 4/88), *Enterobacter* spp. (4.5%, 4/88), Diphtheroid (2.2%, 2/88), *Corynebacterium* spp. (1.1%, 1/88), *Micrococcus* spp. (1.1%, 1/88) and *Citrobacter* spp. (1.1%, 1/88). There was a significant co-relation between the bacterial strains causing bacteremia and covid-19 in hospitalized children.

Conclusion: *Klebsiella* spp. bacteremia had the most frequency among children with covid-19. As a matter of fact, there is limited data about bacterial and Covid-19 co-infections particularly in children. Based on the results of this study, Covid-19 could affect hospitalized children and lead to death.

Keywords: Covid-19; Bacteremia; *Klebsiella* spp.; Co-infection.

Frequency of *Helicobacter pylori* Virulence Genes in Water Resources of Western Iran by Direct PCR

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Background and Aim: *Helicobacter pylori* (*H. pylori*) causes the most common human bacterial chronic infection worldwide. The sero-epidemiological studies show that up to 80% of people in the developing countries have an *H. pylori* infection history. Previous studies indicate the vigorous relationship between *H. pylori* infection prevalence and water hygiene status. This study aimed to detect the *H. pylori* virulence genes by direct PCR on water samples of Sanandaj city, located in the west of Iran.

Methods: Two hundred and fifty water samples were collected from water resources, including wells, Municipal water, river, and dam in Sanandaj city during 6 months from April to September 2019. The preparing stages of water samples was including primary centrifuge, filtration and final centrifuge. To extract potential bacterial DNA, the sediment of water samples was treated by using PowerSoil DNA Isolation Kit (Qiagen Co.). *H. pylori* virulence genes were detected by using direct PCR and specific primers. The *H. pylori* 16s rDNA gene sequence of water samples were aligned and compared with other *H. pylori* 16s rDNA gene sequences registered in GenBank by using Mega7 software.

Results: The frequency of *H. pylori* genome in water samples was 23%, including 20%, 19%, 30%, and 36%, into tap, well, dam, and river water samples, respectively. The highest and lowest frequency rates belonged to well (62.5%, September) and dam water samples (0.0%, August), respectively.

Conclusion: The frequency of *H. pylori* virulence genes in Sanandaj city water samples was remarkable by using direct PCR method.

Keywords: *Helicobacter pylori*; Sanandaj; Virulence genes; Water resources

Frequency of *Legionella pneumophila* Virulence Genes in Hospital Water Supply Systems of Western Iran

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Background and Aim: *Legionella pneumophila* (*L. pneumophila*) is still a common cause of Hospital-acquired pneumonia. In this regard, hospital water supply systems are an important resource of bacterial transfer. The aim of this study was to detect *L. pneumophila* in water supply systems of Sanandaj's hospitals.

Methods: In a cross-sectional study, 100 water samples from hospital water supply systems including shower of bathrooms, laboratory water bath, water cooler tank, and ponds were collected during 6 months, from February to November in 2020. The *L. pneumophila* were examined through culture on BCYE and Hippurate hydrolysis test. Then PCR was used as molecular method by using *Legionella* species primers.

Results: Twelve strains of *Legionella* species were isolated from the water samples through culture on BCYE medium. Furthermore, the prevalence of 16s rRNA, *mip* and *rtxA* genes detected in water samples by PCR were 16%, 4% and 4%, respectively. 4 strains of *Legionella* isolated were determined *L. pneumophila* through hippurate hydrolysis test.

Conclusion: A considerable prevalence of *mip* and *rtxA* genes was detected in environmental strains of *L. pneumophila* isolated from Sanandaj's hospital water supply systems.

Keywords: *Legionella pneumophila*; 16s rRNA; *mip*; *rtxA*; PCR; Hospital Water resources

Graphene Quantum Dots-PVA Nanocomposite as a Platform for Wound Dressing

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Background and Aim: Herein, we report a new platform for wound dressing based on a new nanocomposite material fabricated by synergistic mixing graphene quantum dots (GQDs) and polyvinyl alcohol (PVA). The antibacterial activity of this nanocomposite was investigated against tow model bacteria, *Staphylococcus aureus* as a gram-positive bacterium and *Pseudomonas aeruginosa* as a gram-negative bacterium.

Methods: Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were evaluated for GQD/PVA nanocomposite and then In vivo studies were also assessed in burned wound infection animal model.

Results: The results indicated a persistent level against methicillin-resistant *Staphylococcus aureus* (MRSA) in higher ratios of GQD. The optimum ratio of GQD to PVA was determined based on in vitro studies and MTT assay. *In vivo* studies were also assessed in burned wound infection models and the CFUs in the burn wound were quantified by a standard colony counting method.

Conclusion: Finally, due to the results, the mechanism of antibacterial activity of nanocomposite described as a contact mediated oxidative stress induction.

Keywords: Biological Inorganic Chemistry; antimicrobial materials; active packaging materials; curcumine; *Pseudomonas aeruginosa*; *Staphylococcus aureus*

High Prevalence of Antiseptic Genes in Clinical Isolates of Methicillin-resistant *Staphylococcus aureus* (MRSA) in Isfahan, Iran

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Background and Aim: Biocides are Disinfectants and antiseptics that have been used in hospitals in order to limit the number of microorganisms. But misusing these substances extensively has led to a significant staphylococcal resistance to them. Besides gaining multi-drug-resistant genes, *Staphylococcus aureus* has gathered genes responsible for biocidal resistance, making it a dangerous threat to medical science. The aim of this study is to determine antibiotic resistance patterns and an investigation of antiseptic resistance genes in multidrug-resistant methicillin-resistant *Staphylococcus aureus* (MDR-MRSA).

Methods: for this research, 169 *Staphylococcus aureus* isolates were collected from four teaching hospitals in Isfahan. MRSA isolates were detected by using 30 µg cefoxitin disc and was confirmed by PCR detection of the *mecA* gene. Antibacterial susceptibility test was performed by disc diffusion method by some of common antibiotics on these isolates. Then the existence of antiseptic resistance genes (*qac A/B*, *smr* and *nor A*) was investigated in MDR-MRSA by PCR method.

Results: Out of 169 samples, 54 samples were identified as MRSA. antibacterial susceptibility testing for MRSA indicated the most resistance frequency toward erythromycin (83.3% (45/54)), ciprofloxacin (79.6% (43/54)), clindamycin (77.7% (42/54)), tetracycline (68.5% (37/54)), gentamycin (66.6% (36/54) and rifampin (57.4% (31/54)). all MRSA isolates were susceptible to linezolid and only one isolate was resistant to quinupristin/dalfopristin. Among MRSA isolates, 81% (44/54) isolates were multidrug-resistant (MDR). PCR method revealed that 100% of the samples had *smr* and *norA* genes. also 96.3% (52/54) of the isolated MRSAs had *qacA/B* gene.

Conclusion: The results indicate a growth in the number of multi-drug resistant *Staphylococcus aureus* that have antiseptic resistance genes. Further studies must be considered in order to contribute in better use of biocidal agents or even replace them with more useful substances.

Keywords: methicillin-resistant *Staphylococcus aureus* (MRSA); biocide; antibiotic resistance

High Titer of Antibody against *Streptococcus pneumoniae* LytA Protein in Healthy Individuals

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Background and Aim: LytA, is a surface protein that plays an important role in the pathogenesis of pneumococcal infections. The LytA protein is highly conserved among *Streptococcus pneumoniae* strains and is absent among other *Streptococcus* species. In the present study, the level of antibodies against LytA recombinant protein was evaluated in healthy individuals

Methods: DNA was extracted from *Streptococcus pneumoniae* ATCC 49619 to amplify lytA gene by PCR. The lytA amplicon and pET28a vector were separately double digested using Nde-1 and Xho1 restriction enzymes and then ligated together with ligase enzyme. The recombinant plasmid was transferred into the *Escherichia coli* BL21 strain, and the LytA recombinant protein was purified using Ni-NTA affinity chromatography. Western blot was carried to detect LytA recombinant protein. The titers of anti-LytA antibodies were determined in three age groups;

Results: The lytA gene was highly expressed in *Escherichia coli* BL21 host. The recombinant LytA protein was purified and confirmed by western blotting. Tukey test analysis showed that there were no significant differences among the age groups considering the anti-LytA titer of 10. However, at the anti-LytA titer of 160, significant differences were observed between group 1 vs. group 2 ($p < 0.05$); group1 vs. group 3 ($p = 0.003$), and 2 VS group3 ($p = 0.024$).

Conclusion: Considering the high titer of anti- LytA antibody in healthy individuals, the LytA protein seems to be a highly immunogenic antigen and a potential target for developing vaccines against pneumococcal infections

Keywords: *Streptococcus pneumoniae*; N-acetylmuramoyl-l-alanine amidase; antibody

How Pathogenic Bacteria Colonize on Host Surfaces?

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Background and Aim: The first step for creating disease by harmful bacteria after their entering into the human 's body, is adherence to target tissue. In this review we will describe the ability of pathogenic bacteria for colonizing their main target organs.

Methods: This study is a review article and we collected all data information by searching the specified set of Medical Subject Heading (MeSH) terms such as: Colonization, Pathogens, & Mechanisms of Adherence, in NCBI and PubMed using search engines like as Science Direct, MEDLINE and ISI Web of Science.

Results: All of the different entrance ways of the human body that pathogenic bacteria usually entered from them, covered by mucin. In addition to glycoprotein materials which exist in this layer, many kinds of different substances are there, like as immunoglobins, defensins, cryptdins, regenerating islet-derived protein 3 γ (REG3 γ), etc. each bacterium has a specific mechanism for adherence to its target cell and evade from immune molecules such as IgA by producing IgA- protease and so pathogen- associated molecular proteins (PAMPs) that activates Toll- like receptors (TLR) signaling pathways. Pili, the short hair like appendages are one of the more important structures for binding bacteria to host cells that classified into six groups. For example, UPEC are uropathogenic Escherichia coli with an adhesion factor named PapG, which can recognize and bind to epithelial layer of kidney, such as glycosphingolipids and some of them possess type I pili which binds to D-mannosylated receptors. Neisseria meningitides has type IV pili that has a major role in formation of micro colonies attached to vascular endothelial cells. Some bacteria can change their pili and some of them have antigenic variation. Slime layer and capsule are two major biofilm molecules which can mimics host molecules and evade from immune system.

Conclusion: Bacteria have several factors which help them to adhere their target cell and initiate infection. Studying & collecting information about this subject can help us to finding ways for controlling infectious disease and finding suitable antibacterial substances can limit infectious.

Keywords: Pathogenic bacteria; colonization; adherence factors; receptor

***In vitro* Antibacterial Effect of Ethanol and Hydrogen peroxide Consortium with Preservatives against Clinical Isolates**

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Background and Aim: Health-care professionals are a major source of infectious diseases caused by microorganisms. The application of various antiseptics and disinfectants is one of the most effective preliminary controls of infectious microbial sources. The prime objective of present research work, to make new antimicrobial formulation and to determine the efficacy of antibacterial agents against clinical isolates from burn patients admitted in Burn & Accident hospital, southwest of Iran.

Methods: Different source samples such as blood, wound & biopsy collected from burn patients and they were enriched and cultured repeatedly to get axenic culture, *Bacillus* spp. isolated from environment. Species identification was carried out using phenotypic & molecular methods (16S rDNA sequencing). In vitro antibacterial activity of Ethanol & Hydrogen peroxide (newly formulated) determined by broth microdilution method.

Results: *Pseudomonas aeruginosa*, *Acinetobacter* spp., *Staphylococcus aureus* & *Enterobacter* spp. has been isolated from clinical isolates. MIC of newly formulated Ethanol & Hydrogen peroxide consortium against clinical isolates used in study showed highest potent antibacterial activity and as per the in vitro data revealed that, this formulation would be the best disinfectant to eliminate all the microorganisms including spore. In this research, the use of appropriate concentration ethanol 70%, hydrogen peroxide 0.126%, silicon ester & some preservatives have been considered to improve disinfection & protect the instruments & skin from side effects.

Conclusion: The results revealed that ethanol & hydrogen peroxide consortium has promising antibacterial against tested microorganism, & can be recommended for disinfection process.

Keywords: Molecular identification; Ethanol & Hydrogen peroxide; Disinfectants; Silicon ester

Inhibitory Effects of Potentially Probiotic *Lactobacillus* species against Pathogenic Bacteria Survival in Burn Wounds and Diabetic Foot Ulcer

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Background and Aim: Infection remains the most common complication after burn injury and can result in sepsis and death, despite the use of topical and systemic antibiotics. *Lactobacillus* species have been considered as probiotic microorganisms due to their potential human health properties and was assessed as a bio-control strategy for wound infections. Evaluation of capabilities of native probiotic *Lactobacillus* species for antibacterial peptide production, inhibition of biofilm formation and competition for cell attachment against pathogen bacteria were the aims of this study.

Methods: Anti-bacterial activities of the CFS of *Lactobacillus* strains were assessed by agar-spot test and the microplate method via violet staining was used for biofilm formation. Human skin fibroblast (HFSF-PI3) was used for examination of the *Lactobacillus* adherence and inhibitory activity against attachment of pathogen bacteria.

Results: The agar-spot test results showed that *Lactobacillus* species have a high ability to inhibit *Pseudomonas aeruginosa* compared to other genus and microplate method results also approved it. *Lactobacillus* species have a good attachment to HFSF-PI3 cell line and competition results showed that *Lactobacillus* species had an inhibitory effect against the binding of *Pseudomonas aeruginosa* to the cell line.

Conclusion: Our results suggest that *Lactobacillus* strains may represent an alternative bio-control strategy against wound infections. The antimicrobial effect, inhibit the biofilm formation of pathogenic bacteria, and competition with pathogens for attachment are the three main probiotic effects of *Lactobacillus* species.

Keywords: Probiotics; *Lactobacillus*; Burn Wounds; Diabetic Foot; Pathogenic Bacteria

Investigating the Frequency Positive cultures of the Pulmonary Secretions, the Organisms and Determining Their Antibiotic Susceptibility Patterns in Intubated and Suspected of Pneumonia Patients in ICUs of Shahid Beheshti hospital of Hamadan in 2019-2020

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Background and Aim: One of the common problems of intubation patients admitted to the intensive care unit is hospital-acquired pneumonia, which in addition to increasing the length of stay of patients and the costs of diagnosis and treatment may lead to their death. The aim of this study was to determine the frequency of positive culture of pulmonary secretions, causative organisms and their pattern of antibiotic susceptibility in patients with intubation and suspected pneumonia in the ICU.

Methods: In a descriptive cross-sectional study, 143 intubated patients admitted to the ICU of Shahid Beheshti Hospital in Hamadan with hospital acquired pneumonia during 2019 and 2020 were selected and in terms of the frequency of nosocomial infections, the causative organisms and antibiotic susceptibility were evaluated. After registering in the checklist, patients' data were analyzed using SPSS software version 16.

Results: The mean age of patients with pneumonia was 72.15 years. Out of 144 samples, 54.86% had ventilator-induced pneumonia, 8.33% had tracheitis, 31.25% had colonization and 56.5% had negative culture. Of the 91 patients with pneumonia and tracheitis, 42 were *Acinetobacter*, 15 were *E. coli*, 14 were *Klebsiella*, 10 were *Enterobacter*, 4 were *Pseudomonas* and 6 were Gram-positive. The highest rate of antibiotic susceptibility to colistin was 95.24% in *Acinetobacter*, 66.66% in *Escherichia coli*, 64.28% in *Klebsiella*, 100% in *Enterobacter* and 25% in *Pseudomonas*. Sensitivity to vancomycin with 66.66% was highest in gram-positive bacteria. Experimental antibiotic regimen in most patients was meropenem with vancomycin, meropenem alone or in combination with other antibiotics.

Conclusion: *Acinetobacter* is the most common cause of ventilator-induced pneumonia in intubated patients admitted to the intensive care unit. Ventilator-associated pneumonia-causing microorganisms are highly resistant to most antibiotics used, especially the experimental antibiotic regimen.

Keywords: Nosocomial infection; Antibiotic susceptibility; pulmonary secretion; Ventilator-associated pneumonia

Investigating the Relationship between COVID-19 Disease and Tuberculosis: A Systematic Review

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Background and Aim: COVID-19 disease, at an alarming rate in the world, puts people with underlying diseases at a higher risk of death and, like tuberculosis, is an infectious disease that targets the respiratory system. Little is known about the relationship between COVID-19 and tuberculosis, so it is important to examine the interaction between the two diseases.

Methods: In this study, research related to the prevalence of COVID-19 disease in patients with tuberculosis was reviewed. These articles were excluded from the study by searching for keywords in the PubMed, Scopus, and Embase databases based on inclusion, evaluation, and duplicate criteria.

Results: The results show that while tuberculosis is still the world's leading infectious killer, the COVID-19 epidemic has overtaken all health issues around the world. Also, studies conducted in more than 172 centers in 34 countries showed that 16.5% of patients with more severe clinical symptoms were affected by both diseases at the same time. The inter relationships between the two diseases are significant in three respects: First, since viral respiratory infections and tuberculosis (TB) inhibit host immune responses, these two enhance their detrimental effects and exacerbate their fatal progression, clinical consequences, and death of patients. More severe strains of COVID-19 are devastating in patients with active TB or previous TB whose lung function is impaired. Second, the Covid-19 epidemic and the global response to it will have a profound and long-term impact on TB diagnosis and control, as TB control programs are under pressure due to diversion of resources, and loss of health system focus, leading to reduced quality of care. Tuberculosis and the consequences get worse. Third: The symptoms of tuberculosis and COVID-19 are similar, this can not only confuse the diagnosis, but also cause deviation in the timely diagnosis of tuberculosis due to the fear of COVID-19.

Conclusion: Despite the fact that a significant portion of the world's population is infected with COVID-19, tuberculosis remains a major threat to global health, so tuberculosis and Covid-19 need immediate attention. Tuberculosis should be considered as a risk factor for severe COVID-19 disease and patients with tuberculosis should be given priority for vaccination against COVID-19 prevention measures. New solutions should be devised immediately or the TB control goals will never be achieved.

Keywords: COVID-19; Tuberculosis; Interaction

Investigation of Antibacterial Effect of Nanofiber mats-green Nanogel against Clinical and Standard Strains of Pathogens

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Background and Aim: *Staphylococcus aureus* and *Pseudomonas aeruginosa* are two causative agents of human skin infectious diseases. Because of antibiotic resistance phenomenon in many bacteria, developing new antibacterial agents such as green antibacterial agents have become a new milestone.

Methods: In this study, nanofibers of alginate- polyethylene oxide nanofibers with 177 ($\pm$ 36) nm diameter were prepared. ingredients of *Elettaria Cardamomum* essential oil were determined: α -terpinol acetate, 1-8 cineole, Linalool Acetate, Linalool, α - terpineol, Limonene were identified as six major ingredients. A nanoemulsion-based nanogel of *Elettaria Cardamomum* was prepared and physically impregnated on the surface of the nanofiber's mats. Antibacterial effects of this component at 2.5, 5, and 10 mg/mL concentrations with 24 h exposure were investigated against standard and clinical strains of *Pseudomonas aeruginosa* and *Staphylococcus aureus*..

Results: After 24 h exposure at 2.5, 5, 10 mg/mL concentration, the growth rate of a standard strain of *S. aureus* ~95, ~ 70, ~ 50 % was decreased, respectively. There is no significant reduction in other strains.

Conclusion: According to findings, this nanofiber could be propose as a green potent antibacterial agent in different propose such as wound dressing.

Keywords: *Elettaria Cardamomum*; essential oil antibacterial

MALDI-TOF Mass Spectroscopy Applications in Clinical Microbiology

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Background and Aim: There is a range of proteomics methods to spot and analyze bacterial protein contents such as liquid chromatography-mass spectrometry (LC-MS), two-dimensional gel electrophoresis, and matrix-assisted laser desorption/ionization mass spectrometry (MALDI-TOF MS), which give comprehensive information about the microorganisms that may be helpful within the diagnosis and coverings of infections. Microorganism identification by mass spectrometry is predicted on identifying a characteristic spectrum of every species so matched with an outsized database within the instrument. MALDI-TOF MS is one of the diagnostic methods, which is a straightforward, quick, and precise technique, and is employed in microbial diagnostic laboratories these days and may replace other diagnostic methods. One of the MALDI-TOF MS's crucial applications is bacteriology, which helps identify bacterial species, identify toxins, and study bacterial antibiotic resistance. By knowing these cases, we will act more effectively against bacterial infections.

Results: In the study of Sun et al., they used MALDI-TOF MS to identify species of *Lactobacillus plantarum*, a Gram-positive bacterium, and they identified 34 proteins as the biomarker proteins and used these proteins for identifying species of bacterium in the various cultures. In another study, MALDI-TOF MS was used to identify 99 species of clinical *Streptococci*, a Gram-positive bacterium; they identified 71 *Streptococcus mitis*, 23 *Streptococcus anginosus*, and five *Streptococcus salivarius*. Another study carried out on mycobacteria by Hettick et al. used MALDI-TOF MS to identify various *Mycobacteria* and acid-fast bacteria, such as *Mycobacterium tuberculosis*, *Mycobacterium intracellulare*, *Mycobacterium avium*, *Mycobacterium bovis*, *Mycobacterium kansasii*, and *Mycobacterium fortuitum*.

Conclusion: Rapid diagnosis of infection makes the control of it more straightforward, vital for protecting the community's health. There are various methods for rapid diagnosis of infections. One of them is MALDI-TOF MS, which is a fast, easy, and accurate method. Detecting bacterial proteinaceous agents helps us diagnose disorders quickly, which leads to more rapid treatment of the patient.

Keywords: MALDI-TOF MS; Bacteria; Antibiotic Resistance

Molecular Characteristics of Multiple and Extensive Drug-resistant *Acinetobacter baumannii* Isolates from a Tertiary Care Hospital in Southern Iran.

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Background and Aim: The emergence of multiple drug-resistant (MDR) *Acinetobacter baumannii* (*A. baumannii*) strains have become a global problem. This study aimed to evaluate the occurrence of antibiotic resistance and to investigate the presence of antibiotic resistance determinants among *A. baumannii* isolates obtained from hospitalized patients in Iran.

Methods: This cross-sectional study was performed on 92 *A. baumannii* isolates in the years 2015-2016. Antimicrobial susceptibility testing was carried out by the disk diffusion method. The presence of antibiotic resistance determinants was detected by the PCR method. All the *A. baumannii* isolates were resistant to tested carbapenems, fluoroquinolones and sulfonamide agents, and susceptible to polymyxins.

Results: Of the isolates 92.4% were extensive drug-resistant (XDR) and 7.6% were MDR. PCR screening for the presence of integron genes revealed that class 1 integron presented in 46.7% of isolates and class 2 in 18.5% isolates. Of the investigated antibiotic resistance genes, bla VIM, bla IMP, bla SPM, sul1 and sul2 were positive in 75%, 12%, 12%, 47.8%, and 67.4% of isolates, respectively.

Conclusion: The high prevalence of metallo-beta-lactamase (MBL) and sul genes in our results may indicate the importance of these genes in the dissemination of carbapenem- and sulfonamide-resistant *A. baumannii* isolates. Rational and restricted prescription of carbapenems may be an effective way to minimize the emergence and spread of MBL-harboring strains.

Keywords: *Acinetobacter baumannii*; extensive drug-resistant (XDR); integrons; metallo-beta-lactamase (MBL)

Multiple Sclerosis, Urinary Tract Infections and Infertility

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Background and Aim: Multiple sclerosis (MS) is a chronic autoimmune disease of the central nervous system (CNS) with destroying the axons and the myelin that shows a variable and progressive course. MS patients usually have complications, such as Lower urinary tract dysfunction, Urinary Tract infection (UTI), and sexual dysfunction, which are the most common complications in the patients. MS is also common in the young adults and can lead to significant sexual dysfunction and infertility that increases with the disease progression.

Methods: This article provides an up-to-date overview of clinical manifestations, research, and treatment choices for UTI and infertility in patients with MS. In the last two decades, many advances in the treatment of MS have been made with the advent of immunomodulatory therapies that can potentially slow down the progression of the disease and change its course.

Results: UTI and sexual dysfunction are major risks and concerns in MS patients. The high prevalence, hospitalization rate, and urinary tract infection mortality in MS are worrying, as is the cause-and-effect relationship between UTI and corticosteroid use in outbreaks. Therefore, knowledge of important complications of MS, such as UTI and infertility in this group is important to make early diagnoses, adequate management and prevention.

Conclusion: Therefore, knowledge of important complications of MS, such as UTI and infertility in this group is important to make early diagnoses, adequate management and prevention.

Keywords: Multiple sclerosis; infertility; UTI; sexual dysfunction

Presence of *bla*_{OXA-24/40}-like, *bla*_{NDM} and Expression of Efflux Pumps in Carbapenem Resistant *Acinetobacter baumannii* Isolated from COVID-19 Patients

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Background and Aim: Era of COVID-19 has witnessed wide range of bacterial health-care associated infections in these patients. Increase in the incidence of oxacillinase and New Delhi metallo- β -lactamase-producing carbapenem-resistant *Acinetobacter baumannii* (CRAB) has led to poorer outcome. *A. baumannii* is widely notorious for being resistant to several antimicrobial classes, including cephalosporins, fluoroquinolones and carbapenems, rendering infections difficult to eradicate. Carbapenems no longer find their usage due to increasing prevalence of CRAB. As hospital staff had witnessed immethodical usage of antibiotics mainly, macrolides and carbapenems thus, this study investigated the prevalence of *A. baumannii* in these patients and assessed presence of *bla*_{OXA-24/40}-like, *bla*_{NDM} and expression of efflux pumps in CRAB isolated from COVID-19 patients.

Methods: *A. baumannii* (n=28) isolates were recovered from inpatients afflicted by COVID-19. Identification of *A. baumannii* was confirmed by the amplification of RNA polymerase β subunit (rpoB) gene using PCR. Genetically confirmed *A. baumannii* (n=28) isolates were screened for carbapenem resistance by the Kirby-Bauer and E-test and the presence of *bla*_{OXA-24/40}-like and *bla*_{NDM} genes and the level of expression of efflux pumps (adeB and adeJ) was investigated by Real-time PCR.

Results: Twenty-eight *A. baumannii* isolates were isolated from endotracheal aspirates (n=12), blood stream (n=8), wound (n=6) and urine (n=2) of COVID-19 admitted patients. Of these 14.28% isolates were extensive drug resistant while others multidrug-resistant. Frequency of *bla*_{OXA-24/40}-like and *bla*_{NDM} genes was 21.42% and 7.14% respectively. Over-expression of adeB and adeJ was observed in 17.85% and 14.28% *A. baumannii* strains respectively.

Conclusion: Presence of *bla*_{OXA-24/40}-like and changes in the expression of efflux pumps mainly contributed to the emergence of CRAB in our hospital *A. baumannii* isolates. Intubated COVID-19 patients' positive for *A. baumannii* highlights lacunae in infection control practices demanding strict surveillance strategies and control programs.

Keywords: *Acinetobacter baumannii*; Carbapenemase; Oxacillinase; Efflux pump; COVID-19

Prevalence and Serotype Distribution of *Streptococcus agalactiae* Isolated from Pregnant Women in Khoy, Iran

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Background and Aim: *Streptococcus agalactiae* (*S. agalactiae*) colonizes pregnant women and if transmitted to the newborns during delivery, can lead to serious and fatal diseases such as sepsis, pneumonia and meningitis. The aim of this study was to evaluate the frequency, serotypes and antibiotic resistance patterns of *S. agalactiae* isolated from pregnant women's vagina.

Methods: In this cross-sectional study, 446 vaginal swab specimens were collected from pregnant women. Suspected colonies were confirmed by conventional tests such as hemolytic activity, catalase, Gram staining, Bile Esculin Agar, CAMP, and hippurate hydrolysis. Antimicrobial susceptibility of *S. agalactiae* strains was determined by Kirby-Bauer disk diffusion method. The presence of resistance genes (*ermB*, *ermTR*, *mefA* and *linB*) detected with the Multiplex PCR method. The isolates were serotyped by the sequential multiplex-PCR method

Results: A total of 42 strains were isolated from 446 vaginal swab samples. All strains were sensitive to penicillin, ampicillin and vancomycin, and 45.2 and 28.5% of the isolates were not susceptible to erythromycin and clindamycin, respectively. The *ermTR*, *ermB*, *mefA* and *linB* genes were detected in 11(26.19%), 9(21.42%), 2 (4.76) and 4 (9.52%) isolates, respectively. The most common serotype in this study was III (61.9%) followed by II (14.28%), Ia (11.9%), V (7.14%) and Ib (4.76%).

Conclusion: Serotype III is highly prevalent in Iran and due to its association with invasive diseases, screening of pregnant women and prophylactic treatment of carriers is necessary. The most important resistance mechanism for macrolides is the *ermTR* gene and the M phenotype.

Keywords: *Streptococcus agalactiae*; Molecular Serotyping; Antibiotic Resistance; group B *Streptococcus* (GBS)

Prevalence of Bacterial Co-Infections and Antibiotic Resistance in Patients with Covid-19; a Systematic Review

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Background and Aim: Twenty-five percent of hospitalized pneumonia patients suffered from concomitant bacterial viral infection, and on the other hand, there is little data on the association of coronavirus respiratory disease with bacterial respiratory infection. Due to the widespread prohibition of the use of antibiotics and the recommendation not to use antibiotics in mild COVID 19 patients, we aimed to investigate the prevalence of bacterial infection and bacterial resistance in Covid 19 patients in this review article.

Methods: We searched MEDLINE, EMBASE, PubMed, Cochrane Central Register of Controlled Trials, and Web of Science from inception pandemic to 27 November 2021. The search strategy was as follows; (coinfection covid-19) AND (bacterial infection) AND (drug resistance). We excluded duplicate articles, review articles, case report articles, and articles that did not fit the purpose.

Results: Results: Among the important bacteria of nosocomial infections were *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Enterobacter cloacae*, *Proteus mirabilis*, *Enterococcus faecalis*, *Escherichia coli*, *Staphylococcus aureus*, *Acinetobacter baumannii*. The highest prevalence was *E. coli* (2.54%), *S. aureus* (1.49%), and *A. baumannii* (1.40%). Resistance of these isolates to one or more antibiotics was at least 82.1%, 62.5%, and 71.8%, respectively.

Conclusion: Among the important bacteria of nosocomial infections were *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Enterobacter cloacae*, *Proteus mirabilis*, *Enterococcus faecalis*, *E. coli*, *S. aureus*, *A. baumannii*. The highest prevalence was *E. coli* (2.54%), *S. aureus* (1.49%), and *A. baumannii* (1.40%). Resistance of these isolates to one or more antibiotics was at least 82.1%, 62.5%, and 71.8%, respectively.

Keywords: COVID-19; Coinfection; Bacterial Drug Resistance

Prevalence of *bla*_{CTX-M} Gene in *Acinetobacter Baumannii* Isolates from the Sputum Samples of Intensive Care Units (ICU)

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Background and Aim: *Acinetobacter baumannii* (*A. Baumannii*) has become an extremely troublesome nosocomial infection in ICU wards. Extended-spectrum beta-lactamases (ESBLs) are enzymes that can hydrolyze beta-lactam antimicrobials and neutralize their properties. This study aims to investigate the prevalence of the *bla*_{CTX-M} gene in *A. Baumannii* isolates from the sputum samples of intensive care units (ICU).

Methods: One hundred *A. baumannii* isolates were collected from intensive care units of Rasool Akram hospital Tehran, Iran. Microbiological and biochemical tests were confirmed to identify the isolates, then the antibiogram test was performed for all of the isolates and ESBL isolates were collected. Subsequently, detection of the *bla*_{CTX-M} gene was accomplished by PCR amplification.

Results: Among these isolates, 2 isolates were ESBL positive by the phenotypic method. Molecular characterization by PCR amplification showed 22/91% (22/96) isolates carried *bla*_{CTX-M} genes.

Conclusion: *A. Baumannii* isolates were resistant to extended-spectrum antibiotics, especially beta-lactam antibiotics. Therefore, the study of resistant genes is essential for controlling and monitoring infection in the different wards of the hospital, especially in ICUs.

Keywords: *Acinetobacter baumannii*; Nosocomial Infection; *bla*_{CTX-M}; Antimicrobial Resistance; ICU Wards

Prevalence of *bla*_{CTX-M} Gene in *Klebsiella Pneumoniae* Isolates from the Sputum Samples of Intensive Care Units (ICU)

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Background and Aim: Nosocomial infections are a growing concern in modern-day medicine. This is in part due to the increased prevalence of drug-resistant pathogens. *Klebsiella Pneumoniae* (*K. Pneumoniae*) is one of the most prominent pathogens found in drug-resistant infections all around the world. Drug resistance of *K. Pneumoniae* is associated with the *bla*_{CTX-M} gene that is responsible for the production of β -lactamase. This study aims to determine the prevalence of the *bla*_{CTX-M} gene in *K. pneumoniae* strains isolated from ICU patients.

Methods: One hundred five *K. pneumonia* isolates were collected from intensive care units of Rasool Akram hospital Tehran, Iran. Microbiological and biochemical tests were confirmed to identify the isolates. Then the antibiogram test was performed for all of the isolates and ESBL isolates were collected. Subsequently, detection of the *bla*_{CTX-M} gene was accomplished by PCR amplification.

Results: Ten samples were considered positive by phenotypic ESBL test, and 89.4% (85/95) isolates carried the *bla*_{CTX-M} gene.

Conclusion: *bla*_{CTX-M} gene was frequently prevalent among clinical isolates of *K. pneumoniae* which were collected from ICU wards. Inhibition of the expression of antibiotic resistance genes is essential in the management and control of nosocomial infection. Similar studies can provide invaluable information to better strategize against this problem.

Keywords: Nosocomial Infection; Antimicrobial Resistance; ESBL; *Klebsiella pneumoniae*

Prevalence of Brucellosis in Blood Donors Referred to Malalai Hospital in Kabul, Afghanistan

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Background and Aim: Brucellosis is a zoonotic infection transmitted from animals to humans. Which is considered as a health problem in different cities of Afghanistan. The aim of this study was to evaluate the serum prevalence of brucellosis in blood donors in Malalai Hospital in Kabul

Methods: In a descriptive cross-sectional study, anti-Brucella antibodies were tested in the blood serum of donors referred to Malalai Hospital in Kabul. All blood samples were qualitatively tested for Rose Bengal. Wright and 2ME tests were then performed on positive samples of Rose Bengal. A titer of greater than or equal 1/80 for STAT and a titer of greater than or equal 1/40 for 2ME were taken as criteria of brucellosis. Findings were analyzed using SPSS software and Friedman statistical test.

Results: Of the 2,000 blood donors, 85 (4.25%) had a Rose Bengal test, 49 (2.45%) had a Wright of 1.8 and above, and 26 (1.3%) had a Meme 140 and above. Serum prevalence of brucellosis was calculated to be 1.3%. There was no significant relationship between serum prevalence of brucellosis, age and sex of blood donors. The highest prevalence was in people who came to Kabul from Bamyan and Ghazni and the lowest prevalence was observed in donors living in Kabul. There was a statistically significant relationship between the season and place of residence of blood donors and the prevalence of brucellosis, ($P = 0.026$) and ($p = 0.041$, respectively).

Conclusion: According to the results of this study, the serum prevalence of brucellosis in blood donors who came to Malalai Hospital is higher than expected. However, Brucella is very common in most cities, including Bamyan.

Keywords: Blood Donors; Brucellosis; Seroprevalence; test

Prevalence of Colonization of *Streptococcus agalactiae* Isolated from Urinary Tract Infections in Pregnant Women Referred to Qom Hospital, Northern Portion of the Iran

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Background and Aim: Urinary tract infection is the most common bacterial infection in children and adults, especially females. Among urinary tract infection caused by bacteria, *Streptococcus agalactiae* (*S. agalactiae*) can cause periodic, temporary, or persistent infection in mothers. The aim of this study is the prevalence of colonization of *S. agalactiae* isolated from urine sample in pregnant women referred to Qom hospitals, Northern portion of the Iran.

Methods: In this descriptive study, we worked on 1264 pregnant women who have been clients of Ali-ibn-AbiTaleb hospital medical laboratory. Midstream urine specimens collected from pregnant individuals were passaged on blood agar media and after differential tests and detection of *Streptococcus agalactiae*, an antibiotic susceptibility test has been conducted by using the disc diffusion method according to CLSI protocols.

Results: Out of 1264 pregnant patients, 17.64% were diagnosed with beta-hemolytic *Streptococcus* bacteria, and 13.37% were diagnosed with *S. agalactiae*. The rest of them were colonized with other beta-hemolytic *Streptococcus* family bacteria. The highest resistance was related to Clindamycin and Erythromycin, and the lowest resistance was related to Nitrofurantoin. resistance to Penicillin is about 33%, but mismanagement in the prescription of this antibiotic will result in more resistance.

Conclusion: Regarding this research results, urine culture test better gets done before week 34 and the last weeks of pregnancy. To prevent further *S. agalactiae* antibiotic resistance, we recommend other antibiotics to be used before week 34 of pregnancy rather than Penicillin, and Penicillin treatment need to be postponed to the last weeks of pregnancy.

Keywords: *Streptococcus agalactiae*; urinary tract infection; antibiotic resistance; pregnancy

Prevalence of Cyclomodulin-encoding Genes among the Mucosa-associated *Escherichia coli* Strains from Colorectal Cancer Patients and Control Subjects

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Background and Aim: Colorectal cancer (CRC) is one of the most common cancers in the world. Recent studies have shown that cyclomodulin-positive *Escherichia coli* (*E. coli*) strains are involved in the pathogenesis of CRC. The aim of this study was to compare the prevalence of cyclomodulin-encoding genes among mucosa-associated *E. coli* strains from patients with CRC and healthy controls.

Methods: Between August 2019 and June 2020, 96 subjects, 48 with CRC and 48 without were examined at two referral university-affiliated hospitals in northwest Iran. The colonic tissue samples obtained during colonoscopy was used to identify mucosa-associated *E. coli* isolates. *E. coli* isolates were typed using enterobacterial repetitive intergenic consensus-based PCR (ERIC-PCR). The PCR method was used to evaluate the presence of cyclomodulin- (*cdtB-I*, *cdtB-II*, *cdtB-III*, *cdtB-IV*, *cdtB-V*, *cnf1*, *cnf2*, *cnf3*, *cif* and *pks* genomic island) encoding genes.

Results: In this study some tissue samples did not possess any strain of mucosa-associated *E. coli* and some samples had one or more than one strains, thus a total of 60 mucosa-associated *E. coli* strains from CRC patients and 41 strains from the control group were identified. The prevalence of cyclomodulin-encoding genes in strains isolated from CRC patients was significantly higher than the control subjects ($p < 0.05$). The *cif* gene was predominant in CRC patients (16.6%), followed by *cnf1* (13.3%), and *pks* (10%). Moreover, *cif* (2.4%) was the only cyclomodulin-encoding gene that was detected in the *E. coli* strains from control group.

Conclusion: In conclusion, the gut mucosa of CRC patients was more colonized by cyclomodulin-positive *E. coli* strains in comparison to the control subjects.

Keywords: Colorectal cancer; *Escherichia coli*; Cyclomodulin

Prevalence of Enteropathogenic *Escherichia coli* in Mucosal Samples of patients with Colorectal Cancer and Control Subjects

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Background and Aim: Enteropathogenic *Escherichia coli* (EPEC) is an attaching and effacing (A/E) pathogen that causes diarrhea in acute infection. But recent studies have shown that some strains of EPEC are able to survive and replicate in colonic epithelial cells as a chronic intracellular pathogen and subvert some host cell functions ultimately leading to colorectal cancer (CRC). The aim of this study was to compare the prevalence of EPEC among mucosa-associated *E. coli* strains from CRC patients and healthy controls.

Methods: From August 2019 to June 2020, 45 CRC patients and 45 non-cancerous subjects from two referral university-affiliated hospitals in northwest Iran were enrolled in this study. The biopsy specimens were obtained from the colon and rectum was used to identify mucosa-associated *E. coli* isolates. The boiling method was used to extract DNA from *E. coli* strains. The PCR method was used to detect for the presence of the *eae*, *stx1*, *stx2* and *bfp* genes.

Results: Typical EPEC is defined as strains possessing *eae* and *bfp* genes but no *stx1*, *stx2* genes. In addition to *stx1*, *stx2* genes, atypical strains of EPEC also lack the *bfp* gene. In this study the *eae* gene was detected in 24.4% of *E. coli* strains from CRC patients, but only in 6.6% of strains from control group ($p < 0.05$). Moreover, all strains possessed *eae* gene were negative for *bfp*, *stx1*, *stx2* and genes. Therefore, all EPEC strains were atypical EPEC.

Conclusion: The gut mucosa of CRC patients provides suitable conditions for colonization of EPEC, and these strains may play a role in the pathogenesis of CRC.

Keywords: Colorectal cancer; Enteropathogenic; *Escherichia coli*; PCR

Prevalence of *Helicobacter Pylori* Antibodies Titer with ELISA in Blood and Compared to Stool Antigen in Urmia, Iran, 2021

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Background and Aim: *Helicobacter pylori* (*H. pylori*) is one of the most common human infections in the world. In developing countries, most children (80%) are infected by the age of 10, while in advanced countries, the prevalence of *H. pylori* and gastritis increases with age. This study aimed to evaluate the antibody titer of IgG and IgM anti-*H. pylori* using the ELISA method and to investigate its relationship with stool antigen.

Methods: This study was conducted in 2020 on 100 patients. After blood sampling and serum isolation, IgG and IgM antibody titers were measured by the ELISA method. On the other hand, stool antigens were also tested using the *Helicobacter pylori* Rapid Kit.

Results: showed that 51% of IgG positive and 54% of *H. pylori* Ag stool positive were studied. Other results showed that 33% of patients were both IgG positive and *H. pylori* Ag stool positive. No association was found between *H. pylori* Ag stool and IgG with IgM, which means that the IgM antibody was negative in all patients. That is, *H. pylori* Ag stool is the best and most accurate method for *H. pylori* screening.

Conclusion: Simple methods based on the search for antibodies and antigens with high sensitivity and specificity are available. The advantage of ELISA in addition to its high sensitivity and specificity also determines the antibody class. IgG has a higher diagnostic value and prognosis and represents a chronic stage of the disease. IgM is not very useful in the diagnosis of *H. pylori* and can be said to be an insensitive indicator of acute infection. On the other hand, for detection of *H. pylori* antigen in stool rapids can also be very useful due to its high accuracy and low test time.

Keywords: *Helicobacter Pylori*; antibodies titer; ELISA; stool

Prevalence of Infection with *Klebsiella pneumoniae* Isolates and the Antibiotic Susceptibility to Routine Antibiotics in Hospitalized Patients to the Rasht Velayat Burn Hospital in 2018-2020

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Background and Aim: The use of antibiotics in different hospitals and wards of a hospital varies due to differences in patients, hospital characteristics, and the way a physician or medical staff is trained. Increased resistance may lead to increased use of broad-spectrum antibiotics. Using information about antibiotics and sharing this information with other intensive care units in a hospital or other hospital will ultimately lead to a better understanding of the causes of antibiotic resistance and the rational use of it.

Methods: In a descriptive cross-sectional study, information on all cases of infection with *Klebsiella pneumoniae* (*K. pneumoniae*) isolates caused by burn wounds of patients referred to Rasht hospital in 2018-2020, including age, sex, percentage of burns, ward, duration of hospitalization, VAP, Consequences and antibiotic susceptibility were extracted from their accepted files and the results of antibiotic susceptibility with *K. pneumoniae* isolates causing infection due to burn wound were extracted from laboratory results.

Results: Out of 594 cases of burn infections, 61 cases (10.27%) were identified as *K. pneumoniae* which 44 cases (72.1%) were male and 17 cases (27.9%) were female. The mean age of patients was 39.15 ± 18.48 years. 42 (68.9%) cases of *K. pneumoniae* isolated from ICU and 19 cases (31.1%) were related to surgery. The mean time of hospitalization of patients was 4.18 ± 4.43 days. 5 cases of *K. pneumoniae* isolates (8.2%) are related to patients with ventilator-associated pneumonia (VAP). 35 cases of infection with *K. pneumoniae* isolates (57.4%) were related to recovered patients and 26 cases (42.6%) were related to deceased patients. Antibiotic susceptibility results showed that azithromycin (Resistance rate: 31.8%) and meropenem (36.4%) were the most effective antibiotics against *K. pneumoniae* and high level of resistance related to imipenem (85.7%).

Conclusion: The results current study identified a pattern of antibiotic susceptibility for *K. pneumoniae* isolates in patients admitted to the burn hospital in Rasht province. Our study showed a remarkable rate of burn wound infection, which demanded a more restricted and effective infection control policy. Moreover, our results provided important information for suggestions of proper antibiotics administration based on the local antibiotic susceptibility patterns.

Keywords: Burn wound infection; Antibiotic susceptibility; *Klebsiella pneumoniae*

Prevalence of Nosocomial Infections in Intensive Care Units in Imam Khomeini Hospital of Ilam

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Background and Aim: Nosocomial infections are a major challenge worldwide. An important part of these infections occurs in the ICU, which affects the treatment process, inpatient duration and mortality of patients admitted to this ward. Therefore, identification the causes of infection and antimicrobial susceptibility pattern in the ICU is essential to the process of controlling nosocomial infections in this department .Given the importance of *Escherichia coli* (*E. coli*) and *Klebsiella* bacteria in causing a major ratio of nosocomial infections in the ICU and their specific resistance pattern, determining resistance to ESBLs and carbapenemases as one of the objectives of this study to implement an appropriate strategy Its considered important to deal with such organisms

Methods: In this cross-sectional study, the antimicrobial susceptibility pattern of bacterial isolates from patients with nosocomial infections admitted to the ICU during 6 months was determined by disk diffusion method. Overall, 57 *E. coli* and *Klebsiella* isolated from different infection sites of patients with nosocomial infections were used to determine of ESBLs, Metallo- β -lactamase and carbapenemases resistances. Phenotypic screening and Confirming tests of ESBLs, Metallo- β -lactamase and carbapenemases enzymes was carried out. Detection of ESBL (*bla*_{CTX-M}, *bla*_{OXA11} and *bla*_{SHV}), Metallo β -lactamase (*bla*_{NDM}, *bla*_{VIM} and *bla*_{IMP}) and carbapenemase (*bla*_{KPC}, *bla*_{OXA-48} and *bla*_{OXA-23}) genes was also performed by the PCR method.

Results: A total of 103 different strains bacteria were isolated from 71 patients with nosocomial infections in the ICU. The most frequently isolated bacteria were *E. coli* 28.16%, *Acinetobacter* 14.56% and *Klebsiella* 12.26%. The most common source of infection was a respiratory tract infection of 50.48%, Also, the rate of multidrug resistant isolates was 51.45% (103.53). Phenotypic confirmation tests showed that 43 (75.43%) isolates of *E. coli* and *Klebsiella* produced ESBLs and 9 isolates (15.79%) were identified by MCIM phenotypic test as producers of metallo-betalactamase and carbapenemase. Molecular tests showed high prevalence of the *bla*_{CTX-M} 90.69% and low prevalence of the *bla*_{SHV} 20.93% in ESBLs genes. also *bla*_{NDM} and *bla*_{OXA-23} Genes were detected in 55.55% of isolates and *bla*_{OXA-48} and *bla*_{KPC} genes in 11.11% of isolates. The *bla*_{VIM} and *bla*_{IMP} genes were not detected in any of the isolates.

Conclusion: In this study, gram-negative bacteria with high phenotypic and genotypic resistance were the cause of a high percentage of nosocomial infections in the ICU. Therefore, finding and implementing an appropriate strategy in diagnosing, prescribing medication and monitoring infection control can be effective in reducing the rate of this resistance as well as the rate of nosocomial infection.



Keywords: Nosocomial infection; *Escherichia coli*; *Klebsiella*; ESBLs; Carbapenamase



Prevalence, Antibiotic Resistance Pattern and *in vitro* Analysis of Biofilm Formation of *Staphylococcus aureus* Isolated from Clinical Samples of Gorgan City, Iran

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Background and Aim: *Staphylococcus aureus* (*S. aureus*) is found in 30–50% of individuals, and due to high resistance to harsh environments and antibiotics, it's one of the major causes of nosocomial infections. Biofilm formation is an important virulence factor of *S. aureus* leading to its high resistance to disinfectants, antibiotics, and evasion from host defenses. Therefore, in this study, we aimed to assess the prevalence of biofilm-producing *S. aureus*, their antimicrobial-resistant profile, and characterization of genes involved in the biofilm formation.

Methods: A total of 79 *S. aureus* were isolated from 1000 clinical samples by applying the phenotypic, biochemical, and molecular tests. The biofilm production by isolates was examined by the microtiter assay. Moreover, the expression of genes involved in biofilm production includes PSM A and PSM B was screened by using the real-time PCR. Finally, Kirby-Bauer antibiotic susceptibility testing was performed and interpreted as per the CLSI standard

Results: Out of 79 *S. aureus* isolates, 43 (54.4%) isolates were strong biofilm producers, 21(26.6%) isolates were weak biofilm producers, and 15 (19%) isolates were found to be non-adhesive. The results of real-time PCR showed that 55 isolates (86%) were positive for the PSM A gene and 60 isolates (93.7) were positive for the PSM B gene. Results of DST illustrated that all of the isolates were resistant to two or more antibiotic agents

Conclusion: The high incidence of biofilm-forming *S. aureus* in hospital environments it's could be a major health challenge with serious outcomes for hospitalized patients. Thus, it is necessary to disinfect hospital surfaces and equipment to reduce the risk of infection and spreading of this microorganism.

Keywords: *Staphylococcus aureus*; biofilm; real-time PCR; antibiotic susceptibility

Promising of *Origanum majorana* Essential Oil Antibacterial Effect against Bacterial Pathogens

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Background and Aim: Antibiotic resistance is an important challenge for human health. *Pseudomonas aeruginosa* (*P. aeruginosa*) and *S. aureus* (*S. aureus*) are two important bacteria that cause many human infections. Green antibacterial components are as a main regarding new antibacterial agents against resistant strains.

Methods: In this research, potential of antibacterial effect of *Origanum majorana* essential oil (EO) were investigated. Major ingredients of this EO were Terpinene, 4-ol, cis- sabinene hydrate, γ -terpinene, α - terpinene and Trans- sabinene hydrate were identified as five major ingredients. Nanoemulsion-based nanogel of *Origanum majorana* EO was prepared and physically impregnated on the surface of the paper mats. Antibacterial effects of the prepared prototype at 2.5, 5, and 10 mg/mL concentrations were investigated against clinical and standard strains of *S. aureus* and *P. aeruginosa*.

Results: After 24h exposure, at 2.5, 5, 10 mg/mL concentration with 24 h exposure, the growth percentage of a standard strain of *S. aureus* 100, ~ 90, ~ 80 % was decreased, respectively. growth of other strains shown no significant reduction in the growth rate percentage.

Conclusion: The proposed component could be used as a green antibacterial agent as a surface antibacterial solution and wound dressing pad.

Keywords: Essential oil; Antibacterial

Respiratory Tract Bacterial Co-Infections and Antibiotic Management in Patients with COVID-19 Admitted to ICU in Central Iran. A Follow-Up Study

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Background and Aim: Bacterial co-infections are one of the important challenges in patients with COVID-19 that is associated with a high mortality rate. The current study was designed to assess bacterial co-infections and antibiotic management in COVID-19 patients admitted to ICU

Methods: In a cross-sectional study, 73 intubated COVID patients were included the study. The lung aspirate samples were collected in 2 stages and assessed for bacterial growth by standard methods. Antimicrobial testing was also performed based on the CLSI. Demographic and clinical data were collected. The statistical analysis was down by Chi-square test and Student's t-test and p-value < 0.05 was considered significant.

Results: Fourty men and 33 women with mean age 64.78 ± 13.90 were included in our study. The mean length of hospitalization and stay in ICU were 18.77 ± 12.94 and 13.51 ± 9.83 days, respectively. 84.9% cases were died. 32 patients had bacterial co-infection that was mainly caused by *Klebsiella* sp. and *Acinetobacter* sp. 64.5% MDR gram negative bacilli were isolated. A significant relationship was seen between co-infection COVID-19 and length of hospital stay until intubation.

Conclusion: Bacterial co-infection and mortality rate were relatively high in COVID-19 patients admitted to ICU. It seems infection control committee and antibiotic stewardship programs should be stronger than empiric antimicrobial therapy in hospitals.

Keywords: Bacterial; Co-infections; Antibiotic management; COVID-19 patients

Serum Level of Vitamin D, CRP and Biochemical Parameter in Acute and Chronic Brucellosis Treated with Doxycycline-loaded Solid Lipid Nanoparticles

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Background and Aim: Brucellosis is a common disease in humans and livestock in which treatment and relapse of this disease are one of the significant challenges. This study aimed to evaluate the therapeutic effect of doxycycline-loaded solid lipid nanoparticles (DOX-SLN) on acute and chronic brucellosis and its effect on serum level 25-OH-Vitamin D, C-reactive protein (CRP), and biochemical parameters

Methods: The double emulsion method was used for the synthesis of nanoparticles. Serum vitamin D levels, CRP, total protein, albumin, and globulin were measured in rats involved with brucellosis. The therapeutic effect of DOX-SLN was compared with that of free doxycycline.

Results: The mean size, Zeta potential, PDI, drug loading, and encapsulation efficiency of the synthesized nanoparticles were 299 ± 34 nm, -28.7 ± 3.2 mV, 0.29 ± 0.027 , $11.2 \pm 1.3\%$, and $94.9 \pm 3.2\%$, respectively. Serum vitamin D levels in rats with chronic brucellosis decreased compared to healthy rats. CRP increased in the acute and chronic stages of brucellosis. Serum levels of total protein and albumin were similar in both stages of the disease, but serum globulin levels were increased.

Conclusion: According to the results, the serum level of vitamin D was increased in rats after treatment with DOX-SLN. The use of DOX-SLN as an alternative drug for the treatment of chronic brucellosis is recommended.

Keywords: Brucellosis; Nanoparticles; Vitamin D; CRP; Total protein

Species Diversity, Molecular Characterization, and Antimicrobial Susceptibility of Opportunistic Actinomycetes Isolated from Immunocompromised and Healthy Patients of Markazi Province of Iran

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Background and Aim: Actinomycetes widely exist in nature and these species cause infections in immunocompromised and healthy patients, although they are frequently found as members of the normal microbiota of humans and animals. These subsequent infections are often misdiagnosed as malignancy and tuberculosis. Due to this issue, the present study aimed to determine the presence and diversity of actinomycetes species causing infections in Iranian patients.

Methods: A total of 79 clinical samples collected from five hospitals in Markazi province were analyzed for the existence of actinomycetes using standard protocols for isolation and characterization of the isolates. The conventional tests were used for preliminary identification, the PCR amplification of hsp65 gene, the specific region of the 16S rRNA, and sequence analyses of 16S rRNA were applied for the genus and species identification. MICs of the antimicrobial agent were determined by the broth microdilution method and interpreted according to the NCCLS guidelines.

Results: A total of 17 (21.51%) actinomycetes isolates were recovered from clinical samples. In other analyzed samples, eight (10.12%) gram-positive, 12 (15.18) gram-negative bacteria, and six (7.6) fungi isolates were recovered. The most prevalent actinomycetes species were *Mycobacterium fortuitum* (17.64%), *Nocardia Mexicana* and *S. heliomyces* (11.76% each), and 10 species, ie, *Nocardia farcinica*, *Mycobacterium lehmannii*, *Mycobacterium flavescens*, *Arthrobacter crystallopoietis*, *Nocardia neocaledoniensis*, *Mycobacterium phocaicum*, *Mycobacterium abscessus*, *Mycobacterium arupense*, *Mycobacterium setense*, and *Nocardia cyriacigeorgica* made up the single isolates. Results of DST illustrated that all of the isolates were susceptible to Amikacin, Levofloxacin, Ofloxacin, and Ciprofloxacin, whereas all of them were resistant to Rifampicin and Doxycycline.

Conclusion: In conclusion, increasing isolation of actinomycetes found in various clinical cases merits special attention by health authorities in developing countries. In health centers, action should be taken to increase awareness of appropriate diagnostic criteria and management guidelines for actinomycetes diseases. Furthermore, an increase in the number as well as the quality of national and regional reference laboratories may facilitate more accurate diagnosis of actinomycetes diseases.

Keywords: actinomycetes; 16SrRNA; drug susceptibility pattern; immunocompromised

Stroke and Urinary Tract Infection: Reconsidering the Strategies to be Implemented

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Background and Aim: Stroke is the leading cause of major long-term disability and urinary tract infections (UTIs) are one of its complications. Though many other bacterial infections are evident in these patients, nevertheless, these patients are more vulnerable to UTI due to increased risk from immunosuppression, bladder dysfunction, Foley catheter use and the fever and systemic inflammatory response associated with UTI. As a consequence, patients are forced to stay longer in hospital, face poor neurological outcomes and social and economic burden. This review attempted to discuss the risk factors and the interventional strategies to be implemented so as to avoid the infection. **Methods:** PubMed”, “Google Scholar” and “Medline” were searched using the MeSH terms stroke, fever, infection, stroke/complications, risk factors and urinary tract infection.

Results: Research studies have pointed that several influencing factors like sex, smoking, NIHSS score, interleukin-6, and hemoglobin play a crucial independent role in the development of UTI in stroke patients. Alternatively, smoking has been found a protective factor of urinary tract infection. Female has been observed as a risk factor for UTI. Elevated levels of interleukin-6, higher admission NIHSS score, and higher mRS and increasing age has been proposed as independent risk factors to predict UTI. Reducing the use of Foley’s catheter is hypothesized to decrease the risk of UTI progression. Usage of antibiotic-coated catheters have been suggested to prevent the bacterial adherence to catheter surfaces. *Escherichia coli*, *Klebsiella pneumoniae* and many gram positive bacteria are the etiological agents. Multidrug- resistance is an additional concern in therapeutics.

Conclusion: UTI is a common complication in stroke patients. The condition is associated with urinary catheterization and many risk factors have been found to be associated with the development of UTI. Interventional strategies are required to prevent either development or prevention to improve the quality of life of a patient.

Keywords: Risk factors; post-stroke infection; Stroke; Urinary tract infection; Outcomes

Study of CBC, CRP and ESR Changes in People with Brucellosis Referred to Gonabad Community Health Centers during the Years 2016-2019

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Background and Aim: Brucellosis is a major health and economic problem in many parts of the world, including the Middle East. Blood disorders such as anemia, leukopenia and thrombocytopenia can be seen in brucellosis. laboratory findings of this disease are different in different studies and populations. Therefore, this study was performed to investigate the changes in CBC, CRP and ESR in people with brucellosis referred to Gonabad community health centers during the years 2016-2019.

Methods: The present study is a cross-sectional study that was performed on 221 patients with brucellosis. According to the national guidelines for brucellosis, titers greater than 1/80 for Wright and more than 1/40 for 2-ME were considered as positive cases of brucellosis. Using the checklist, information related to CBC, CRP and ESR test parameters were collected and after entering the data into SPSS software version 16, the analysis was performed.

Results: The results showed that out of 221 patients studied, 58.4% were male and 41.6% were female. The mean age was 44.9 ± 19.8 years for men and 49.3 ± 17.3 years for women. High ESR was seen in 43.4% and positive CRP in 59.7% of patients. Leukopenia in 8.6%, leukocytosis in 9%, neutropenia in 6.8%, neutrophilia in 9.5%, lymphopenia in 3.6%, lymphocytosis in 10%, anemia in 17.2% and thrombocytopenia in 9.5% of patients were seen. 5% of patients had bicytopenia and pancytopenia was not seen in any of the patients. In this study, there was no relationship between wright titer and age ($P = 0.431$), incidence of leukopenia ($P = 0.474$), neutropenia ($P = 0.475$), lymphopenia ($P = 0.352$) and anemia ($P = 0.705$) But there was a significant relationship between wright titer with sex ($P = 0.004$), high ESR.

Conclusion: Brucellosis in endemic areas should be considered in the differential diagnosis of patients presenting with thrombocytopenia. Also in high Wright titers, CRP is more valuable indicator than ESR.

Keywords: Brucellosis; CBC; ESR; CRP

The Antibacterial Effect of the Collagen-hydrogel Scaffold of the Mouse Bone Marrow and Adiposed Mesenchymal Stem Cells on Burn Tissue Infected by *Pseudomonas aeruginosa* *in vivo*

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Background and Aim: Patients with burns are predisposed to a variety of microbial infections. Despite effective treatments for these infections, septicemia and pneumonia caused by burn wound infection can lead to death in some patients. *Pseudomonas aeruginosa* is the most common bacterium that causes burn infections and, the emergence of multidrug-resistant strains of Pseudomonaseae is one of the most concerning factors in treatment. Recently, the use of mesenchymal stem cells in the form of collagen hydrogel scaffolds has been proposed as a suitable option for the treatment of burn wound infection.

Methods: First, the stem cells from mouse adipose and bone marrow tissues we extracted were confirmed by the surface markers using flow cytometry analysis and encapsulate in collagen scaffolds. Then, following a burn infection in mice, Collagen-encapsulated mesenchymal stem cells applied to dress the wound. Finally, the bacterial load was monitored in infected mice by standard colony count test after one-week.

Results: The adipose mesenchymal stem cells had significant antimicrobial properties due to the reduction of bacterial load, while the bone marrow mesenchymal stem cells indicated a healing role on the burned wounds.

Conclusion: According to the results of this study and the emergence of bacterial antibiotic-resistance, the use of the collagen-hydrogel scaffold of the mouse bone marrow and adiposed mesenchymal stem cells is recommended as a suitable and new option for the treatment of *Pseudomonas aeruginosa* induced burn infections.

Keywords: Collagen; Scaffold; Mesenchymal Stem Cells; Antibacterial; Burn; *Pseudomonas aeruginosa*

The Emergence of Linezolid Resistant *Staphylococcus epidermidis* in the COVID-19 Hospitalized Intubated Patients in North Khorasan, Iran

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Background and Aim: In the COVID-19 pandemic from 2019 to date, we confront secondary bacterial and viral infections in SARS-CoV2 infected patients, especially hospitalized patients. Coagulase-negative staphylococci, are commensals of the human body and can lead to infections in immunocompromised patients. The antimicrobial resistance is increasingly reported in coagulase-negative staphylococci, especially in *Staphylococcus epidermidis* (*S. epidermidis*). One of the most critical problems is resistance to linezolid in *S. epidermidis*, observed in Europe since 2014. The aim of this study was to evaluation of bacterial Co-infections and determination of antimicrobial resistance pattern of co-infection isolated strains in North Khorasan, Iran, in the last six-month period.

Methods: After microbiological evaluation of pulmonary samples of hospitalized intubated patients with signs of bacterial pneumonia, we found co-infection in 11 of 185 patients with *S. epidermidis*, *Staphylococcus aureus*, and *Acinetobacter baumannii*, respectively. Interestingly seven of nine *S. epidermidis* isolates were linezolid resistant. For identification of the isolates at the species level, we used phenotypic methods and also the Polymerase Chain Reaction (PCR) for the *atfE* gene. Selected isolates were characterized by determining their antimicrobial resistance patterns and using molecular methods including SCCmec typing, detection of *ica*, *mecA*, *vanA*, and *cfr* genes.

Results: All isolates were resistant to methicillin, and Seven isolates were resistant to linezolid. It should be noted that all nine isolates were positive for the *ica* gene. Nine of 11 isolated have belonged to the SCCmec I, and two belonged to the SCCmec IV. It should be noted that all patients had the underlying disease and six patients died.

Conclusion: The increasing linezolid resistance in bacterial strains becomes a real threat for patients, and monitoring such infections combined with surveillance and infection prevention programs is very important to decrease the number of linezolid-resistant staphylococcal strains.

Keywords: *Staphylococcus epidermidis*; COVID-19; Antibiotic resistance; linezolid; vancomycin

The Global Prevalence of Daptomycin, Tigecycline, and Linezolid-Resistant *Enterococcus faecalis* and *Enterococcus faecium* Strains from Human Clinical Samples: A Systematic Review and Meta-Analysis

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Background and Aim: The predominant species of the Enterococcus, *Enterococcus faecalis* (*E. faecalis*) and *Enterococcus faecium* (*E. faecium*) cause great variety of infections. Therefore, the expansion of antimicrobial resistance in the Enterococcus is one of the most important global concerns. This study was conducted to investigate the prevalence of resistance to linezolid, tigecycline, and daptomycin among enterococcal strains isolated from human clinical specimens worldwide.

Methods: Several databases including Web of Science, EMBASE, and Medline (via PubMed), were carefully searched and reviewed for original research articles available in databases and published between 2000 and 2020. A total of 114 studies worldwide that address *E. faecalis* and *E. faecium* resistance to linezolid, tigecycline, and daptomycin were analyzed by STATA software.

Results: The overall prevalence of antibiotic-resistant *E. faecalis* and *E. faecium* was reported to be 0.9 and 0.6%, respectively. *E. faecalis* and *E. faecium* were more resistant to the linezolid (2.2%) and daptomycin (9%), respectively. The prevalence of tigecycline-resistant *E. faecium* (1%) strains was higher than *E. faecalis* strains (0.3%). Accordingly, the prevalence of linezolid resistant *E. faecalis* was higher in Asia (2.8%), while linezolid-resistant *E. faecium* was higher in the America (3.4%). Regarding tigecycline-resistance, a higher prevalence of *E. faecalis* (0.4%) and *E. faecium* (3.9%) was reported in Europe.

Conclusion: In conclusion, this meta-analysis shows that there is an emerging resistance in Enterococcus strains. Despite the rising resistance of enterococci to antibiotics, our results demonstrate that tigecycline, daptomycin, and linezolid can still be used for the treatment of enterococcal infections worldwide.

Keywords: *Enterococcus faecalis*; *Enterococcus faecium*; tigecyclin; daptomycin; linezolid

The Relation of Bacterial Vaginosis with the Serum Level of 25-Hydroxy Vitamin D in Women Referred to private clinics and gynecology clinic of Allamah-Bohloul-Gonabadi hospital during 2019- 2020

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Background and Aim: Bacterial vaginosis is one of the most common vaginal infections among women of reproductive age so that 40-50 percent of women at this age are affected. Factors such as micronutrient deficiencies such as vitamin D can increase the risk of bacterial vaginosis. The prevalence of bacterial vaginosis in childbearing age also the prevalence of vitamin D deficiency in Iranian women is high. So, this study was designed to investigate the relationship between bacterial vaginosis and serum vitamin D levels in women referred to the gynecology clinic of Allama-Behloul-Gonabadi Hospital and private clinics in 2019-2020.

Methods: In this case-control study, 250 married women were divided into the case group (with a complaint of vaginal discharge suspected of bacterial vaginosis) and the control group (healthy women with routine gynecological and obstetric care) were included in the study. Finally, the collected data were analyzed using conditional logistic regression model by the software of SPSS version 16 and STATA version 12.

Results: The results of multiple conditional logistic regression showed that after adjusting the effect of confounding variables, compared to people with adequate serum levels of vitamin D, whom are deficient in vitamin D or their vitamin D is inadequate, have a 4.3 and 3.6 times higher chance of developing bacterial vaginosis, respectively ($P = 0.011$ and $P = 0.020$, respectively). Also, people who smoke and hookah are 6.15 times more likely to get bacterial vaginosis than those who do not ($P = 0.014$). People who experienced first sexual contact at an older age had a lower chance of developing bacterial vaginosis so that for every one-year increase in the age of first sexual contact, their chance of getting it was 0.14 less ($P = 0.004$). Also, the chances of contracting bacterial vaginosis in people who used natural contraception and condoms were 0.80 and 0.78 less than those who did not have contraception ($P = 0.006$ and $P = 0.016$, respectively).

Conclusion: The results of this study showed that there is a significant relationship between bacterial vaginosis and serum levels of vitamin D in women referred to the gynecology clinic of Allameh Behloul Gonabadi Hospital and private clinics in 2019-2020. As a result, targeted screening may be effective in preventing diseases such as vaginosis and its complications.

Keywords: Bacterial vaginosis; 25-hydroxyvitamin D; Vitamin D deficiency

The Role of Gut Microbiota in the Carcinogenesis of Colorectal Cancer

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Background and Aim: Colorectal cancer (CRC) is one of the deadliest cancers in the world. Accumulating evidence has shown that gut microbiota is involved in CRC carcinogenesis. The aim of this study was to review the role of gut microbiota in the pathogenicity of CRC.

Methods: We've searched PubMed, Google scholar, SID and Magiran databases from the year 1990 to 2021 for keywords such as gut microbiota, CRC, bacteria, carcinogenesis and inflammation among published articles.

Results: Alteration in the composition and function of gut microbiota (dysbiosis) are associated with various diseases such as diabetes, autism, allergies and CRC. A significant number of studies have reported that the composition of the gut microbiota of people with CRC was completely different from those with healthy subjects. In various studies, levels of some bacteria such as colibactin-producing *Escherichia coli*, *Fusobacterium nucleatum*, *Streptococcus gallolyticus*, enterotoxigenic *Bacteroides fragilis* and *Helicobacter pylori*, were detected significantly higher in tumorous microenvironment compared to healthy tissues. The carcinogenic mechanism of these bacteria is not well understood in different animal models, but it seems that they are involved in the initiation and development of CRC by promoting chronic inflammation and exploiting virulence factors.

Conclusion: Bacterial microbiota plays a key role in CRC carcinogenesis but more research is needed to better understand their precise carcinogenic mechanisms.

Keywords: Gut microbiota; Colorectal cancer; Carcinogenesis

The Survey on *Acinetobacter baumannii* Antibiotic Resistance Pattern Obtained from Clinical Specimens of Shahid Beheshti Hospital in Kashan

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Background and Aim: *Acinetobacter* is a group of gram-negative coccobacilli that are widely distributed in nature and in the hospital environment and cause a wide range of nosocomial infections such as pneumonia, bacteremia, meningitis, skin and soft tissue infections, wound and burn site infections. *Acinetobacter baumannii*, as a species of this family that is responsible for various nosocomial infections, has shown resistance to various antimicrobial agents. The aim of this study was to identify and evaluate the patterns of antibiotic resistance of *Acinetobacter baumannii* isolates obtained from clinical samples of Shahid Beheshti Teaching Hospital in Kashan, Iran.

Methods: This descriptive cross-sectional study was performed on 102 *Acinetobacter* species isolated from hospitalized patients in Shahid Beheshti Hospital in Kashan. Standard biochemical tests were used to identify at the species level and PCR was used to identify the blaOXA51 gene to confirm and identify these isolates. Antibiotic susceptibility of 102 isolates was determined by standard method and according to CLSI criteria. MDR was defined as resistance to three common classes of antibiotics and XDR was defined as resistance to imipenem or meropenem in addition to the previous groups.

Results: Findings of our study showed that 96.1% of the patients received meropenem during their hospitalization, followed by vancomycin (78.4%), ciprofloxacin (72.5%) and levofloxacin (70.6%). Also, the findings of our study showed that the highest resistance was related to the antibiotics, piperacillin (1/93%) and meropenem (93.1%) and the antibiotics ciprofloxacin (91.2%) and imipenem and gentamicin (each with 89.2%) were in the next categories. they take. Also, the most sensitive among the studied antibiotics were related to minocycline (64.7%) and then tobramycin (30.4%). Finally, it was found that most of the samples are in the definition of XDR resistance pattern (92.2%). Also, no pattern related to MDR resistance pattern was found among the samples taken.

Conclusion: The results of our study showed that clinical isolates of *Acinetobacter* family, especially *Acinetobacter baumannii* as a hospital opportunistic pathogen have high resistance to antibiotics, especially ciprofloxacin and meropenem.

Keywords: *Acinetobacter baumannii*; Nosocomial infections; Antibiotics; Drug resistance

Trends of Healthcare Associated Bacterial Infections in COVID-19 Patients and Biofilm Production in Pathogen

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Background and Aim: During the setup of organization that was forced by the COVID-19 emergency, attention was paid to infection control measures. Increment in the bacterial infections is an ongoing concept in patients with severe COVID-19 infection after hospital admission. Evidence on the effect of the COVID-19 pandemic on healthcare-associated infections (HAIs) is still limited. In this study, we investigated the HAI type bacterial infections occurring in COVID-19 patients and the impact of biofilm formation by these organisms.

Methods: A total of 79 bacterial isolates obtained from various clinical infections in COVID-19 patients were taken into the study. These isolates were obtained from blood, catheterized urine, wound and endotracheal aspirates. Isolation and identification were done by utilizing the conventional bacteriological methods followed by antibiotic susceptibility assay by Kirby-Bauer method. Biofilm formation was analyzed in 96-well flat bottom polystyrene plates, under static conditions and biofilm specific genes were assessed genetically.

Results: During the period of 15 months of COVID-19 epidemic, *Escherichia coli* (n=18), *S. aureus* (n=15), *Acinetobacter baumannii* (n=28) and *Klebsiella pneumoniae* (n=18) were isolated from catheterized urine, endotracheal aspirate and blood specimens of COVID-19 patients. *Escherichia coli* was the bacterial pathogen isolated from catheterized urine send from all 18 patients. Sputum was not sent from any COVID-19 patient. *Enterococcus* spp., *S. epidermidis*, *Proteus* spp., *Stenotrophomonas maltophilia* and *Burkholderia cepacia* were not isolated from any COVID-19 patients while were etiologic agents of infections in other patients. Biofilm capacity was as follows: *Escherichia coli*-14.84% were strong biofilm producers with fimH gene positivity in 9.24% isolates, *S. aureus*-12.45% were biofilm producer with all isolates positive for *icaA* and *icaD* genes, 15.96% *Acinetobacter baumannii* were strong biofilm producers with all isolates positive for *bap* gene.

Conclusion: Presence of HAIs needs additional rethinking on the necessity of more controlled infection control measures. Biofilm positivity may be the foundation of infections.

Keywords: *Acinetobacter baumannii*; Staphylococcus; *Klebsiella pneumoniae*; *Escherichia coli*; Hospital acquired infection; COVID-19

Urinary Incontinence: Phenomenon of Dysbiosis in Urinary Microbiota

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Background and Aim: Urinary incontinence (UI), experienced by middle-aged to older women, has been overlooked in microbiological aspect. Urine which was once thought sterile, is now known to comprise its own microbiota. The female urinary tract however, shows variation in species and abundance depending upon the age, hormonal status and underlying urological condition. The expanded quantitative urine culture (EQUC) techniques and New Generation Sequencing (NGS) has facilitated and broadened our knowledge on this subject. This review attempted to characterize the microbiota with regard to UI and discuss the potential impact of bacteria on this condition.

Methods: Four search engines were selected to search the best literature research- Google scholar, PubMed, Science direct and Scopus. “Female urinary tract”, “Urinary Incontinence”, “Microbiota”, “Microbiome”, “Urinary tract infection”, and “microbial communities” were used for each search.

Results: Human urinary tract harbors a resident microbial community. More than 100 species from more than 50 genera are thought to reside in the human urinary tract. UI affect most frequently women of more than 44 years. Women afflicted with this condition suffers from sudden, intense urge to void which detracts the patient’s quality of life. When this condition turns chronic, socioeconomic burden adds psychological load on personal life. Though clinically known as a neuromuscular defect, the UI is actually a dysbiosis of urinary microbiota. The clinical diagnosis of UI requires exclusion of urinary tract infection. Decreased *Lactobacillus* and increased *Gardnerella* content is the basis of UI. Other bacterial flora which are focused in UI is presence of *Aerococcus*, *Arthrobacter*, *Staphylococcus*, *Actinomyces* and *Sneathi* to name a few. *L. gasseri* is predominantly detected in UI while, *L. crispatus* maintains the healthy urinary environment.

Conclusion: Microbiome is more diverse in UI. Those who do not respond to the treatment have greater bacterial diversity than who responds to the treatment.

Keywords: Microbiota; Urinary Incontinence; Urinary tract; EQUC; 16S rRNA

Virulence Genes of *Escherichia coli* Isolates from Urine samples Collected from Males in Kerman City

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Background and Aim: This study was purposed to determine the virulence genes of *Escherichia coli* isolates from males suspected to urinary tract diseases in Kerman city.

Methods: In this work, 16 *Escherichia coli* isolates were considered; all of these isolates have obtained from men suspected to urinary tract diseases. The isolates were reconfirmed using by biochemical tests. Presence and prevalence of selected virulence genes including *iucD*, *pap*, *sfa* and *hly* genes were determined by PCR.

Results: Among the examined isolates 12/16 (75%) were positive for one of the *sfa*, *hly*, *iucD* and *pap* genes. Six isolates (37.5%) contained *sfa* gene, 6 isolates (37.5%) had *iucD* gene, 4 isolates (25%) were positive for *pap* gene and none of the isolates were positive for *hly*. Totally 6 gene profiles were obtained, including: *iucD/sfa* (3 isolates; 18.75%), *pap/sfa* (2 isolates; 12.5%), *pap/iucD* (1 isolates; 6.25%), *sfa* (2 isolates; 12.5%), *pap* (2 isolates; 12.5%) and *iucD* (2 isolates; 12.5%). Four isolates (25%) were negative for all studied genes.

Conclusion: The results showed that most *Escherichia coli* strains obtained from male urine samples carry some important genetic virulence factors such as *sfa* and *iucD* genes.

Keywords: virulence genes; *Escherichia coli*; male; urine



Medical virology



Antiviral Effects of Punicalagin against Influenza A virus Infection in MDCK cells

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Background and Aim: The Influenza A virus is a serious threat to human health. Influenza complications are mild to serious, and can cause death in some cases. A great deal of attention has been paid in recent years to the development and use of new antiviral compounds for drug resistance in certain strains of the influenza virus and the clinical implications. The aim of this study was assessing the effect of Punicalagin on influenza virus A/Puerto Rico/8/34 (H1N1; PR8) in Madin-Darby canine kidney (MDCK) cells.

Methods: The anti-Influenza A virus activity of Punicalagin was evaluated in this experimental research using the Hemagglutinin assay (HA) and 50% tissue culture infective dose (TCID₅₀) method.

Results: The results showed that Punicalagin decreased virus titers ($P < 0.05$) and had an inhibitory effect on virus hemagglutination and the activity of viral polymerase.

Conclusion: Since punicalagin has antiviral effects in inhibiting hemagglutination and virus titration as well as virus binding in vitro, more animal studies and clinical trials are required to use the compound for treatment and prevention. It appears that this compound can be used as a complementary medication in conjunction with existing anti-influenza drugs in severe influenza infection to minimize the titer of the virus.

Keywords: Punicalagin; Anti-influenza; Influenza A virus; Antiviral activity

Human Respiratory Syncytial Virus in Children Less than 5 Years Old Hospitalized in Tabriz Childrens Hospital

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Background and Aim: Acute respiratory tract infection (ARTI) is a common reason for hospitalization in children that continues to threaten public health. ARTI is commonly caused by viruses, including human respiratory syncytial virus (HRSV), human Parainfluenza virus (HPIV), human bocavirus virus (HBoV), human coronavirus (HCoV), influenza virus (IFV), and adenovirus (HAdV). RSV is a common cause of acute respiratory tract infections in infants and toddlers that is a cause of bronchitis and pneumonia. The aim of in this study, is to detection of RSV among children less than 5 years old hospitalized in Tabriz Children's Hospital.

Methods: A total of 100 children less than 5 years old who had shown ARTI symptoms were recruited at the time of hospitalization from Tabriz Children's Hospital. Viral RNA was extracted from all samples using the RNX-PLUS kit and then complementary DNA (cDNA) synthesis was performed using cDNA synthesis kit according to manufacturer's instructions. cDNA from all samples was amplified by semi- Nested PCR technique by using the primers for N gene RSV. All amplified products were visualized on 1.5% agarose gel and the presence of a 180 base pair amplicon considered as RSV.

Results: Of 100 enrolled children (less than 5 years old), 44 were girls and 56 were boys. No significant difference was observed in gender. The age distribution of the study population was as follows: less than 12 months (53%), 13–36 months (33%), and 37–60 months (14%). RSV recognized in 18% (18/100) in patients with acute respiratory tract infection. 10 (55.6%) of the RSV-positive samples were boys, while 8 (44.4%) were girls. Most patients (61.1%) with RSV infection were less than 12 months old, and also RSV infections had occurred mainly during winter.

Conclusion: According to our findings, RSV can be one of the important respiratory pathogens in young children in northwestern Iran. Consequently, implementing a routine diagnosis for respiratory pathogens can improve the management of respiratory infections in children and help combat ARTI.

Keywords: RSV; ARTI; children

Molecular Prevalence of Human Papillomavirus Infection among Iranian Women with Breast Cancer

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Background and Aim: The etiology and molecular mechanisms involved in the development of breast cancer still remain poorly understood. Some epidemiological studies have shown an association between human papillomavirus (HPV) and breast cancer. However, the findings are controversial.

Methods: Our study was aimed to investigate the presence of HPV DNA in breast carcinomas of Iranian women. In total, 72 samples of formalin-fixed paraffin-embedded (FFPE) tissues of breast cancer collected between December 2014 and April 2016 were examined. HPV DNA detection was performed by nested-PCR assay. Next, positive samples were subjected to genotyping by the CLART HPV2 microarray system. All statistical analysis was carried out using SPSS V.18.0.

Results: HPV DNA was detected in 4/72 (5.55%) samples. Clinical factors were not statistically associated with HPV presence. However, CLART HPV2 microarray assay failed to determine the genotype of any positive samples.

Conclusion: The low frequency of HPV detected in our study does not support an association between breast carcinoma and HPV infection. However, it is possible that HPV may be responsible for breast carcinogenesis only in small percentage of all breast cancer.

Keywords: Human papillomavirus; Breast carcinoma; Iran; HPV; Breast cancer

The Study of Complete Sequence of sTAg of BK virus in Patients Receiving a Kidney Transplant in Ahvaz

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Background and Aim: Today, BKV infection is one of the most common infections in renal transplant recipients, and BK polyomavirus-associated nephropathy (BKVPN) is seen about 10% of patients who have had kidney transplants, which causes renal dysfunction and even rejection of the transplanted organ. BKV is common in serology and it is unclear whether a specific subtype or variant will play a role in its pathogenesis. sTAGs are another main proteins in the BK virus replication. **Aim:** In this study, we determined the sequence of the sTAG genes isolated from patients who received kidney transplantation and were admitted to hospital due to fever and increased creatinine.

Methods: Long PCR method with specific sTtAg primers was used. Then, purified fragments were cloned and sTAG regions of each sample were compared with homologous sequences in the Gene bank.

Results: Comparing the gene sTAG with reference strains of Dunlop, MM and SJH-LG-253, we found that the nucleotide difference was genetically very close to the Dunlop strain followed by the SJH-LG-253 strain, and is different from the MM in some genes, sTAG (Respectively 1, 7 and 1 nucleotides).

Conclusion: In this study, the complete sequences of sTtAg were aligned and compared to the standard strains of Dunlop, MM and SJH-LG-253. They were had very little difference in the sTtAg regions. And these genes are not suitable for determined the polymorphism.

Keywords: BK virus; sTAG; Kidney transplantation; Ahvaz

A Comparative Study of Clinical Signs and Laboratory Findings in Covid-19 Patients in Yasuj according to PCR Result in 2020.

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Background and Aim: Covid-19 is the most dangerous virus of the Corona viridae family, the first outbreak of Covid 19 (SARS-COV-2) was reported in 2019 in Wuhan, China. The aim of this study was to compare the clinical symptoms, laboratory and demographic findings of Covid 19 patients admitted to Shahid Jalil Hospital in Yasuj based on the results of RT-PCR test in 2020.

Methods: This study was a retrospective descriptive study, which was performed by investigation of the medical records of 330 patients. Patients had been confirmed as Covid19 by CT scan report and physician decision while some of them had positive real time PCR (RT-PCR) results. Data obtained from laboratory and clinical records of patients were analyzed using SPSS software version 22. Pearson Chi-Square and Fisher's Exact test were used to compare qualitative variables.

Results: Out of 330 patients, 173 (52.4%) were males and 157 (47.6%) females. The mean age of patients was 47.4 years and their age range were between 2 and 93 years. The average of hospital stay time was 6.3 days. The results of the present study showed that 83.9% of CT scan patients were positive. Impaired sense of smell was the most common clinical symptom among patients (73%). Then there was a disturbance in taste, cough and dyspnea of breath with a prevalence of 71.8%, 71.2% and 63.3%, respectively. Comparison of clinical signs in two groups patients PCR-RT+ and PCR-RT - showed that fever, headache, olfactory disturbance, taste disturbance, nightmare and chest heaviness between these two groups showed a statistically significant relationship. Analysis of laboratory results of the present study showed that, there was a statistically significant correlation between RT-PCR and neutrophil results, white blood cells, BUN and platelets.

Conclusion: The present study showed some of clinical and paraclinical findings of Covid19 may be associated with load of virus that can effect on the PCR result test. Loss of taste and smell in the early stages of the disease, where PCR is low due to low viral load, can be useful in diagnosing Covid 19 disease. Typical findings of chest CT scans seem to play a key role in screening and diagnosing people with negative RT-PCR results.

Keywords: Covid 19; RT-PCR; CT scan

A Comparison between Cryotherapy and Thermal Ablation to Treat Human papillomavirus (HPV) Infections

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Background and Aim: Human papillomavirus (HPV) infection is one of the most transmitting infections among viral diseases. Many people may have the HPV virus without any appeared symptoms but in some cases, it can cause health problems such as genital warts or various cancers. Fortunately, HPV infection can be prevented by vaccination and if the virus infects an individual, there are some other methods to cure it. This study aims to compare two cryotherapy and thermal ablation as treatments for HPV infection.

Results: According to collected data from researches, thermal ablation (including CO2 laser, Pulsed dye laser (PDL), Er-Yag, and Nd-Yag laser) had between 50-64% and cryotherapy had about 60% of success during these experiments. Also, some observations demonstrate the positive effect of using interferon alongside these treatments. Furthermore, HPV DNA was detected in cryo-treated lesions but was not detected in hyperthermia-treated lesions.

Conclusion: Thermal ablation had nearly the same result as cryotherapy but without any signs of HPV DNA. Also, PDL was the method that had the least adverse effect. Using interferon made the treatment more operative so it is suggested. Currently, one of the research projects was not powered to establish the affinity between the techniques, results from the ongoing RCT need to be confirmed.

Keywords: Human papillomavirus; cryotherapy; laser therapy; thermal ablation; genital wart

A Multi-center Retrospective Study of Clinical Features, Laboratory Characteristics, and Outcomes of 166 Hospitalized Children with Coronavirus Disease 2019 (COVID-19); A Preliminary Report from Iranian Network for Research in Viral Diseases (INRVD)

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Background and Aim: The objectives of this study were to analyze the clinical features and laboratory profiles and risk factors associated with critical illness of children with SARS-CoV-2. The objectives of this study were (i) to analyze the epidemiological characteristics (ii) to describe the clinical features and laboratory profiles and (iii) to investigate the risk factors associated with critical illness of 166 children with SARS-CoV-2 infection admitted to hospitals collaborative to the Iranian Network for Research in Viral Diseases (INRVD).

Methods: One hundred sixty-six COVID-19 Iranian pediatric patients were recruited through a collaborative research network between March and May 2020. Demographics, clinical, laboratory, and radiological results were obtained from patient files.

Results: Of 166 patients, 102 (61%) and 64 (39%) were males and females, respectively. Ninety-six (57.8%) and 70 (42.2%), had moderate and severe conditions, respectively. Thirty (18%) of patients died. The common symptoms were fever (73%), cough (54%) and shortness of breath, headache decrease in neutrophil and platelet counts; increase values in lactate dehydrogenase (LDH), decrease in the blood PH and HCO₃ were significantly associated with the disease severity. 54% and 56% of patients showed abnormal radiographic appearance in Chest X-ray and in chest computed tomography (CT) scan, respectively. Sixty-one (36.7%) of patients referred to ICU. The coexistence of comorbidity was the main factor associated with ICU admission, shock, arrhythmia, acute kidney injury, acute respiratory distress syndrome (ARDS), acute cardiac injury, and death.

Conclusion: We describe a higher than previously recognized rate of COVID-19 mortality in Iranian pediatric patients. Epidemiological factors, such as relatively high case fatality rate in the country and the presence of underlying diseases were the main factors for the high death rate.

Keywords: Severe Acute Respiratory Syndrome Coronavirus-2; COVID-19; Children

A Retrospective Study on the Gam-COVID-Vac (Sputnik V) COVID-19 Vaccine Short-term Efficacy and Side Effects in Healthcare Providers from Iran

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Background and Aim: The Gam-COVID-Vac (Sputnik V) COVID-19 vaccine is one of the approved vaccines which has been used in Iran. In the present retrospective study, we evaluated the short-term efficacy and side effects of Sputnik V vaccine against COVID-19 in Iranian healthcare providers.

Methods: We evaluated the side effects of vaccination with the Sputnik V vaccine after the second dose of vaccination in healthcare providers. Furthermore, the produced antibody levels were evaluated in this group of vaccine recipients. Vaccine recipients were divided into two groups based on the previous history of SARS-CoV2 infection.

Results: The mean Optic Density (OD) levels for SARS-CoV-2 IgG were higher in recipients with the history of COVID-19. The mean OD of the SARS-CoV-2 IgG antibody 3 months after the second dose in recipients with no confirmed history of SARS-CoV-2 infection and with the history of COVID-19 were 4 ± 2.9 and 6.6 ± 2.2 , respectively. There were 18 (27.7%) patients with vaccination breakthrough in our study.

Conclusion: The Sputnik V seems to induce high levels of antibody after the second dose; however, it has 27.7% vaccination breakthrough. Booster dose is recommended 6 months after the second dose of vaccination.

Keywords: COVID-19; Vaccine; Sputnik V

Acute and Post-acute Phase of COVID-19: Analyzing Expression Patterns of miRNA-29a-3p, 146a-3p, 155-5p, and let-7b-3p in PBMC

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Background and Aim: When a new pathogen, such as severe acute respiratory syndrome coronavirus 2, appears all novel information can aid in the process of monitoring and in the diagnosis of the coronavirus disease (COVID-19). The aim of the current study is to elucidate the specific miRNA profile which can act as new biomarkers for distinguishing acute COVID-19 disease from the healthy group and those in the post-acute phase of the COVID-19 disease.

Methods: The expression level of selected miRNAs including let-7b-3p, miR-29a-3p, miR-146a-3p and miR-155-5p were evaluated in peripheral blood mononuclear cells (PBMCs) of COVID-19 patients, in both the acute and post-acute COVID-19 phase of the disease and healthy groups, by real-time PCR assays. Specificity and sensitivity of miRNAs was tested by receiver operating characteristic (ROC) analysis in COVID-19 patients.

Results: The expression level of all miRNAs in COVID-19 patients was significantly higher than in the healthy group. Therefore, the expression pattern of miR-29a-3p, miR-146a-3p and let-7b-3p in the post-acute COVID-19 phase was significantly different from the acute COVID-19 phase. ROC analyses demonstrated that miR-29a-3p, -155-5p and -146a-3p may serve as the novel biomarker for COVID-19 diagnosis with high specificity and sensitivity. In addition, miR-29a-3p, and -146a-3p can maybe act as novel biomarkers for distinguishing acute from post-acute phase of COVID-19 disease.

Conclusion: The difference in miRNA expression pattern between COVID-19 patients and those in the healthy group, and between acute COVID-19 with post-acute COVID-19, suggested that cellular miRNAs could be used as promising biomarkers for diagnosis and monitoring of COVID-19.

Keywords: COVID-19; Biomarker; SARS-CoV-2; microRNA.

Assessment of the Role of *Epstein Barr virus* and Vitamin D in the Lichen Planus Patients and Healthy Controls

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Background and Aim: Lichen planus (LP) is a chronic inflammatory disease that affects mucocutaneous including skin, genital and oral mucosa. Although the main cause of LP is elusive, several risk factors such as genetic, drugs, and viral infections are suggested as disease triggers. The aim of this study was to investigate the presence of Epstein–Barr virus (EBV) and vitamin D level patients as two risk factors in LP Patients (pre-and post-treatment) in comparison with healthy controls.

Methods: A case-control study was designed on 20 patients with lichen planus (12 males and 8 females; mean age 39 ± 1.49 years) and 20 healthy controls (5 males and 15 females; mean age 36.15 ± 2.40 years). Peripheral blood mononuclear cells were isolated and examined for Epstein-Barr virus load by Real-Time PCR. To determine IgG antibody titers towards EBNA-1 and Vitamin D level by enzyme-linked immunosorbent assay plasma samples were used.

Results: Statistically, no significant difference was found in terms of EBV load in patients with LP when compared to healthy controls (P -value < 0.05). Evaluation of IgG antibody levels toward EBNA-1 And plasma level of vitamin D did not show any statistically significant difference between patients with LP in comparison with healthy controls (P -value < 0.05).

Conclusion: Our results did not support any role of EBV infection in LP patients although further comprehension studied with a large group of samples are warranted to evaluate the plausible role of EBV infection in the patients.

Keywords: EBV; Vitamin D; lichen planus

Association of ABO and Rhesus Blood Groups with the Severity of SARS-CoV-2 Infection: A Cross-Sectional Analytical Study

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Background and Aim: different blood group types as risk factors for Coronavirus Disease 2019 (COVID-19) infection, the association between blood groups and the COVID-19 infection. Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2), a new strain of RNA viruses causes coronavirus disease in human. Though some studies suggested it may not be the same in various societies with different genetic statuses. Also, no studies so far have investigated the relationship between COVID-19 severity and ABO blood groups in Iran and developing countries according to the World Health Organisation (WHO) criteria. To evaluate the association between blood types and the severity of COVID-19 infection.

Methods: This cross-sectional study enrolled 171 patients infected with SARS-CoV-2 {49 severe (severe or critical) and 122 non severe (mild to moderate)}, who were admitted to hospitals of Mazandaran University of Medical Sciences, Sari, Iran from April 2020 to June 2020. To evaluate the distribution of the blood group, 171 COVID-19 patients were compared with the reported data of Iranian population blood groups. The severity of COVID-19 infection was determined based on WHO criteria including clinical symptoms, radiological findings, and signs of organ dysfunction. The associations between ABO blood groups and the severity of COVID-19, were evaluated using Pearson's Chi-square.

Results: The distribution of the blood group in 171 patients with COVID-19 was not different compared to the reported general Iranian population blood group (p -value=0.344). Evaluation of the association between ABO blood groups and the severity of COVID-19 showed that patients with blood group type B developed severe COVID-19 infection compared to other blood types who showed mild or moderate conditions (p -value=0.048). Mortality due to COVID-19 was not statistically different between the ABO blood group and Rh (p -value=0.96 , p -value=0.27 respectively), but the frequency of patients with Rh-negative that needed intubation and mechanical ventilation was higher compared to Rh-positive patients (p -value=0.003).

Conclusion: A positive correlation was found between blood type B and COVID-19 severity. Also, mechanical ventilation was significantly more in Rh-negative patients.

Keywords: Intubation; Iran; Severity; Severe acute respiratory syndrome coronavirus 2

Comparison of the Diagnostic Value of One-Step and Two-Step Real-Time Reverse Transcription Polymerase Chain Reaction (Real time RT-PCR) to Detection of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2)

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Background and Aim: The gold standard test for the diagnosis of SARS-CoV-2 is the one-step RT-qPCR method, which requires expensive probes and one-step reagents and not always available.

Methods: In the current study, a cost-effective technique based on SYBR green to detection of SARS-CoV-2 was evaluated. So that the specific primers for S and N genes were designed for use in the SYBR green-based two-step RT-qPCR method and then, the cross-reactivity test was performed with positive samples for other coronavirus and respiratory viruses. Moreover, the analytical sensitivity test was carried out with 8 dilutions (10-fold) of SARS-CoV-2 for one-step and two-step RT-qPCR. Lastly, the SARS-CoV-2 clinical samples (n= 210) were tested by these two methods and Receiver operating characteristic (ROC) analysis was performed to investigate incremental diagnostic value of each genes in the study methods

Results: The two-step RT-qPCR did not show any amplification of the positive samples of other respiratory viruses; it was able to detect until 7th dilutions of the SARS-CoV-2 sample. Furthermore, two-step qPCR detected 90% of the positive samples correctly. ROC analysis revealed a diagnostic ability of two-step RT-qPCR method for SARS-CoV-2 with an area under ROC curve of ≥ 0.7 ($p < 0.05$) and relatively high sensitivity and specificity. Combination of N and S genes increased the sensitivity up to 88%, specificity up to 86% and area under the ROC curve up to 0.85 (95% confidence interval (95% CI): 0.72 to 0.93, $p=0.0461$)

Conclusion: Our findings indicated that the two-step method has comparable sensitivity and specificity to the one-step method. Therefore, this method can be considered as alternative diagnostic method for diagnosis and monitoring of COVID-19 patients.

Keywords: SARS-CoV-2; RT-qPCR; SYBR Green; TaqMan

Comparison of the Frequency of IL6-174G> C (rs1800795) Polymorphism in Patients with Covid-19 and Healthy Individuals

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Background and Aim: SARS-CoV-2 is the result of the spread of respiratory disease, which first occurred in Wuhan, China. In severe cases, there may be an initial delay in antiviral responses, followed by an increase in inflammatory cytokines and the movement of monocytes and neutrophils into the lungs, leading to a cytokine storm. These cytokines, including interleukins IL-1, IL-6, IL-12, and TNF- α . lead to increased vascular permeability, leading to respiratory failure. Due to the role of IL-6 gene polymorphism in respiratory diseases, and the polymorphism of this gene may be one of the factors involved in the deterioration of the Covid-19, so we evaluated the prevalence of polymorphism -174G> C (rs1800795) interleukin 6 gene in patients with Covid-19 admitted to Shohadaye Ashayer Hospital in Khorramabad city.

Methods: The mean age of Covid-19 positives was 53.75 ± 15.27 with 85 subjects, and the mean age of Covid-19 negative subjects was 51.41 ± 22.37 with 85 subjects. In the present study, to investigate the polymorphism of -174G> C (rs1800795) interleukin 6 gene by PCR-RFLP method, genomic DNA samples were extracted from peripheral blood leukocytes using a DNA extraction kit. The PCR reaction was then optimized using specific primers and the PCR products were enzymatically digested using NlaIII enzyme.

Results: In this study, in 57 (67.1%) of Covid-19 positive individuals, there was no polymorphism of -174G> C (rs1800795) interleukin 6 gene as GG genotype, and in 28 (32.9%) polymorphism as GC and CC genotypes were reported. Regarding healthy individuals without Covid-19 infection, in 55 people (64.7%) no polymorphism of -174G> C (rs1800795) interleukin 6 gene as GG genotype, and 30 people (35.3%) with the polymorphism as genotypes GC and CC, which were not statistically significant ($P = 0.872$). Among patients with pulmonary complications infected with Covid-19, in 33 patients (70.2%) there was no polymorphism of -174G> C (rs1800795) interleukin 6 gene as GG genotype, and 14 patients (29.8%) had polymorphism as GC and CC genotypes. In the group of people with pulmonary complications and healthy in terms of Covid-19, 17 people (47.2%) had no polymorphism and GG genotype, and 19 people (52.8%) had polymorphism as CC and GC genotypes. There was a statistically significant difference ($P = 0.043$).

Conclusion: The prevalence of pulmonary complications among individuals without polymorphism -174G> C (rs1800795) interleukin 6 gene as GG genotype is significantly different from individuals with the polymorphism as GC or CC genotypes due to the effect of this polymorphism on the rate of cytokine storm in this group of patients.



Keywords: SARS-CoV-2; Polymorphism; Interleukin 6



Coronavirus (SARS-CoV2) Drug Repurposing Based on System Biology

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Background and Aim: Since December 2019, the Novel Coronavirus, known as COVID-19, has been spreading over the world. It is a significant threat to every country's community healthcare system. It cannot be pointed how important it is to understand more about COVID-19's properties and interactions with human host cells in order to achieve successful COVID-19 therapies. Patients with mild sickness recover within a week; however, those with severe disease have major respiratory issues as a consequence of alveolar damage, which eventually leads to death, especially in the elderly with pre-existing disorders. Early identification and treatment are critical due to the disease's importance and high mortality rate. The antiviral medications used to treat coronavirus infections are only designed for short-term usage. Additionally, clinical studies for effective drugs and vaccinations take an extended period of time. Additionally, managing Corona viruses in vivo is often difficult and dangerous. The goal of this study was to identify the genes implicated in SARS-CoV2 and to repurpose prospective drugs for SARS-CoV2 therapy using co-expression network analysis. This research is using co-expression network analysis to find possible treatments for SARS-CoV2. These medications were offered as innovative possible therapies for Covid-19 and should be evaluated in future clinical research.

Methods: The Biogrid database was utilized to identify the genes that were associated with Covid-19. In the following step, we reconstruct the co-expression network using STRING database and visualize it using cytoscape software, and after that we create modules with the help of cytoscape software and the clusterviz plugin, and in the following step, we identify potential drugs using the DGIDB database, and after that we create a Drug-GENE network, finally we introduce high degree drugs into this network.

Results: First 260 differentially expressed genes were considered to be major genes. The STRING database was then used to create a gene co-expression network based on this basic gene list. Therefore, After the disconnected genes were eliminated, a network of 1189 genes was created. After analyzing the network in Cytoscape software and gene co-expression networks have been clustered, we discovered one significant module with 159 genes in the co-expression network. To find drugs that target genes in the module, the DGIDB database was applied. As a result, the genes for the modules were initially imported into DGIDB in distinct batches, In order to create a single drug-gene network, all of the drug-gene interactions were combined and gartered. Using this database, 4220 drugs were found for co-expression modules. Then, A drug-gene network was constructed using the

cytoscape software. In the drug- gene network, some genes have more than one drug. Also some drugs regulate two genes. Thus, these drugs are more important in drug- gene network.

Conclusion: We find out 122 covid-19 related genes affected by potential drugs and 6 genes have high degree in our network (AR, NFE2L2, VDR, HIF1A, MTOR and EGFR). Then we identify 29 potential drugs that have high degree in our network (RIBAVIRIN, ASPIRIN, METHOTREXATE, CISPLATIN, HYDROCORTISONE, SIROLIMUS, CYCLOSPORINE, INFLIXIMAB, SUNITINIB, SORAFENIB, DOXORUBICIN, METHIMAZOLE, ATORVASTATIN, PRAVASTATIN, INSULIN, METHYLPREDNISOLONE, TRETINOIN, PACLITAXEL, STREPTOZOCIN, DIGIXIN, PROGESTERONE, OMEPRAZOLE, SIMVASTATIN, STAUROSPORINE, ALDESLEUKIN, FENOFEBRATE, and CARBAMAZEPINE).

Keywords: Coronavirus; SARS-CoV2; COVID-19; drug repurposing; co-expression network; system biology; STRING database; DGIDB database



Correlation Between Single Nucleotide Polymorphism of Programmed Cell Death 1 Gene with Human T-cell Leukemia Virus Type 1 (HTLV-1)

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Background and Aim: Human T-cell leukemia virus type 1 (HTLV-1) is the first retrovirus that cause of disease in human. Programmed cell death-1 (PD-1) created exhaustion phenotype in T cells and disrupts antiviral responses so in this case-control research, for the first time, correlation between single nucleotide polymorphism of rs36084323 of PD-1 gene promoter with HTLV-1infection has been studied.

Methods: Genomic DNA of 81 HTLV-1 asymptomatic carriers (ACs)and 162 healthy controls (HCs) blood donors in Khorasan Razavi province were extracted and PD-1 rs36084323 polymorphism were genotyped by Polymerase chain reaction-restriction fragment length polymorphism (PCR- RFLP).

Results: The results indicated that ACs had higher frequency of GG genotype (59.3%) and HCs had higher frequency of GA+AA genotype(48.1%) in rs36084323 polymorphism of PD-1 gene but there was no significantly(P=0.27).

Conclusion: Our results point to a lack of association between rs36084323 polymorphism with susceptibility to HTLV-1 infection in the Iranian population.

Keywords: Genetic polymorphism; HTLV-1; Programmed death-1; viral load

COVID- 19 Reinfection in a Healthcare Worker after Exposure with High Dose of Virus: A Case Report

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Background and Aim: A novel coronavirus, severe acute respiratory syndrome coronavirus (SARS-CoV-2), has spread worldwide and leading the World Health Organization to proclaim a pandemic. To date, no specific vaccine or effective treatment has been found for this disease. Decreasing immunity happens in patients who have recovered from SARS-CoV-2 disease.

Methods: On June 21th 2020, a 36-year-old healthcare worker in charge of collecting samples at the Corona Diagnosis Center in Tabriz, Iran. On day 3 after symptom onset, he was tested positive for SARS-CoV-2 by qualitative real-time reverse transcriptase-polymerase chain reaction assay (qRT-PCR) of the ORF1N gene (BioGerm, China) in nasopharyngeal swab according to WHO interim guidance. On 25th August, when sampling a patient, coughing droplets sprayed on the face and eyes of this healthcare worker and caused to redness, inflammation, and eye infection. Sampling of the eye and nasopharyngeal was performed and RT- PCR was done for both ORF1ab and N.

Results: The result of RT- PCR was positive for both ORF1ab and N after eye infection in August. Therefore, the healthcare worker became re-infected with a very large volume of SARS-CoV-2 that was spread in his eyes.

Conclusion: This case demonstrated that re-infection can occur even just after a few months of recovery from the first infection. The finding indicates importance of continuous protection in healthcare workers.

Keywords: COVID- 19; pandemic; reinfection; SARS- CoV- 2; Case report

COVID-19 Infection in Children with Type 1 Diabetes

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Background and Aim: Coronavirus disease 2019 (COVID-19) spread around the world quickly. Chronic diseases, including diabetes (T1D), have been identified as a risk for COVID-19 complications. The significant evolving evidence in diabetes and COVID-19 is related to type 2 diabetes (T2D). Due to the limited information about COVID-19 in children with T1D, this study aimed to investigate COVID-19 in children with T1D.

Methods: This observational study was performed on 120 children under 18 years of age with T1D from February 2020 to September 2021. Data were collected from the clinics of Yazd Diabetes Research Center. All outpatients with T1D who were periodically visited at the center for follow-up diabetes were included in the study. The patients with new-onset diabetes during this period were excluded. Only children with suspected symptoms of COVID-19 infection or whose families were infected underwent COVID-19 PCR.

Results: Out of 120 children with T1D, eleven children (9%) were tested for COVID-19. PCR test of six children (5%) was positive. All children with COVID-19 infection had mild symptoms of COVID-19. Only one of them was hospitalized in the emergency department due to his mother's concern, and after 9 hours, he was discharged with normal chest X-rays and oxygen saturation, and the rest were treated on an outpatient basis. After proper medical treatment, these children with T1D and COVID-19 had no side effects.

Conclusion: Despite the concerns of parents of children with T1D about the effects of COVID-19 on their children, we found no evidence that children with T1D develop severe COVID-19.

Keywords: COVID-19; Type 1 Diabetes; Children

COVID19: Interaction Between Virus Spikes and Cell Receptor

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Background and Aim: Emerging of severe acute respiratory syndrome coronavirus 2 (SARS CoV-2 / Covid-19) has caused widespread worldwide outbreaks and a major public health problem. Understanding and recognition the viral structure of COVID-19 can help us to design vaccines and drugs for controlling and preventing this new acute respiratory syndrome. The attachment step in viral life cycle occurs when virus enters to body and penetrate into target cells. In this systematic review we want to explain how it will be occurred.

Methods: Research articles published concerning diagnosis of coronavirus disease, were analyzed and discussed to develop new clinical diagnosis tools In this systematic review we collected all data information by searching in NCBI and PubMed Central (PMC) and we use search engines like as: Science Direct, MEDLINE and ISI Web of Science. These Data were gathered from full- length articles.

Results: SARS-CoV-2 infects the host using the angiotensin converting enzyme (ACE2) receptor, which is expressed in several organs, including the lung, heart, kidney, Corona virus entry into target cells by its surface spike named "S" Protein. S protein is a type 1 glycoprotein that consists of S1 and S2 subunits and is present on the virion surface as a trimer. The S1 region is involved in receptor binding and contains N and C-terminal domains(S1-NTD and S1-CTD)respectively that may both act as receptor-binding domain (RBD),with the major determinants of cell tropism residing in S1-CTD. . the COVID-19 also might interact with some of the previously known host targets like as: CD26, Ezrin, Cyclophilins.

Conclusion: Our knowledge about the life cycle and pathogenicity of COVID-19 are increased Recognition All steps in virus replication can help us to controlling of this epidemic disease. Several strategy must be used for eradication of COVID-19. Prevention by social distancing, using face mask and herd immunity by vaccination, rapid diagnosis of patients and treatment by effective drugs such as RNA-dependent RNA .polymerase (RdRp) could help us to control and eradicate COVID-19.

Keywords: Coronavirus; COVID-19; Cell receptor; ACE2

Cytomegalovirus Active Infection in Persons Infected with Human Immunodeficiency Virus

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Background and Aim: Cytomegalovirus (CMV), one of the most common opportunistic pathogens in patients infected with human immunodeficiency virus (HIV), can cause the diseases such as encephalitis, pneumonia, and chorioretinitis. This study aimed at molecular studying of CMV infection in individuals infected with the human immunodeficiency virus.

Methods: In this study, 50 venous blood samples from HIV-infected individuals were taken. Patients were divided into two categories: patients under treatment with and without antiretroviral drugs. Plasma were separated from blood samples and examined for the presence of cytomegalovirus genome by PCR. Patients were divided into two group of under treatment with and without antiretroviral drugs.

Results: Of 50, 28 (% 56) were men and 22 (% 44) were women. CMV genome was identified in 8 samples (16%), and the molecular prevalence of CMV infection was 21.4% (n= 6) in males and 9.1% (n = 2) in females.

Conclusion: Given the frequency of Cytomegalovirus Active Infection in HIV-infected individuals under antiretroviral therapy, we should be careful about the treatment of Cytomegalovirus Active Infection.

Keywords: Active Infection; Cytomegalovirus; Human Immunodeficiency Virus; Shiraz; PCR

Detection of Epstein–Barr Virus in among Women Breast Cancer in Tabriz

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Background and Aim: After cervical cancer, breast cancer is the most common type of cancer among women. The cause of this cancer has not yet been fully identified, and several factors, including various environmental factors, including infectious agents, may play a role.. The role of viruses in breast cancer has not yet been well established, however, studies of the association and the presence of EBV in breast cancer, have had inconsistent, from 0% to 50%. The aim of this study, was investigating the EBV EBER gene in paraffin sample women with and without breast cancer in Tabriz.

Methods: In our study, we collected 80 paraffin-embedded malignant breast cancer tissue samples and 80 samples with non-cancerous tissues from the pathology lab in Tabriz. Deparaffinization and DNA extraction was performed using protocol mentioned in previous studies . The EBER region of the EBV genome was used as the primer for the detection of EBV in all samples by PCR .

Results: In our study, EBER gene of EBV was detected in 11.2% (9 out of 80) of breast cancer tissues and none in the non-cancerous tissues. The highest frequency of EBV – EBER in breast cancer samples was observed in Invasive Ductal Carcinoma II (10.9%) and in ≤ 50 age group (13.2%).

Conclusion: Basie of our finding, our results showed a possible role of EBV infection in breast cancer in Tabriz. These patients probably are infected with EBV at an early age, that the virus remains in their bodies, and that it may have contributed to cancer progression so However the controversy remains. Additionally, the exact mechanism of the potential role of EBV in breast cancer is yet to be elucidated.

Keywords: EBV; Breast cancer; Invasive Ductal Carcinoma II

Detection of Human Papillomavirus (HPV) Infections in Iranian Women with and without Abnormal Cervical Cytology by Multiplex PCR

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Background and Aim: Infection with certain human papillomaviruses (HPV) is strongly associated with the development of dysplasia and cancer of the cervix uteri. The average global prevalence of HPV infection has been reported to be almost 1.4–25.6% in different geographical regions. To date, more than 200 HPV genotypes have been identified, and more than forty of these have been shown to cause anogenital infections in humans. The aim of this study was to estimate the prevalence of HPV in 350 cervical samples from patients who underwent annual routine gynecological exams by Multiplex PCR from different geographical regions in Iran.

Methods: Cervical cells were collected from 12 different city in Iran and were stored at 4°C until DNA extraction. The extraction of total DNA was performed according to the manufacturer's instruction (Add bio, Korea) and HPV genotyping by multiplex PCR method. The data were recorded using Microsoft Excel 2007 (Microsoft Corp, Redmond, WA, USA) and analyzed with the SPSS software (version 16; SPSS Inc., Chicago, IL, USA). P value < 0.05 was considered statistically significant.

Results: In total, Three hundred and fifty women between 18 and 77 years (mean age: 34 years) were screened, 112 (32 %) were HPV-DNA positive. A total of 23 low-risk and 65 high-risk HPV isolates were identified. HPV high-risk type 16 showed the highest prevalence (19.64%), while in low risk samples, HPV type 6 showed the highest prevalence 33.03%. Demographic data of positive HPV patients showed that 4.46%, 6.25%, 5.37%, 3.57%, 6.25%, 22.32% and 3.57% with presence of raised lesions/discharge, abnormal bleeding, history use oral contraceptives pills, abnormal pap smear, menstrual irregularities, delivery and abortion, respectively.

Conclusion: HPV detection is important for the selection of prevention strategies and improving approaches to combating cervical cancer. Our Multiplex PCR assay will be useful to address questions related to viral persistence at the genotype level, the kinetics of viral load and disease recurrence. Evidence is accumulating that HPV genotyping may be useful for patient management in the future.

Keywords: Multiplex PCR; Human papillomavirus; cervical cancer

Determining the Association of Polymorphism C1596T(rs74810361) in the Promoter of IP-10 Gene with Infection of Acute Respiratory Syndrome-Corona Virus- Type 2(Sars-Cov-2)

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Background and Aim: The appearance of acute respiratory syndrome caused by SARS coronavirus 2 in human populations caused the outbreak of a global epidemic in 2019. The body releases cytokines to confront the virus. In severe Covid-19 patients, serum levels of il -7, il -6, MIG, MCP -3, MCP -1, IP -10, MIP -1, G -CSF, M -CSF are higher than in mild cases. Genetic factors, in addition to patient's age, as well as underlying diseases can play a role in susceptibility or resistance to the virus. variations in ip-10 expression (chemokine cxcl-10 or interferon-gamma-induced protein) are associated with inflammatory diseases such as infectious disease, immune dysfunction, and tumorigenesis. Therefore, in this study we examined the polymorphism C1596T (rs74810361) in the ip-10 gene promoter.

Methods: In this case-control study, genomic DNA of 50 covid-19 patients from Babol and 50 control groups from blood donors in Tehran were extracted, and polymorphism C1596T (rs74810361) were genotyped by polymerase chain reaction-restriction fragment polymorphism (PCR-RFLP). The accuracy of genotyping data for the SNP obtained from PCR/restriction fragment length polymorphism analysis was validated by direct sequencing of random sample of patients.

Results: the results indicated that no significant accompany between rs74810361 polymorphism and the corona virus disease.

Conclusion: Therefore, treatment and prevention of the progression of this pandemic disease is one of the most important goals of disease control in the world. This study can examine one of the risk factors that is the genetic background of individuals as a background for predicting viral responses and personal medicine and thus controlling the mortality rate from infection with this virus.

Keywords: ip-10(cxcl10); polymorphism C1596T; sars-cov-2

Determining the Association of Polymorphism C1596T(rs74810361) in the Promoter of IP-10 Gene with the Severity of Acute Respiratory Syndrome-Corona Virus- Type 2(Sars-Cov-2)

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Background and Aim: COVID-19 is coated single-stranded RNA beta-corona virus that was reported in China in late 2019 and then became a global pandemic. According to WHO reports, the course of the disease is divided into the following stages: asymptomatic stage, early stage of infection (mild) in the form of fever less than 38 degrees, sore throat, nausea, vomiting and 93% oxygen level, stage two (Respiratory phase) into 2 groups: moderate (between 90-93%) and severe (less than 90%) and increased lung involvement by more than 50%. The third stage (inflammation exacerbation phase) that requires special care and the oxygen level is 88%. Hyperinflammation is associated with the severity of COVID-19, which is characterized by cytokine release syndrome (CRS). Some inflammatory cytokines, such as IL-6, IL-10, chemokines, as well as ip-10, cxcl-10 increases in covid-19 patients. Chemokine cxcl-10 is a cytokine belonging to the c-x-c chemokine family. Changes in cxcl-10 expression are associated with inflammatory diseases such as infectious disease, immune dysfunction, and tumorigenesis. SNP in the ip-10 promoter can regulate ip-10 expression by altering its binding affinity and thus affect disease progression. In this study correlation between single nucleotide polymorphism of rs74810361 and severity of symptoms in covid-19 disease has been studied.

Methods: Genomic DNA of 50 covid-19 patients in babol city were extracted and polymorphism C1596T (rs74810361) were genotyped by polymerase chain reaction-restriction fragment polymorphism (PCR-RFLP). The accuracy of genotyping data for the SNP obtained from PCR/restriction fragment length polymorphism analysis was validated by direct sequencing of random sample of patients.

Results: the results indicated that there was no significant accompany between rs74810361 polymorphism and the corona virus disease.

Conclusion: Therefore, treatment and prevention of the progression of this pandemic disease is one of the most important goals of disease control in the world. This study can examine one of the risk factors that is the genetic background of individuals as a background for predicting viral responses and personal medicine and thus controlling the mortality rate from infection with this virus.

Keywords: ip-10; severity; corona virus disease; C1596T polymorphism

Different Aspects of the Ethical Challenges of the COVID-19 Pandemic

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Background and Aim: The Covid-19 Pandemic is a global warning. In the event of future pandemics or COVID-19 waves, it is critical to focus on the ethical problems that have been raised and continue to be raised throughout this COVID-19 tension. There are ethical challenges to COVID-19 in many ways. How governments treat people in relation to drug quotas or mandatory and optional vaccinations, ethical issues related to drug research and testing and reporting of their results, equal access of countries to drugs, prioritization of jobs and individuals to receive vaccines, how to treat patients and their families, Forcing people alone and things like that. All ethical dimensions must be studied.

Results: The medical field's typical ethical standards have inevitably been thrown off. Healthcare personnel, patients, and their families experience are worry or disarray as a result of such interruption. Despite all of the attempts to develop equitable laws, many of the policies that have been implemented can be criticized for being insensitive to parts of society's demands. COVID- 19 vaccinations remain a critical weapon in the fight against the world's deadliest modern-day epidemic. However, the vaccine is not available to everyone.

Conclusion: There are still several ethical considerations about preparedness, knowledge sharing, intellectual property rights, and environmental health between countries. The most important principles we require to fight the epidemic are local and global solidarity, honesty, and collegiality. Bioethicists and other healthcare professionals must absorb the lessons of COVID-19 and put them into practice as quickly as possible in order to be better prepared for the next pandemic, which is unavoidable.

Keywords: COVID-19; ethical challenges; ethical principles; Public health

Downregulation of gsk3 β and Upregulation of urg7 in Hepatitis b-Related Hepatocellular Carcinoma

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Background and Aim: Hepatitis B virus (HBV) is the leading cause of hepatocellular carcinoma (HCC). The exact molecular contributors to the development of HBV-related HCC are not yet completely understood. Recent studies demonstrated that the deregulation of the Wnt pathway is highly associated with the development of HCC. Besides, HBV is known to have roles in the deregulation of this pathway. The present study evaluated the molecular aspects of the Wnt pathway in HBV-related HCC in liver tissue samples.

Methods: Viral characterization was done by identifying the HBx mutations and the assessment of intrahepatic viral load using an in-house real time PCR assay. The expression of Wnt pathway genes was evaluated using real-time PCR and methylation-specific PCR.

Results: The intrahepatic viral load was significantly higher in tumor samples than in normal tissues ($P = 0.0008$). Aberrant expression was observed in Wnt-1, Wnt-7a, FZD2, FZD7, β -catenin, URG7, c-Myc, SFRP5, and GSK3 β , among which Wnt1, FZD2, SFRP5, Gsk3 β , and URG7 were associated with HBV. HBx mutations at positions I88, L116, and I127 + F132 were associated with the decreased expression of GSK3 β and overexpression of URG7 and Wnt1. Alterations in the expression level of β -catenin, as well as some mutants of HBx, were correlated with the level of c-Myc.

Conclusion: HBV-related HCC seems to be mostly coordinated with epigenetic behaviors of HBx, such a multi-functional peptide with suppressing/trans-activating functions.

Keywords: HBV; HCC; Wnt pathway; HBx; mutation; gene expression

Downregulation of miR-143/145 Cluster in Breast Carcinoma Specimens: Putative Role of DNA Oncoviruses

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Background and Aim: The DNA oncoviruses, including human papillomavirus (HPV) and Epstein-Barr virus (EBV) are among the most important infectious agents involved in breast carcinogenesis. These oncoviruses have a broad disrupting effect on cellular miRNAs and their functions, by which they contribute to carcinogenesis. In this investigation, we evaluated the correlation between HPV and EBV and the expression level of tumor suppressor miRNAs (miR-143 and 145), clinical outcomes, and their association with stimulating inflammatory cytokines in patients with breast carcinoma.

Methods: In our case-control study, 35 cancerous tissues and 35 adjacent non-cancerous tissues were collected from 35 patients. Nested-PCR was set up for the detection of HPV and EBV genomes, and RT-qPCR was used for miRNA expression in the case and control groups. In addition, serum specimens were obtained from all patients (n = 35) and healthy controls (n = 35) to determine the IL-8 serum concentration.

Results: We found HPV and EBV in 14.2% (10/70) and 7% (5/70) of all samples, respectively. The distributions of positive samples in the case and control groups were 25.7% (9/35) and 2.9% (1/35) (P = 0.006) for HPV and 11.4% (4/35) and 2.9% (1/35) (P = 0.164) for EBV, respectively. Besides, RT-qPCR showed that miR-143 and miR-145 were significantly downregulated in HPV and EBV-infected cases compared to non-infected ones (P < 0.05). Data also indicated that the promotion of metastasis status was related to miR-143/145 downregulation and HPV infection (P = 0.003). No significant difference was found in serum IL-8 concentration concerning viral infections.

Conclusion: Our results suggested the possible involvement of viral infections in breast carcinogenesis and adverse clinical outcomes by downregulating miR-143/145

Keywords: Breast Neoplasms; Human Herpesvirus; Human MicroRNA-143; Human MicroRNA-145; Papillomaviridae

Effect of Baseline Resistance-Associated Substitutions on Thalassemia Patients with Chronic HCV Infection: A Two- Year Follow-Up

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Background and Aim: Direct-acting antivirals (DAAs) against hepatitis C virus (HCV) infection showed the presence of resistant-associated substitutions (RASs). The aim of the present study was to carry out a follow-up of patients with baseline RASs to report the impact of RASs on DAA therapy outcome.

Methods: In a cohort study, we analyzed NS5A and NS5B RASs among nine thalassemia cases by baseline RASs. In a 2-year follow-up, we analyzed viral markers and biochemical and hematological parameters of the participants and their sustained virologic response (SVR). Statistical analyses were performed using SPSS software version 22.

Results: RASs for HCV subtype 1a included M28V, L31M, and H58P. For subtype 1b: L28M, R30Q, S24F, and C316N. And for subtype 3a: C316S, and S24F. In patients with cirrhosis ($n = 5$), ALT ($p = 0.001$) and AST ($p = 0.007$) levels were significantly reduced after treatment, and creatinine level slightly increased ($p = 0.025$). However, no significant data was observed in non-cirrhotic patients following the treatment.

Conclusion: The present study did not show any adverse effects of DAA therapy among patients with thalassemia suffering from chronic HCV infection with baseline RASs. Furthermore, reduction in ferritin and liver stiffness levels after DAA therapy could show the efficacy of DAA in such patients.

Keywords: Direct acting antivirals (DAAs); Resistant associated substitutions (RASs); Chronic HCV infection

Evaluation of Anamnestic Response to Hepatitis B Booster among First-Year University Students Aged 18 to 20 Years Immunized During Infancy in Shiraz, Southwest of Iran

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Background and Aim: The antibody titer falls several decades after vaccination against the hepatitis B virus (HBV) during infancy. Hence, booster doses are required to reinforce the antibody levels, especially in high-risk groups like healthcare students. Therefore, this study aimed to evaluate the anamnestic response to hepatitis B boosters among anti-HBs antibody (anti-HBsAb)-deficient first-year university students in Shiraz, southwest of Iran, all of whom were immunized during infancy.

Methods: In this cross-sectional study, 164 first-year healthcare students of Shiraz University of Medical Sciences with anti-HBs < 10 mIU/ml were referred to the student health clinic for the administration of three 20 mg HBV vaccine booster doses. One month after the third dose, ELISA was performed for evaluating the anti-HBsAb titers.

Results: Out of 164 students, 77 (47%) were male, and 87 (53%) were female. ELISA results indicated that all students showed an anamnestic response (anti-HBs Ab > 10 mIU/ml), with a geometric mean concentration (GMC) of 246.38 mIU/ml. No significant difference was observed between genders in the level of the response ($P = 0.58$). The moderate and high response rates were 1.22% (2/164) and 98.78% (162/164), respectively.

Conclusion: The results indicated that an appropriate anamnestic response was induced in all students, representing the high efficacy of the HBV booster doses.

Keywords: Hepatitis B virus; vaccine; booster dose; anti-HBs Ab titer; immunity

Evaluation of Genomic Changes in Neuraminidase Gene and Resistance to Oseltamivir Influenza Virus B Isolated from Patients with Acute Respiratory Infection Referred to Shiraz Teaching Hospitals During 2017-2018

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Background and Aim: One of the most important viruses for acute respiratory infection is influenza virus which has three common types including A, B and C. Influenza virus type B has been less studied because of low grade pathogenicity and genetic diversity. As antiviral therapy by Oseltamivir is widely used in these cases, it may induce mutation and further resistance variants. In this regards, we aimed to investigate neuraminidase inhibitor resistance mutations in influenza B virus isolates obtained from patients with acute respiratory infection referred to Shiraz teaching hospitals, Shiraz, Iran, during 2017-2018.

Methods: This cross-sectional study performed on 235 children under 15 years by acute respiratory infection. After RNA extraction, one-step Real-Time PCR by designed specific primers and probe for Influenza B virus was carried out. Then, the positive samples were amplified by a nested PCR and the amplicons sent to Nucleotide Sequencing (One BEST co., Korea). Raw data trimmed and analyzed by CLC workbench 5 and used for resistance mutation detection.

Results: Out of 235 specimens, 23 were identified by influenza B infection. Of them, 17 were sequenced and found that the most common mutations were K220N and N340D, however they were not described previously as resistance mutations to Oseltamivir. Phylogenetic tree showed that our strains were serotypes B / Brisbane / 60/2008 (Victoria category) and B / Perth / 211/2001 (Yamagata category) in relation to Clade 1 and Clade 2.

Conclusion: According to the results, investigated region of neuraminidase gene have not shown any resistance mutation to neuraminidase inhibitors. However, broad spectrum resistant surveillance studies by more sample size and whole genome sequencing strategies should be done to comprehend the result.

Keywords: Influenza B; Neuraminidase inhibitor; Drug resistance; Mutation

Evaluation of Immune Status among First-Year University Students Aged 18 to 20 Years Immunized During Infancy in Shiraz, Southwest of Iran

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Background and Aim: Hepatitis B virus (HBV) vaccination is a safe and cost-effective strategy to control and prevent HBV infections. The anti-HBs antibody (anti-HBsAb) titer reduces over time, resulting in decreased immunity to HBV. Therefore, this study evaluated the anti-HBsAb titer among first-year university students immunized during infancy in Shiraz, southwest of Iran.

Methods: In this cross-sectional study, blood samples were collected from 679 first-year university students admitted to Shiraz University of Medical Sciences in 2016. All students had received standard HBV vaccination in infancy. Anti-HBsAb titers were evaluated using ELISA.

Results: Out of 679 students, 47.4% (322/679) were males, and 52.6% (357/679) were females. The mean age was 18.84 years. The results showed that 68.9% (468/679), 23.1% (161/679), and 7.4% (50/679) of students had anti-HBsAb titers < 10 mIU/mL, 10-100 mIU/mL, and >100 mIU/mL, respectively. The mean anti-HBsAb titer was 25.4 mIU/mL. Statistical analysis showed no significant difference between protective antibody levels in male and female groups ($P = 0.55$).

Conclusion: This study showed that 68.9% of first-year university students admitted to Shiraz University of Medical Science do not have a protective level of anti-HBsAb. Therefore, booster doses are recommended for this group.

Keywords: Hepatitis B virus vaccine; anti-HBs Ab titer; immunity

Evaluation of the Effect of Covid-19 Virus on Pregnant Women and Fetuses

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Background and Aim: Pregnant women are one of the groups most at risk for Covid-19 virus. This study examines the findings related to the impact of coronavirus on pregnant women and their fetuses, diagnostic methods, safety protocols in pregnant women and postpartum care.

Methods: The present study is a kind of systematic review based on the information collected from searching Internet resources in Persian language databases using the keywords Covid-19, pregnancy, pregnant women and fetus, from the papers accepted at the conference, World-renowned protocols, updated protocols and accurate statistics extracted. Initially, 100 articles were reviewed, but after applying the necessary filters, this number was reduced to 6.

Results: This study examined the findings related to the effect of corona during pregnancy on pregnant women and their fetuses, and Results: also provided information on how to diagnose the disease in pregnant women and their care protocols. The information of this article is based on a review of the results and updated and valid statistics.

Conclusion: Due to the global prevalence of this virus, protocols and health care applied to other people should be done for pregnant women as well. It is also necessary to take preventive measures at the community level in relation to pregnant women and monitoring systems for pregnant and non-pregnant women to be specifically monitored and followed up during pregnancy.

Keywords: Covid-19; pregnancy; pregnant women; Fetus

Evaluation of the Frequency of Angiotensin Converter Enzyme-2 (ACE2) Gene Polymorphism in COVID-19 Patients

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Background and Aim: SARS-CoV-2, a new member of the human coronavirus, was first developed in Wuhan, China in 2019. SARS-CoV-2 binds to the ACE2 receptor on the surface of host cells through the spike proteins. ACE2 is expressed on the epithelial cells in the lung, intestine, kidney, and blood vessels. The angiotensin-converting enzyme (ACE or ACE1) converted Angiotensin-I to Angiotensin-II, and ACE2 as the negative regulator of the RAS hydrolyzes Angiotensin-II into Ang-1-7. ACE2 gene has a high degree of genetic polymorphisms. Since the entry of SARS-CoV-2 is through the ACE2 receptor, ACE2 polymorphism may affect the severity of the disease and the potential effect of the ACE2 polymorphism is unknown in COVID-19, therefore, this study aimed to investigate the frequency of the ACE2 polymorphism in COVID-19 patients.

Methods: ACE2 allele and genotype rs2106809 polymorphism in 144 COVID-19 patients (72 males and 72 females) was analyzed using PCR-RFLP methods.

Results: The result of our study showed that the frequency of the ACE2 genotype A-A, A-G, G-G in males 76.38 % (55/72), 0% (0/72), 25% (18/72), and females 36.1%(26/72), 43% (31/72), 26.3% (9/72) respectively.

Conclusion: The data obtained from this study may be helpful for the relationship between this polymorphism and COVID-19 disease and maybe this polymorphism can be considered as a risk factor for disease severity.

Keywords: Covid-19; ACE2; Polymorphism

Evaluation of Varicella-Zoster Virus in Leukemias by PCR Molecular Method

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Background and Aim: Leukemia begins with the atrophy of bone marrow and progresses through the destruction of myeloid or lymphoid lineages over time. In immunocompromised patients and children with leukemia, there is a potential for Varicella-Zoster virus. In this study, among the DNA-based methods, PCR has been used to evaluate the DNA of Varicella-Zoster virus in the serum of patients with leukemia, which is one of the most accurate methods approved by the WHO.

Methods: The PCR test was optimized for VZV detection and was evaluated in terms of specificity and limit of detection (LOD). The DNA was extracted from 100 serum specimens of patients with leukemia and 100 healthy control specimens (positive and negative) from Tehran laboratories by phenol-chloroform. The product of PCR was detected using Agarose gel electrophoresis method.

Results: The varicella-zoster virus didn't observe in all healthy controls, but it was seen in nine percent (9%) of patients' specimens. The PCR test was optimized and 216-bp product was observed in agarose gel electrophoresis (Fig 1). In specificity test, there was no product other than the VZV specimen (Fig 2). A detection limit of 10 Copy / Reaction was reported (Fig 3).

Conclusion: In leukemia, besides environmental and genetic factors, infectious agents such as some viruses are effective. According to this study, VZV, after several periods of proliferation and accumulation in the skin, can be considered as an effective infectious agent in leukemia due to its severe pathogenicity.

Keywords: Varicella-Zoster virus; PCR; Leukemia

Explore Susceptibility to Covid 19 in Pregnant Women

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Background and Aim: Coronavirus as a cause of acute respiratory disease has risks for people with weakened immune systems, including pregnant women. Because of pregnancy is a special state in women and Pregnant women are susceptible to infection, and impaired mechanical and immunological functions. At present study we determined the causes of susceptibility of pregnant women to coronavirus using the review of other articles.

Methods: At present study , articles selected from different databases including Pubmed, Google scholar, Medline, Web of science. Published papers were searched in PubMed using the following keywords in combination : Risk factors , Covid 19 , Pregnancy, Pregnant women , Cytokine storm. We included published papers between Aug/2020_ Dec/2021. Two steps were performed to screen the articles for the present study. At first screened the titles and abstracts of papers and then assessed the full text of selected studies in details.

Results: Physiological changes in pregnant women such as increased diaphragm, increased oxygen consumption and swelling of mucous membranes of respiratory tract caused to prone to respiratory disease and pneumonia. Perinatal is result of hypoxemia in pregnant women, other complications including fetal distress, preterm delivery, respiratory distress, thrombocytopenia. On the other hand, hormonal changes in this group of patients, such as increase in progesterone through changes in nasal mucus, lead to colonization of virus in the mucosa of the respiratory tract and reduce its elimination. Increased levels of inflammatory cytokines such as , IFN- γ , IL-6, TNF- α and inflammatory chemokines known as cytokine storms, increase fetal abnormalities including neurodevelopmental disorders.

Conclusion: Pregnant women are at risk, although pregnancy has not been proven to be a risk factor for Covid 19, but Covid 19 is dangerous for both mother and baby and should be prevented by observing hygienic standards.

Keywords: Coronavirus; Pregnancy; Cytokine storm; Covid 19

High-throughput and Exactness CRISPR-Based Technologies in Diagnostics COVID-19

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Background and Aim: With the advent of COVID-19, Accurate and rapid recognition of SARS-COV-2 virus is a major challenge in the epidemic. Among the diagnostic tools, the CRISPR-Cas system has a special place, and the use of CRISPR-based technology is gaining expansive significance for the construct of diagnostic kits where it works by recognizing a particular genetic sequence followed by cutting the reporter molecule added to the reaction mixture.

Methods: This is an overview study. The keywords “CRISPR/Cas”, “COVID-19”, and “Detect” were used for the search in PubMed, Scopus, Google Scholar, and Web of Science databases for studies published.

Results: Detection of COVID-19 by CRISPR is much more efficient than RT-PCR. Detection of COVID-19 by RT-PCR takes 4-8 hours, but the CRISPR-based technologies detect it within a few minutes. In addition, in the Crisper technique, RNAs are designed according to the target sequence. Therefore, it can specifically identify a specific gene of the SARS-COV-2 virus. CRISPR-based virus diagnosis detection is a method that can test multiple specimens in parallel or examine multiple viral targets. For this reason, CRISPR-based technologies are very accurate, specific, and time-consuming in detecting COVID-19, even in the differential diagnosis of SARS-COV-2 virus from other viruses such as influenza A, influenza B, and respiratory syncytial viruses are involved, differential diagnosis of these viruses for treatment with the onset of the cold season and the presence of these viruses is a major concern for physicians. We examined the details of CRISPR-based technologies for the detection of COVID-19, Initial assessments had reinforcement primers designed against conserved viral regions and Cas13 crRNAs or Cas12 that would specifically bind a particular COVID-19 (Table 1) .

Conclusion: Given the potential of the Crisper system for rapid and specific detection of viruses and even different variants of SARS-COV-2 virus, the use of this technology can be a great help in controlling COVID-19 disease.

Keywords: SARS-COV-2; RT-PCR; Detection; CRISPR-Cas

How Correlation Between Parameters is Changed During COVID-19?

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Background and Aim: There are numerous studies which show that some parameters are important in COVID-19 but there is not any study to show the correlation of these parameters. In this study 7 lab parameters including WBC, PLT, RBC, NEU, LYM, LDH and CRP were statistically investigated in order to show their importance in diagnosing of COVID-19 and discover if any new relationship between these parameters is exists.

Methods: Lab parameters of 111 patient with severe COVID-19 and 120 healthy people were used. The significance of each biomarker was tested using ANOVA followed by Duncan test and was observed a significant difference between levels of NEU, LYM, LDH and CRP among two groups. Then in order to evaluate predictive ability of each parameter ROC curve and AUC analysis was applied on among two groups. In the next step the correlation between parameters were studied.

Results: NEU, LDH, CRP and NEU/LYM ratio are good predictors in prediction of RT-PCR results. The correlation analysis show that as a result of disease, a significant correlation is appeared between WBC with NEU, LYM and LDH and between CRP and LDH parameters.

Conclusion: Risk factors in severity of covid-19 are WBC, CRP, LDH, LYM and NEU. The correlation of these parameters is changed significantly during infection in severe cases.

Keywords: Blood biomarker; COVID-19; Correlation analysis; severity

In Silico Chimeric Synthetic Peptides Containing Immunodominant Epitopes from the Envelope gp46 Glycoprotein and Tax Protein of Human T-Lymphotropic Virus Type 1 (HTLV-I) in Fuse to IgG2aFC Fragment as a Candid Vaccine

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Background and Aim: Human T-lymphotropic virus type 1 is a human oncogenic retrovirus that causes diseases, such as Adult T-cell leukemia-lymphoma and myelopathy/tropical spastic paraparesis, in less than 5% of infected individuals. The envelope protein of the virus is essential for its binding and entering the host cell. Tax phosphoprotein plays a pivotal role in the continuous proliferation of cells infected with HTLV-1. Cellular immunity plays a key role in destructing HTLV-1-infected cells. The immunodominant regions of Tax and envelope antigens, with the ability to activate cellular immunity, are important vaccine candidate proteins. To increase the uptake, processing, and delivery of antigens by APCs, cytotoxic T-cells, and to induce strong cellular immunity, effective leukocyte surface receptors such as FcIgG are required, which increases the efficiency of CTL cells and controls viral load or destructs the virus. Due to the absence of any efficient Therapy for these diseases, introducing an effective vaccine against the virus is an emergent requirement.

Methods: The DNA sequences of glycoprotein 46 envelope (SP4 and SP2), Tax (11-19 and 178-186) as immunodominant determinants fused to the carboxy-terminal of the CH2-hinge and CH3 moieties of the mouse Fcγ2a Then were refined and validated through using 3D protein structure prediction software as the programs Phyre2, Ramachandran plot respectively.

Results: The recombinant fusion proteins were designed by using 3D protein structure prediction software. Three templates were designed and modeled in silico.

Conclusion: By binding the FC fragment of the mouse IgG2a protein to the end of the chimeric peptide, the efficiency of the vaccine candidate protein is dramatically increased more than 500 times which predicts high levels of INF-γ and IL-12 in immunity simulations. This Fcγ2α binding chimeric peptide can be used in the prevention, laboratory diagnosis, and treatment of HTLV-I.

Keywords: HTLV-1; FcIgG2a; Tax; Envelop

In-Silico Analysis on Potential Anti-SARS-CoV-2 Protease Agents by Structure-Based Docking and Cheminformatics

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Background and Aim: SARS-CoV-2 (COVID-19) outbreak encountered investigators to a complicated problem considering its control and treatment. There are many proposed drugs without definite effects used routinely in clinical settings by physicians. The aim is to evaluate four suggested chemicals using virtual analysis tools based on the drug-screening approach and application of cheminformatics, pharmacotoxicology, and docking software and servers.

Methods: Based on Ekins et al., 2020 published work that emphasizing four potential chemicals, rizatriptan, dasabuvir, pravastatin, and empagliflozin, after screening of millions of drugs, we done this extra work for a definitive virtual analysis. These drugs are food and drug administration (FDA) approved therapeutic agents, routinely prescribed by clinicians. We obtain the 3D structure of COVID-19 Main Protease (M Pro) from protein data bank (PDB) with PDB code: 6LU7, as the target of binding site screening. Besides, cheminformatics, pharmacotoxicology, and human proteins targets for each drug were evaluated using SwissADME interface, SwissTargetPrediction web server (<http://www.swisstargetprediction.ch/>), toxicity estimation software tool (T.E.S.T) and Toxtree-v3.1.0.1851 offline software. Ultimately, comparison and integration of results showed the best potential chemical.

Results: The best docking score (DOS) for dasabuvir was -139.399, followed by rizatriptan with DOS= -125.707, empagliflozin with DOS= -102.183 and pravastatin with DOS= -99.6642. In addition, quantitative structure-activity relationship (QSAR) and pharmacotoxicology evaluations showed the drug dasabuvir has acceptable criteria than the other three chemicals, in estimations. Human protein target-exploration showed that rizatriptan interacts with G protein-coupled receptor and kinase enzymes, Pravastatin targets the 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase. Empagliflozin interacts with sodium/glucose cotransporters. But, dasabuvir targets human protein with too low scores.

Conclusion: Virtual screening applied on four potential anti-COVID-19 showed the drug dasabuvir may be a safer and efficient agent, regarding pharmacotoxicology, and therapeutic purposes. However, virtually screened agent/s should be evaluated by experimental models for ultimate confirmation.

Keywords: 3C-like proteinase; Coronavirus; Cheminformatics; Molecular Docking Simulation; Quantitative Structure-Activity Relationship

Infertility in Patients with SARS-CoV-2 Infection, Review Study

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Background and Aim: Coronaviruses are a large family of viruses and subspecies of coronaviruses that occur naturally in mammals and birds, yet seven human-transmitted coronaviruses have been discovered. The latest type, Coronavirus Syndrome-Acute Respiratory Syndrome (SARS-CoV-2), broke out in December 2019 in Wuhan, China. The corona virus soon infected the whole world. There are various clinical signs associated with this virus, one of the strangest of which is infertility. The aim of this study was to evaluate the presence of infertility in patients with SARS-CoV-2 infection.

Methods: All studies that worked on the presence of infertility in patients with SARS-CoV-2 infection were searched from databases such as PubMed, Science Direct, Web of Science, Scopus and Google Scholar Magiran, scientific databases, Iran Medex and Iran Doc . Abstracts and results were evaluated and the results were extracted and analyzed.

Results: In this review study, we extracted 14 articles and abstracts and the results showed that the rate of infertility with a significant difference is higher in men than women and it has not yet been conclusively determined that corona is the cause of infertility, but what has been determined The fact is that the corona virus reduces the number of sperm in men.

Conclusion: The results of this study have shown that coronavirus is suspected of causing infertility in women and additional studies are needed to investigate the number of more people and the mechanism of infertility.

Keywords: Coronavirus; infertility; sperm; review study

Inhibition of H1N1 Influenza Virus Infection by Zinc Oxide Nanoparticles: Another Emerging Application of Nanomedicine

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Background and Aim: Currently available anti-influenza drugs are often associated with limitations such as toxicity and the appearance of drug-resistant strains. Therefore, there is a pressing need for the development of novel, safe and more efficient antiviral agents. In this study, we evaluated the antiviral activity of zinc oxide nanoparticles (ZnO-NPs) and PEGylated zinc oxide nanoparticles against H1N1 influenza virus.

Methods: The nanoparticles were characterized using the inductively coupled plasma mass spectrometry, x-ray diffraction analysis, and electron microscopy. MTT assay was applied to assess the cytotoxicity of the nanoparticles, and anti-influenza activity was determined by TCID₅₀ and quantitative Real-Time PCR assays. To study the inhibitory impact of nanoparticles on the expression of viral antigens, an indirect immunofluorescence assay was also performed.

Results: Post-exposure of influenza virus with PEGylated ZnO-NPs and bare ZnO-NPs at the highest non-toxic concentrations could be led to 2.8 and 1.2 log₁₀ TCID₅₀ reduction in virus titer when compared to the virus control, respectively ($P < 0.0001$). At the highest non-toxic concentrations, the PEGylated and unPEGylated ZnO-NPs led to inhibition rates of 94.6% and 52.2%, respectively, which were calculated based on the viral loads. There was a substantial decrease in fluorescence emission intensity in viral-infected cell treated with PEGylated ZnO-NPs compared to the positive control.

Conclusion: Taken together, our study indicated that PEGylated ZnO-NPs could be a novel, effective, and promising antiviral agent against H1N1 influenza virus infection, and future studies can be designed to explore the exact antiviral mechanism of these nanoparticles.

Keywords: Antiviral activity; Zinc oxide nanoparticle; H1N1 influenza; Polyethylene glycol

Investigation of CTNNB1 Gene Mutations and Expression in Hepatocellular Carcinoma and Cirrhosis in Association with Hepatitis B Virus Infection

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Background and Aim: Hepatitis B virus (HBV), along with Hepatitis C virus chronic infection, represents a major risk factor for hepatocellular carcinoma (HCC) development. However, molecular mechanisms involved in the development of HCC are not yet completely understood. Recent studies have indicated that mutations in CTNNB1 gene encoding for β -catenin protein lead to aberrant activation of the Wnt/ β -catenin pathway. The mutations in turn activate several downstream genes, including c-Myc, promoting the neoplastic process. So, we aimed to evaluate the pattern of mutations in the CTNNB1 gene in association to liver cirrhosis and HCC among an Iranian population.

Methods: The present study evaluated the mutational profile of the CTNNB1 gene and expression levels of CTNNB1 and c-Myc genes in HBV-related HCC, as well as in cirrhotic and control tissues. Mutational analysis of the β -catenin gene and HBV genotyping were conducted by PCR and subsequent direct sequencing. The expression level of β -catenin and c-Myc genes was assessed using real-time PCR.

Results: Among the HCC cases, 18.1% showed missense point mutation in exon 3 of CTNNB1, more frequently in codons 32, 33, 38 and 45. The frequency of mutation in the hotspots of exon 3 was significantly higher in non-viral HCCs (29.4%) rather than HBV-related cases (12.7%, $P=0.021$). The expression of β -catenin and c-Myc genes was found upregulated in cirrhotic tissues in association with HBV infection. Mutations at both phosphorylation and neighboring sites were associated with increased activity of the Wnt pathway.

Conclusion: The results demonstrated that mutated β -catenin caused activation of the Wnt pathway, but the rate of CTNNB1 gene mutations was not related to HBV infection. HBV factors may deregulate the Wnt pathway by causing epigenetic alterations in the HBV-related HCC.

Keywords: HBV; HCC; β -catenin; CTNNB1; mutation

Molecular and Serological Prevalence of HCMV in Iranian Patients with Breast Cancer

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Background and Aim: Human cytomegalovirus (HCMV) is prevalent viral infection involved in several human cancers including breast cancer. The presence of HCMV genome in breast cancer tissue and footprint of viral last exposure patient's serum are considered as important factor in the process of breast cancer development. This study aimed to investigate molecular and serological epidemiology of HCMV in patients with breast cancer in Iran for first time.

Methods: In our case-control study, 98 samples of breast tissue, including 49 cancerous (case) and 49 adjacent non-cancerous tissue were collected (control). In addition, we collected sera samples from all patients (n=49) and healthy individual (n=49). Seroprevalence of HCMV was assessed by Enzyme-linked immunosorbent assay (ELISA) and detection of HCMV genome was performed using Nested-PCR method.

Results: HCMV genome found in 16.3% (8/49) of cases tissue and 2% (1/49) of controls tissue. In patients group, the levels of anti-CMV IgG and IgM were 93.9% and 2% compared to 69.4% and 4.1% in healthy individuals, respectively. There was a statistically difference between the anti-CMV IgG in patients and healthy control ($p=0.002$). We found 75% of (6/8) HCMV genome positive PCR samples were also positive for their anti-CMV IgG in cases which was statistically significant ($p=0.01$).

Conclusion: Our result showed significant presence of HCMV genome and anti-CMV IgG in patients, supporting the role of HCMV in breast cancer.

Keywords: Breast Neoplasms; Cytomegalovirus; Iran; Prevalence

Molecular Detection and Genotyping of Human Cytomegalovirus in Kidney Transplant Recipients under Ganciclovir Prophylaxis in Iran

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Background and Aim: Human Cytomegalovirus (HCMV) infection is one of the most common complications in renal transplant recipients. Despite effective treatments strategies for the HCMV infection in renal transplant recipients, this infection is still one of the causes of renal transplant rejection. Objective: To evaluate the frequency of the glycoprotein B genotypes of HCMV among renal transplant recipients.

Methods: A total of 246 renal transplant recipients contributed to this cross-sectional study. Viral DNA was extracted from these plasma samples and the presence of HCMV genome was assessed by Semi-nested PCR with specific primers for HCMV B glycoprotein gene. Sanger sequencing analyses were performed to determine HCMV genotypes and Mega x software was utilized for HCMV nucleotide alignment and construction of a phylogenetic tree.

Results: Human cytomegalovirus DNA was detected in 11 (4.47%) recipients. According to the phylogenetic analysis, HCMV gB3 was 50% among renal transplant recipients, followed by gB4 30% and gB1 20% however, the gB2 genotype was not detected.

Conclusion: This study showed that the HCMV infection in our patients is relatively low since all transplant recipients received appropriate prophylaxis, thereby antiviral prophylaxis should be considered for all patients at risk of HCMV infection after renal transplantation. Also, gB3 was the most predominant genotype among our renal transplant recipients that was associated with a higher incidence of severe HCMV infections. In addition, an increase in serum creatinine level was detected in patients at the time of detection of HCMV infection. No association was found between age and gender and HCMV infection.

Keywords: Human Cytomegalovirus; renal transplantation; Genotyping; glycoprotein B

Molecular Epidemiology of Human Respiratory Syncytial Virus (HRSV) in Iranian Military Trainees with Acute Respiratory Symptoms in 2017

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Background and Aim: Respiratory syncytial virus (RSV) is the leading cause of lower respiratory tract infection in many populations, including military recruits receiving basic training. Therefore, this study was set out to determine the molecular epidemiology, genotype and phylogenetic features of RSVs in patients with respiratory infection as a case study.

Methods: In this study, military barracks of Tehran, Iran, between January to March 2017 exposed to respiratory diseases were used for sampling. Throat swabs were taken, a reverse transcriptase-polymerase chain reaction (RTPCR) assay was performed to identify RSV and then the genotyping and phylogenetic analyses of RSVs in patients with a respiratory infection.

Results: using RT-PCR. Sequencing showed the incidence of type A (2.5%, n=10) to be much higher than type B (0.25%, n=1); Sore throat was the most common symptom among RSV patients. Phylogenetic analysis revealed that the majority of strains from the studied samples were more consistent with those from the Philippines and the US strains.

Conclusion: This study is the first to document RSV as a major cause of acute respiratory illness among military trainees in Iran. The prevalence of RSV is substantial in the cold season and the prevalence of genotype A is dominant in the country, leading to take essential steps in preparing a preventive vaccine against this viral infection.

Keywords: Respiratory tract infection; Human respiratory syncytial virus; Military trainees; Reverse transcription polymerase chain reaction; Genotyping

Molecular Mechanisms of Merkel Cell Polyomavirus

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Background and Aim: Merkel cell polyomavirus (MCPyV) is a double-stranded DNA virus, helps to develop most Merkel Cell Carcinoma (MCC). MCC is an invasive skin cancer that is related to aging and immunosuppression. Most MCV viruses are detected in MCC tumors. About 80% of MCC tumors become infected with MCV, and the virus integrates with a monoclonal pattern into the host genome. MCC is caused by the integration of Merkel cell polyomavirus and the expression of viral T antigens or sunlight exposure. The MCPyV genome is divided into three main regions: 1) the non-coding regulatory region 2) the early region and 3) the late region.

Results: The early region contains large T (LT) antigen, small T (sT) antigen, the 57 kT antigen, and a protein called alternative LT open reading frame (ALTO). The late region contains the capsid proteins, VP1 and VP2. Continuous expression of MCPyV T antigens from the integrated viral genome is necessary for the survival of MCC tumor cells.

Conclusion: These mechanisms support MCPyV in the development of MCC. In the present review, the authors attempt to review the molecular mechanisms involved in the development of MCC.

Keywords: Merkel cell polyomavirus; Merkel cell carcinoma; monoclonal pattern; T antigen

Multicenters Study of Hepatic and Cardiac Parameters in Covid-19 Patients from Three Ethnic Groups of Iran

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Background and Aim: The liver and cardiovascular system disorders are not common in Covid-19 patients, but it has been illustrated that the patients suffering from these complications are exposed to a higher rate of mortality and disease progression. The main reason for targeting these organs is the expression of SARS-CoV-2 receptors on the gastrointestinal tract and cardiovascular systems. Hepatic injuries can drive to increased levels of liver enzymes, including ALT, AST, and LDH. Abundant levels of AST, LDH, and CPK can be indicators of cardiac injuries.

Methods: The current study is comparing 366 individuals who are divided into two groups; the case group (Covid-19 patients) and the control group (healthy individuals) in which we examined hepatic and cardiac function parameters. Moreover, the clinical characteristics of the participants, races and their correlations with studied parameters were assessed. The Participants were from varied ethnicities, including Lor, Fars, and Turkoman races.

Results: The correlations between hepatic and cardiac function parameters and ethnicity indicate that Fars individuals are more susceptible to the disease progression, liver and heart damages. The increased mean of the ALT level in the case group (102.36) compared with the control group (68.32) resulted in no significant correlation. Other parameters, including CPK, LDH, and AST, showed an increase in the control group values when compared to the case group; however, the differences were not significant.

Conclusion: Considering the involvement of cardiac and hepatic organs by SARS-CoV-2, paying special attention to the disorders of these organs through assessing the hepatic and cardiac function parameters can lead to the enhancement of the patient's recovery and survivals. Application of tests, drugs and treatment procedures during the infection may lead to the minor hepatic complications and constant monitoring of hepatic and cardiac function parameters and clinical characteristics can pave the way for more sufficient treatment of SARS-CoV-2 infection.

Keywords: Covid-19; SARS-CoV-2; Hepatic; Cardiac

Nanomaterial Based Diagnosis for Detection of COVID-19

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Background and Aim: Early diagnosis of the disease is one of the most important steps. In medicine, laboratory diagnosis is an important process. Diagnosis is a basic need that can provide information to the physician for proper disease management. Medical diagnosis focuses on a good and accurate diagnostic test. The Covid-19 pandemic has been threatening the world for nearly two years, and today nanoparticles can speed up medical diagnosis.

Methods: We searched Google Scholar, Pubmed and Scopus found 35 articles. 12 articles were selected with specific keywords. Among the Covid-19 nano diagnostic methods, the following can be mentioned: Graphene-based field-effect transistor biosensing devices, COVID-19 rapid test cassette, Chiral zirconium Quantum dots, CRISPR-based detector lateral flow assay.

Results: Nanotechnology can be used to create a new nano diagnostic system for COVID-19. Among the positive features of nanodiagnostics can be the reduction of false-negative results, increase the efficiency of diagnostic testing, detection even in mild or asymptomatic cases, accurate identification of a target Specific in multiple genes, detection at low sample concentrations, and rapid detection at low cost.

Conclusion: Using nano diagnosis-based can increase the speed, availability, sensitivity, and specificity of the diagnostic methods especially in a low-income country.

Keywords: Nano diagnostics; COVID-19; Nanomaterials; Nanotechnology

NGS in the COVID-19 Pandemic, from Alpha to Omicron

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Background and Aim: It has been about two years since the last days of December 2019, when COVID-19 first appeared in Hubei, China, and the SARS-CoV-2 virus is still spreading around the world after many mutations. Next-Generation Sequencing (NGS) technology has dramatically reduced sequencing time and costs in the past few years, making it possible to sequence billions of nucleic acid fragments simultaneously. In this article, we discuss the significant role of this technology in combating the COVID-19 pandemic.

Methods: Through a review of scientific databases such as PubMed, Web of Science, Google Scholar, and Scopus, we will have extracted a number of articles by the year 2021. We used the keywords Next-Generation Sequencing, NGS, COVID-19, Variants and diagnostic assay as our main search terms.

Results: NGS is a simple and accurate way of identifying new strains of coronaviruses and other pathogens without prior knowledge of the organism. Increasing concerns about the rapid spread of new types of SARS-CoV-2, such as B.1.1.7 [Alpha], B.1.617.2 [Delta], and B.1.1.529 [Omicron], require Sequencing to highlight the rapid detection of mutations further and prevents the spread of new strains.

Conclusion: SARS-CoV-2 was detected through sequencing early on in the outbreak. Public health officials, drug developers, researchers, and scientists rely on NGS to provide evidence crucial to their work and track global transmission routes of viruses. This technology makes it possible to detect SARS-CoV-2 virus more accurately and sensitively, and identify mutations faster to prevent the spread of new strains. In addition, NGS can be used to identify potential therapeutic targets for COVID-19, respiratory infections, and antimicrobial resistance alleles.

Keywords: Next-Generation Sequencing; NGS; COVID-19; Variants and diagnostic assay

Occult Hepatitis C Virus Infection among Hemodialysis Patients: An Iranian Experience

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Background and Aim: Patients with chronic kidney failure and those under chronic hemodialysis (CHD) treatment are at high risk of infection with hepatitis C virus (HCV). Incidence of occult HCV infection (OCI) in CHD is controversial and the real burden of HCV in this population may be affected by the rate of OCI. Thus, this study evaluated the molecular assessment of OCI in CHD in an Iranian population.

Methods: All the subjects on CHD in the South Khorasan province of Iran were called for participation in the study. Whole blood sample was taken with serological, clinical, and demographic information being recorded. HCV-RNAs were checked in serum and peripheral blood mononuclear cells (PBMC) using an in-house semi-nested PCR assay. Viral load was determined by a real-time PCR based quantification kit. Sequencing was performed to determine genotypes.

Results: Overall, 120 cases participated in the study; 57.5% were male and the rest were female. In sera samples, no positive case for HCV-RNA was found. In the PBMC samples, 2/120 (1.6%) were positive for HCV-RNA (95% CI, 0.002 to 0.059)); the mean age among OCI positives was 37.5 ± 19.2 which was significantly lower than OCI negatives ($p=0.026$). Only one case had detectable viral load which was 49 IU/mL. The only HCV genotype identified was 1a.

Conclusion: This study showed that there is the risk of OCI among CHD patients, and very low and not-detectable viral loads of OCI warrant more follow up molecular testing for earlier diagnosis and treatment in the era of DAA.

Keywords: HCV; OCI; occult; genotype; dialysis; hemodialysis

Polymorphism Analysis of Interleukin-37 Genes at Position rs4241122 in Hepatitis B Infections with Different Outcomes

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Background and Aim: Regarding the important role of cytokines in influencing the outcomes of hepatitis B virus (HBV) infections, in this study we aimed to investigate the association between polymorphisms of interleukin-37 (IL-37) and the outcomes of HBV infection.

Methods: In this study, we enrolled 300 subjects with chronic HBV infection, including those with cirrhosis/hepatocellular carcinoma (C/HCC), chronic active hepatitis B (CAH) infection, or asymptomatic carriers (AC), and 58 individuals whose infection was spontaneously cleared (SC). The recency of HBV infection was also checked through ELISA assays for HBsAg and HBcAb. Genomic DNA was extracted, and IL-37 genotyping was performed using ARMS-PCR.

Results: The mean age of the participants was 47.61 ± 13.41 years. Out of 358 subjects, 244 were male and 114 were female. The results of ELISA assay showed that all 58 sera samples of the SC group were negative for HBsAg and positive for HBcAb, confirming the spontaneous clearance of HBV infection. The frequency of genotypes and alleles of IL-37 at position rs4241122 were not statistically different among the four studied groups ($P > 0.05$).

Conclusion: According to the results, variation of IL-37 at position rs4241122 does not appear to influence the outcome of HBV infection.

Keywords: HBV; Polymorphism; IL-37

Possible Role of Reovirus in the Celiac Disease

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Background and Aim: Celiac disease (CD) is a recurrent, lifelong autoimmune disease that targets the proximal small intestine with numerous additional systemic intestinal manifestations in genetically predisposed individuals. CD is an important health problem and a significant number of people might suffer from the disease. It has been suggested that viruses play a probable role in the development of CD in humans.

Results: Viral infections change immune tolerance, resulting in diseases like CD. Viral infections have been suggested to elicit pathological processes leading to the initiation of T helper (TH1) immunity celiac disease. Viruses like Adenovirus, Enterovirus, Rotavirus, and Reovirus were previously linked to CD.

Conclusion: Recent surveys also reported that Reoviruses may contribute as an environmental modifier for CD development. The T1L reovirus strain-induced broader transcriptional changes in the mesenteric lymph nodes, where immune responses to dietary antigens are induced. The probable molecular mechanisms of virus-host interactions and possible pathogenesis pathways will be reviewed through the present abstract.

Keywords: Celiac Disease; Reovirus; Autoimmune reactions; Viral infection; Pathogenesis trigger

Prevalence of Human Cytomegalovirus in Breast Cancer: A Systematic Review

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Background and Aim: Breast cancer is the most common malignancy among women around the world, with several risk factors contributing to its progression. The role of infectious agents such as viruses in the progression of this cancer is relatively well-known, as a result of various studies in different geographical regions that reported the prevalence of cytomegalovirus in breast cancer. In this systematic review, we tried to examine the prevalence of cytomegalovirus in breast cancer

Methods: We conducted a comprehensive search of three databases: PubMed, Scopus, and Google Scholar up to January 21, 2021.

Results: Out of 584 articles, 29 articles including 3158 samples were finally reviewed, of which 16 articles used PCR methods, 8 articles used serology method and 13 articles used other methods such as immunohistochemistry and in situ hybridization. The prevalence of the virus was higher in Asian and African countries such as Iraq and Egypt than other European and South American countries. Among these, the highest prevalence of the virus in different samples of patients with breast cancer was related to Iraq.

Conclusion: The results of our study showed the higher prevalence of the virus in patients with breast cancer rather than normal subjects and reveal the role of the virus in the development of breast cancer.

Keywords: Breast Neoplasms; Cytomegalovirus; Prevalence; Systematic review

Prevalence of WU and KI Polyomaviruses in Negative SARS-CoV-2 Patients with Respiratory Symptoms in Hamadan, Iran

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Background and Aim: Respiratory infections are the most common disease worldwide. Findings have shown that viral infections are the most important cause of respiratory infections. WU and KI polyomaviruses (WUPyV and KIPyV) have potential pathogenic role in respiratory infection. Global prevalence of WUPyV and KIPyV are between 1-16.4 and 0.45-12.14% respectively. The aim of this study is prevalence of WUPyV and KIPyV among SARS-CoV-2 suspected patients with negative PCR tests.

Methods: In this study 602 nasopharyngeal swab samples were collected. Viral nucleic acid was extracted by High Pure Viral Nucleic Acid Kit (Roche, Germany) and real-time PCR has been done to SARS-CoV-2. Negative samples were collected and PCR reaction has been done by home made primer to WUPyV and KIPyV. In this study DNA fragment of the virus was synthesized and cloned in the PUC57 vector as a positive control. Analytical sensitivity was determined by conversion of ng of plasmid DNA to copy number.

Results: Descriptive statistical analysis has been done by Graphpad prism V.8. In this study the men and women were 53% and 47% respectively. The children Under 5 years old were 3.8% of the total population. Elderly patients with more than 60 years were 35% of the population. Analytical sensitivity of the PCR reaction was 80 copy per reaction. WUPyV and KIPyV were not detected in these samples.

Conclusion: Prevalence of WUPyV and KIPyV are very different in the human population. The viruses have not already been detected in Hamadan province. The aim of study was determination of WUPyV and KIPyV prevalence in SARS-CoV-2 negative patients. It seems that the other viral respiratory infection may be considered.

Keywords: WU and KI polyomaviruses; SARS-CoV-2; Respiratory infections; polymerase chain reaction

Relation Between the rs10204525 and rs2227981 Polymorphisms of PDCD1 Gene with Human T-Cell Leukemia Virus Type 1 (HTLV-1) and Proviral Load

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Background and Aim: Human T-cell leukemia virus type 1 (HTLV-1) which cause two serious diseases in human and proviral load (PVL) of virus can predict the progression of disease. Programmed cell death-1 (PD-1) is a regulatory protein that created exhaustion phenotype in T cells and influence in immune system response during viral infection. In this research, association between rs10204525 and rs2227981 polymorphisms, in exon 5 of PD-1 gene, with susceptibility to HTLV-1 infection and PVL in Iran's population studied.

Methods: In this case-control study, PD-1 rs10204525 and rs2227981 polymorphisms were evaluated in 162 healthy individuals and 81 HTLV-1 asymptomatic carriers (ACs). These polymorphisms were genotyped by Polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) and PVL was detected by quantitative real-time PCR (Q-RT-PCR).

Results: Frequency of GA and AA genotypes in rs10204525 polymorphism were higher in ACs group (82.7%) than control group (26.5%) significantly; and GA + AA genotypes were significantly associated with HTLV-1 infection (OR = 13.244, 95%CI = 6.755–25.968, p = 0.000); but CT + TT genotypes in rs2227981 polymorphism, were as a protective factor against HTLV-1 infection (OR = 0.473, 95%CI = 0.279–0.813, P = 0.009). However, there was no significant difference between these polymorphisms and HTLV-1 PVL.

Conclusion: The results indicated that there is an association between rs10204525 and rs2227981 polymorphisms with susceptibility to HTLV-1 infection in Iran's population.

Keywords: Gene polymorphism; HTLV-1; Programmed cell death-1; Viral load

Relation Between the rs4143815 Polymorphism in PDL1 Gene with HTLV-1 Proviral Load among Iranian Blood Donors

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Background and Aim: PDL1, ligand of immune checkpoint PD1, regulated the immune response and effect on chronic viral infection. Genetic differences including polymorphisms and mutations can play a vital role in Human T-cell leukemia virus type 1(HTLV-1) infection and proviral load (PVL). PVL is significantly higher in symptomatic patients compared to asymptomatic carriers (ACs); thus PVL would be a prognostic marker of HTLV-1 disease progression.

Methods: So in this case-control study that contained 81 ACs and 162 healthy controls (HCs), the rs4143815 polymorphism of PDL1 gene, a site for binding to miR-570, was evaluated with allele-specific polymerase chain reaction (AS-PCR) and PVL level measured by quantitative real-time PCR (q PCR). Association of rs4143815 polymorphism with HTLV-1 and PVL were analyzed by SPSS-22 statistical software based on chi-square test and T-test.

Results: The results showed that ACs had higher frequency of CC genotype ($p=0.031$) and C allele ($p=0.027$) compared to HCs also, PVL was significantly higher among ACs with C allele ($p=0.020$).

Conclusion: Our results suggest that the rs4143815 polymorphism can play important role in HTLV-1 infection and PVL.

Keywords: Gene polymorphism; Human T-lymphotropic virus 1; PD-L1; Viral load

SARS-CoV-2 Antibody Response with Sinopharm Vaccine in Patients with Cancer: A Case-Control Study

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Background and Aim: Recent studies indicate that serum neutralizing antibody levels are highly predictive for protection against COVID-19 infection (15-18). So in this study we aimed to assess safety and efficacy of BBIBP-CorV (Sinopharm) in groups of cancer patients by using clinical complications of vaccinated individuals after each dose of vaccination and assessment of serum levels of neutralizing antibodies [SARS-CoV anti-spike recombinant receptor binding domain (anti-sRBD), and anti-nucleocapsid (anti-N) IgG].

Methods: In this case-control study, participants were recruited among the patients referring to Firoozgar Hospital, within the period of Apr to Sep 2021 in Tehran, Iran. All the patients were registered to receive BBIBP-CorV (Sinopharm) vaccine against COVID-19. Two groups of patients with a history of cancer considered as case group and without a history of cancer considered as control group.

Results: 152 individuals (67.1% females) with mean age of 46.71 ± 15.36 years old and mean Body Mass Index (BMI) of 24.96 ± 5.46 kg/m², entered into this study. All the participants received the first dose of BBIBP-CorV (Sinopharm) vaccine and 63 (41.4%) patients reported past history of COVID-19 infection. The participants were divided into two groups. 107 individuals with a history of cancer were selected as cases and 45 without a history of cancer were picked up as controls.

Conclusion: Antibody response were significantly lower among cancerous patients compared to the controls after full vaccination with BBIBP-CorV (Sinopharm), Males, those with BMI ≥ 25 and without past history of COVID-19 infection and hematologic cancers were more possible to have no antibody response.

Keywords: COVID-19; Sinopharm; Antibody response; Cancer

Selective APC Targeting for Making a Novel Multi-Immunodominant Fc-Fusion 2 Recombinant Vaccine Against HTLV-1

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Background and Aim: HTLV-1 is the causative agent for two life-threatening diseases: adult T cell leukemia/lymphoma (ATLL) and HTLV-1-associated myelopathy (HAM/TSP). Due to the lack of any effective treatment, introducing an effective vaccine is an urgent need.

Methods: The DNA sequences of the 11-19 and 178-186 amino acids of HTLV-1-Tax and SP2 and P21 were used as immunodominant determinants fused to the carboxy-terminal of Fcγ2a or His-tag. These constructs were then produced in the Pichia pastorisGS115 strain. The immunogenicity and the protective properties of these recombinant fusion proteins were assessed in the HTLV-1 infected-mouse model.

Results: The immunogenicity assessments showed a significant increase in IFN-γ and IL-12 by tTax-tEnv-mFcγ2a (1537.2±292.83pg/ml) compared to tTax-tEnv-His (120.28±23.9). The IFNγ and IL-12 levels in the Fc group were negatively correlated with pro-viral load (PVL) (R=-0.82, p=0.05), respectively. Meanwhile, IL-4 increased by tTax-tEnv-His (21.16±1.76 pg/ml) compared to tTax-tEnv-mFcγ2a (13.7±1.49) (P=0.019), with a significant negative correlation with PVL (R=-0.95, P=0.001).

Conclusion: Both Fcγ2a and His tag constructs had the same protective effect against HTLV-1 infections in the challenging experiments. However, the Fc fusion construct has been more effective in inducing Th1 responses. The comparison of Fc-tag and His-tag multi-immunodominant HTLV-1 vaccine candidates showed that both specific humoral and cellular immune responses are necessary for proper protection.

Keywords: HTLV-1; Vaccine; Fc fusion vaccine; recombinant protein; subunit vaccine

Seroprevalence of HSV1,2 in 15 to 35 Years Old Population of Mashhad City in Iran

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Background and Aim: Herpes simplex virus (HSV) has two types including HSV1 and HSV2. It is proposed that only 20 to 25 percent of the patients who have anti-HSV antibodies have a history of genital or orolabial herpes. Therefore, this study was performed to assess the seroprevalence of HSV1 and HSV2 in 15 to 35 years old population of Mashhad city in Iran.

Methods: This cross-sectional study was conducted on 916 cases composed of 15 to 35 years old population of Mashhad city in Iran. By using the Enzyme-linked immunosorbent (ELISA) method, the presence of HSV1 and HSV2 antibodies were assessed. The extracted data were analyzed using SPSS software version 20.

Results: Totally, 916 people were enrolled including 681 (74.3%) cases with positive test and 235 (25.7%) cases with negative test. However, there were no cases of positive IgM and all were positive IgG. The mean age of the positive cases in our population was significantly higher than negative cases (26.2 years vs. 23.3 years; $p < 0.05$).

Conclusion: Totally, our study showed a high seroprevalence of HSV infection. However, there were no cases of IgM positive antibody in this our 916 cases.

Keywords: HSV; IgG; IgM; Mashhad; Seroepidemiology

Survey on the Presence of rs179008 and rs179009 Polymorphisms of TLR7 Gene and Its Relationship with Disease Severity in Patients with Covid-19

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Background and Aim: With the pandemic of the new corona virus, several studies were begun on this virus. In this study, two polymorphisms of TLR7 gene rs179008 and rs179009 that play an effective role in resistance and susceptibility to viral infections such as HCV, HBV, HIV, CMV were investigated. TLR7 is capable of detecting ss_RNA in innate immunity. It is mainly expressed in Bcells, monocytes and macrophages and pDCs. After binding to the TLR ligand, large amounts of pDCs (IFN α) induce a strong antiviral response in adjacent cells. Both IFN α induction and adaptive immunity are mediated by pDCs, which are considered important for TLR7 agonists to fight viruses, so to optimize and personalize treatment and selecting patients who benefit most from TLR7 agonist therapy. It is important to note how genetic and environmental factors affect the response of pDCs to TLR7 agonists. TLR7 polymorphism (Gln11Leu), sex, and chronic virus all affect pDC IFN production. For example, Gln11Leu polymorphisms are directly associated with higher virus acquisition and loading, as well as faster disease progression in people with hepatitis C and HIV infection and basal cell carcinoma. TLR7 32A / T polymorphism in HIV-1 women was associated with disease progression compared to non-infected women.

Methods: test was performed on 160 CBC samples from people with Covid-19, which included 110 patients with mild disease and 50 patients with acute disease in the ICU ward. extraction was performed. Then, specific primers were designed using gene runner software. the main (outer) primer was designed to amplify the SNP locus, which was a 369-nucleotide product and as a control. Then, two specific primers were designed to check the presence of snp, the 3'OH of the primers was placed on the SNP. in normal conditions and in the absence of snp, the primer was fully connected and reacted with the outer primer. We will have a product and if the snp primer is not fully connected, we will not have a product. so in normal conditions rs179009 primer will react with the outer forward primer and give a 144 nucleotide product and also rs179008 will react with the outer reverse primer and give a product of 87 nucleotides and finally a 369 nucleotide product as a control. This method does not determine whether the sample is heterozygous or homozygous, just shows the presence or absence of snp, base on studies this mutations is commonly homozygous.

Results: In this study, out of 160 samples, 110 were in mild condition and 50 were in ICU. In Mild group, 102 patients (92.7%) had no mutation and 8 (7.3%) had rs179008 mutation. Similarly, in Severe group, 48 patients (96%) had no mutations and 2 patients (4%) had mutations. Also, rs179009 mutation was observed in only one of the samples. In total, the prevalence of rs179008 mutation was

6.25% in 160 patients and rs179009 was 0.65%. Therefore, a statistical relationship between severity and disease and this mutation was not observed in patients with Covid-19. Also, there is no significant relationship between rs179008 mutation and sex in this study (presence of mutation in women: 7.9, men: 4.8%) Similarly, a study by Sonja I. Buschow et al. Showed that the rs179008 allele is predominantly absent in Asians and is more common in Caucasians. According to the articles, the prevalence of rs179009 in Asia and East Asia is reported to be 5-15% and rs179008 is about 0-5%. Therefore, these two polymorphisms are not influential factors in the severity of covid-19 pathogenicity due to the effect on the function of innate immune cells, especially pDCs, in interferon production

Conclusion: Considering that these two polymorphisms played an important role in the resistance and susceptibility of individuals to viral infections, especially HCV, this study was performed which showed that due to the role of these two polymorphisms in innate immunity, especially in pDCs in production Interferon is not a logical relationship between the prevalence of these two polymorphisms and the severity and susceptibility of individuals to this disease. Also, a logical relationship between gender and disease severity was not observed in this study. But a study by Seillet et al et al. Concluded that pDC function is also influenced by gender: Male pDCs produce less IFN α by TLR7 than female pDCs. This difference is due to the difference in the expression of X chromosome genes in the TLR7 stream, along with hormonal effects. Due to the presence of some patients with underlying diseases such as hypertension, hyperlipidemia, asthma, diabetes due to the small number of cases in our study, the statistical relationship is only between people with diabetes and Asthma was observed. For better results, more studies are needed in the higher statistical community.

Keywords: covid-19; corona virus; TLR7; polymorphism; rs179008; rs 179009

The Effect of Medicinal Plants on the Treatment of Coronavirus 2019 (COVID-19)

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Background and Aim: Coronavirus disease 2019 (COVID-19) is a life-threatening disease. The lack of specific drug molecules or therapeutic strategies against this disease is more devastating than it. Also, modern medicine has recognized its success thanks to traditional medicine whose effectiveness Derived from Medicinal Plants This study focuses on medicinal plant extracts as promising approaches against COVID-19. The aim of this study was to evaluate the effect of medicinal plants on the treatment of COVID-19.

Methods: This study was performed by searching for keywords such as coronavirus, COVID-19, and medicinal plants invalid databases such as pub med and google scholar.

Results: Studies have shown that coronaviruses have sensitive single-stranded RNA fragments with average dimensions between 60 and 140 nm in diameter. The elderly and patients with other medical conditions (diabetes and heart disease) become severely ill. Symptoms of this disease mainly include fever, cough, sore throat, runny nose, and difficulty breathing. Factors of production, hospital transmission, proliferation in the lower respiratory tract, and viral immune system are risk factors and serious problems in the general health of this disease. Essential elements of eucalyptus oil on the main viral proteinase have been designed using the docking method and it is suggested that it be considered as the main inhibitor of coronavirus proteinase (SARS CoV-2) in the evaluation of polyphenols in green tea.

Conclusion: Inhibition of viral proteases prevents the proliferation of viruses. Therefore, detection of plant anti-proteases due to fewer side effects will play an effective role in preventing the proliferation and spread of the virus.

Keywords: Coronavirus; COVID-19; medicinal plants

The Relationship Between Age and SARS-CoV-2 Viral Load in SARS-CoV-2 Infected Patients

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Background and Aim: The coronavirus disease (COVID-19) pandemic is an ongoing global health crisis that has led to the death of millions of individuals with the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) being its cause. The risk and outcomes in many diseases are affected by age and it has been shown that COVID-19 is no exception. Moreover, an important predictor of disease severity and mortality in SARS-CoV-2 is viral load. Therefore, in this study, we aimed to evaluate the relation between SARS-CoV-2 viral load and age.

Methods: This cross-sectional study evaluated patients hospitalized in Ghaem Hospital, Mashhad, from August 2020 to July 2021 with clinical symptoms of Covid-19, a positive RT-PCR test, and radiology tests positive for Covid-19. RNA extraction was carried out using a commercial kit (ROJE technologies, Iran). RdRP and N genes of the virus as target genes, as well as human RNAase P gene as an internal control gene were evaluated by real-time PCR method.

Results: A total of 484 patients aging 18 to 99 with the mean age of 61.25 years were evaluated. 258 (53.3%) of our subjects were males and 226 (46.7%) were females. The Ct value ranged from 10.0 to 40.0. Ct values showed no statistically significant difference between male and female subjects ($P=0.127$). Furthermore, correlation analysis showed that there was no correlation between the subjects' age and Ct value. ($r=-0.071$, $n=484$, $p=0.117$).

Conclusion: No statistically significant difference was found in viral loads between age groups. It was demonstrated that males under 25 had a higher mean Ct value in comparison to females with no statistical significance. Our findings further improve our knowledge of possible preventive measures and therapy targets. It is highly recommended that future studies evaluate the correlation between viral load and patients' symptoms, as well as their age, in larger populations.

Keywords: COVID-19; age; real time PCR; Viral load; SARS-CoV2

The Role of Mesenchymal Stromal/Stem Cell Therapy in COVID-19 Patients: A Systematic Review

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Background and Aim: New coronavirus disease 2019 (COVID-19), is considered a pandemic disorder, which in turn has attracted the attention of researchers in different fields to explore reliable therapeutic strategies for the management of patients with COVID-19. Nowadays, regarding the rapid spread of COVID-19; and lack of confirmed treatment for this infectious disease, despite many efforts to develop therapeutic approaches for COVID-19. There is compelling evidence that demonstrated mesenchymal stromal/stem cell (MSC) therapy can be a good option for the management of COVID-19 patients. MSCs have multiple biological properties (including the potential of differentiation, high safety and effectiveness, secretion of trophic factors, and immunoregulatory features) that make them effective for the treatment of diverse diseases. However, some studies have questioned the positive role of MSC therapy in the treatment of COVID-19.

Methods: In the current systematic review study, we searched the Google Scholar, Web of Sciences, PubMed, and Scopus databases to understand the crucial role of mesenchymal stromal/stem cell therapy in COVID-19 patients between July 2010 and November 2021. After conducting the literature review, 137 appropriate investigations were included.

Results: Our findings showed regarding the features of MSCs to overcome viruses, immunomodulatory properties, and their potential for tissue regeneration, MSCs therapy has become a hot topic among researchers. Besides, MSC therapy has not demonstrated any harmful side effects on patients. Meanwhile, there are several challenges exist related to cell-based treatment. The safety, efficacy, and timing of MSC administration, suitable and effective dose, and route preparation for infusion source of MSC delivery are under investigation as well. Thus, MSC therapy could be a promising therapeutic approach to managing COVID-19 patients.

Conclusion: Because of the novelty of MSC- based therapy in the management of COVID-19 patients, there are controversial opinions regarding how this type of treatment could be effective. Further in vitro, in vivo, and clinical trial investigations are required to confirm and refute this claim.

Keywords: New coronavirus disease 2019; Mesenchymal stem cell; Cytokine storm; Immunoregulatory; Pneumonia

The SARS-CoV-2 (COVID-19) Pandemic in Hospital: Disinfectants; Efficiency

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Background and Aim: The current COVID-19 pandemic that is caused by SARS-CoV-2 has led all the people around the globe to implement preventive measures such as environmental cleaning using alcohol-based materials. The present study was conducted to determine the persistence of the virus to two common types of disinfectants and its operators in the hospital.

Methods: All the samples were collected from identical surfaces. Positive or negative results are reported after qPCR confirmation. In the present study, the efficiency of two disinfectants, namely alcohol-based hand rubs (ethanol 70%) and sodium hypochlorite (0.001 %) were examined with regard to inactivation of SARS-CoV-2 on inanimate surfaces of different wards of the hospital at both 5 and 15 min after disinfection.

Results: The SARS-CoV-2 could survive on inanimate surfaces including the nursing station, reception desk, cell phones, door handles, and floors for 5 min after using alcohol-based hand rubs (ethanol 70 %), while in the case of the reception desk, the presence of SARS-CoV-2 was positive after 15 min. With regard to sodium hypochlorite (0.001 %), SARS-CoV-2 could survive on inanimate surfaces including the nursing station, reception desk and the floors at both 5 and 15 min after disinfection, but not on cell phones and door handles.

Conclusion: The level of viral load of patients or infected people in the hospital was high and perhaps several times the amount of disinfectant should be used, especially on high-touch surfaces, to ensure that presence of SARS-CoV-2 on inanimate surfaces is negative. It can be hypothesized that SARS-CoV-2 can survive on different materials for different durations. Based on the obtained findings, it can be suggested that the interval between disinfection operations should be decreased and the disinfectants could be replaced with more efficient ones.

Keywords: COVID-19; SARS-CoV-2; Inanimate surfaces

Coronary Artery Disease in Patients with Coronavirus Disease 2019 (COVID-19): A Systematic Review and Meta-Analysis

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Background and Aim: Patients with underlying cardiovascular disorders such as coronary artery disease (CAD) are more prone to severe forms and multiple complications of COVID-19. The present study aimed to investigate CAD in patients with COVID-19 in the form of a systematic review and meta-analysis.

Methods: Main electronic databases including Medline (via PubMed), EMBASE, and Web of Science were carefully searched and reviewed for original research articles published between 2019 and 2021. A total of 109 studies worldwide that address CAD in patients with COVID-19 were selected and analyzed.

Results: Following search and screening processes 109 relevant publications were selected for analysis. The meta-analysis of prevalence studies indicated that the frequency of CAD among patients with COVID-19 was reported in 10 countries with an overall frequency of 12.4 % [(95% CI) 11.1-13.8] among 20079 COVID-19 patients. According to case reports/case series studies, 50.9% of COVID-19 patients suffered from CAD. Fever was the most common symptom in these patients (47%). 36.5% of these patients also had hypertension.

Conclusion: The results obtained during the present study show that the simultaneous presence of COVID-19 and CAD, especially in men and elderly patients, can increase the risks and complications of both diseases. Therefore, careful examination of the condition of this group of patients for timely diagnosis and treatment is strongly recommended.

Keywords: COVID-19; Coronary Artery Disease; CAD; Meta-analysis

Cutaneous Manifestations of the Coronavirus Disease 2019 (COVID-19): A systematic Review and Meta-Analysis

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Background and Aim: Since the onset of the 2019-nCoV disease (COVID-19), many skin manifestations have been reported in COVID-19 patients. This study aims to provide a systematic review and meta-analysis of various skin manifestations among patients with COVID-19 through case reports/case series and prevalence studies.

Methods: A systematic literature search strategy was conducted by reviewing original research articles published in Medline, Web of Science, and Embase databases in 2020. Statistical analysis was performed using STATA software, version 14.0 (Stata Corporation, College Station, Texas, USA) to report the global prevalence of skin manifestations among patients with COVID-19.

Results: Forty-three studies (35 articles were case reports/case series, and 8 articles were prevalence studies) were included in our study. A meta-analysis of prevalence studies showed that skin manifestations among patients with COVID-19 were reported in four countries (China, Thailand, France, and Italy), with an overall prevalence of 1.0% [(95% CI) 0.1–1.9] among 2,621 patients. Evaluation of the results of the case reports/case series revealed that, out of 54 patients with COVID-19, 48 patients (88.8%) showed skin manifestations. Erythematous rash (59.1%) and urticaria (14.8%) were the most common skin manifestation reported in studied patients.

Conclusion: Infection with 2019-nCoV may lead to skin manifestations with various clinical symptoms. These clinical features combined with clinical symptoms of COVID-19 may aid in the timely diagnosis of patients with COVID-19.

Keywords: corona virus; 2019-nCoV; COVID-19; skin manifestations; meta-analysis

Transcriptional Signature Analysis of PBMCs in Early Stage of Hepatitis B Related Hepatocellular Carcinoma

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Background and Aim: Hepatocellular carcinoma (HCC) is a common malignant tumor with high morbidity and mortality. The appropriate prediction of diagnosis and prognosis is of great emphasis to improve the survival rate of patients. This study aims to evaluate gene expression levels of previously identified transcripts for chronic hepatitis B (CHB) and HCC patients.

Methods: To evaluate previously analyzed transcriptome array data, we selected seven differentially expressed genes (DEGs) that were common in normal vs CHB and CHB vs HCC patients (CD44, SP3, USP8, E2F2, UFM1, IRF2BP2, and TIA1). To this goal, the study included individuals with treatment naïve CHB (n=30), primary HCC (n=25), and healthy control (n=15). Subsequently, the expression of genes was assayed using qRT-PCR. Phylogenetic analysis was applied using direct sequencing on HBsAg.

Results: In HCC patients, 60 % (n = 15) were positive for HBeAg. None of the CHB patients were positive for HBeAg, but all were positive for anti-HBe. HBV load was significantly higher in HCC patients than CHB subjects. All patients were ethnically Iranian and HBV genotype D. There was a higher expression of five transcriptional signatures (CD44, SP3, USP8, E2F2, and UFM1) in patients with HCC than CHB and healthy subjects, which was more approximately consistent with the results of the initial microarray data analysis.

Conclusion: The findings indicate that transcriptional signatures may be associated with HCC pathogenesis and serve as diagnostic biomarkers for monitoring and early diagnosis of HCC.

Keywords: Chronic hepatitis B; Hepatocellular carcinoma; Differentially expressed genes

Molecular Identification of Staphylococcus Spp. Virulence Genes Isolated of Patient Lungs Hospitalized in Intensive Care Unit in the City of Kerman in the Year 2020

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Background and Aim: Staphylococcus aureus is one of the most common nosocomial infections. Today there are reports that the prevalence of methicillin resistance in Staphylococcus aureus is increasing in different parts of the world. The aim of this study was to investigate the molecular identity of Staphylococcus aureus genes isolated from lung secretions of patients admitted to the intensive care unit

Methods: This cross-sectional descriptive study was performed on 60 hospital samples. First, biochemical tests were performed on the samples to isolate and confirm the genus Staphylococcus aureus. After bacterial enrichment and isolation and DNA extraction, mec A and Luk S / F-PV genes were evaluated by multiplex PCR.

Results: The results showed that from a total of 60 samples, 20 isolates of Staphylococcus aureus were isolated. In these 20 isolates of Staphylococcus aureus, the frequency of Luk S / F-PV gene was reported in 12 isolates and the frequency of mec A gene was reported in 8 isolates, of which 4 isolates had both genes

Conclusion: According to the results of this study, it can be said that due to the importance of Staphylococcus aureus as the most important pathogen in developing countries and due to increasing consumption and resistance to antibacterial agents, it poses a serious threat to public health. Multiple PCR is a simple, sensitive, low-cost, relatively fast, and highly specific method that can detect multiple genes in bacterial strains simultaneously

Keywords: Staphylococcus aureus; mec A; Luk S / F-PV

The Role of Probiotics as Anti-Inflammatory Agents in Covid-19

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Background and Aim: In December 2019, the Coronavirus 2019 (COVID-19) disease, caused by a novel coronavirus SARS-CoV-2 strain, emerged in Wuhan, Hubei province, China and shortly spread across the globe. To date, more than 9.2 million people have contracted COVID-19 infection with over 470,000 deaths worldwide. The highest mortality and morbidity in SARS-CoV-2 infection occurs in elderly and those with Co-morbidities including obesity and diabetes. It is noteworthy that these conditions may be associated with the gut microbiota dysbiosis. Therapeutic options are limited as one of the several factors that may contribute to severity of infection is gut microbiome status. Specifically, it is essential to have safe and effective interventions to prevent, reduce susceptibility and alleviate the severity of COVID-19. Interventions targeting the gut microbiome may have systemic antiviral effects in SARS-CoV-2 infection.

Methods: The current understanding constitutes the notion that the gut microbiome can both regulate and be regulated by invading viruses, enabling either stimulatory or suppressive effects. Some commensal gut bacteria can contribute to defense against potential pathogens by communicating with human cells and promoting favorable immune interactions. while the advantageous effect of probiotics (defined as live microorganisms that when administered in adequate amounts confer a health benefit on the host) was originally thought to stem from progress in the intestinal microbial balance, there is now substantial evidence that probiotics can also prove helpful by modulating host immune responses.

Results: Probiotics have been shown to shape T cell subsets responses , stimulate antimicrobial peptide production by Paneth cells and direct Th17 cells differentiation in the small intestinal tract. Laboratory studies have reported that certain probiotics have anti-viral effects including those against other forms of coronavirus. Further research has reported the potential of probiotics to interact with ACE2, the host receptor of SARS-CoV-2 virus. Paenibacillus bacteria were shown to naturally produce carboxypeptidases homologous to ACE2 in structure and function. Some clinical trials have demonstrated that specific probiotics can reduce the frequency and duration of common upper respiratory tract infections, especially in children as well as some adults and long-term nursing home residents. As empirical antibiotics are widely used in the early phase of COVID-19 outbreak and may lead to more severe and unfavorable dysbiosis, reinforcement of colonic microbiota using probiotics has been proposed to reduce susceptibility to consequent secondary co-infections. In an anti-inflammatory response, some probiotic strains can induce regulatory T cells, via dendritic cell modulation in the gut mucosa, inducing IL-10, TGF- β , and enhancing the IgA excretion and gut barrier function.



Conclusion: SARS-CoV-2 associated-immune dysfunction and alterations in the gut microbiota present an exciting prospect for studies and discovery of microbial therapeutics to prevent and treat SARS-CoV-2 infection. There is a insistent need to characterize specific microbial species, prebiotics, or a combination that can boost immunity, and better recognize mechanisms of precision symbiotic with anti-viral immunity for SARS-CoV-2 infection. Significantly, different methodologies should be adopted to study associations between dietary and microbiome effects and susceptibility to SARS-CoV-2 infection and disease severity.

Keywords: Covid-19; Probiotics; anti-inflammatory



A New Approach to Microbiome and Probiotics in Therapy Diseases

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Background and Aim: Now that we are under attack by viruses, The Coronavirus Disease-2019 (COVID-19) epidemic by SARS-CoV-2 is spreading worldwide, and by March 1, 2020, 67 countries including corona, we need to pay more attention to the bacteria that have coexisted peacefully in our digestive system for millions of years. Probiotic bacteria live in the gastrointestinal tract, mouth and throat, providing conditions that prevent the growth and multiplication of viruses as well as bacteria and fungi, and leave micronutrients that are effective in strengthening the body. However microbiota are involved in autoimmune and inflammatory diseases, allergies, metabolic disorders, neurological obesity and psychological disorders.

Methods: This study data collection by articles in internal databases such as, Irandoc, SID and external databases including Science Direct, Scopus, PubMed, Elsevier, Google Scholar

Results: Direct relationship between the presence of beneficial microbiota, development of immune function has it. Therefore, adjusting the microbiota balance is one of the most important strategies to improve health, microbiome balance caused by regulating the level and type of autoantibodies, beta-cell autoimmunity and appetite regulating hormones and the cellular immune response.

Conclusion: Researchers have for the first time been able to identify the microbial patterns that are associated with the severity of coronary heart disease by examining the relationship between intestinal microbiome and the severity of coronary heart disease.

Keywords: Probiotic; microbiome; COVID-19; biofilm; microbiota

Antibacterial Activity of Herbal on Helicobacter Pylori Infection

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Background and Aim: Helicobacter pylori (HP) is the most prevalent gastric pathogens, causing gastric dysfunction, peptic ulcer and carcinoma. Existing options for HP eradication treatments are severely limited. Antibiotic resistance, as a rapidly growing problem, may interfere with the success of eradication therapy. Plants as a source of medicinal compound have continued to play important role in the maintenance of human health since ancient times. The aim of this review study was to determine the effectiveness of Cinnamon, Garlic, Ginger, Honey and Thyme against HP.

Methods: The current review was conducted in 2020 year by reviewing all full text of English language articles with subject effect of Cinnamon, Garlic, Ginger, Honey and Thyme on HP that were published in PubMed and Google Scholar databases. For this purpose, the papers with keywords pylori and Cinnamon, pylori and Garlic, pylori and Ginger, pylori and Honey and pylori and Thyme in their title were selected.

Results: this systematic review included 147 articles, after the removal of repetitive, not available, non-English and Farsi language, review articles and thesis; finally were reviewed 70 full text articles. All articles extracted from PubMed are presented in the supplementary table.

Conclusion: The results demonstrated although the many of studies suggest anti-HP effects of Cinnamon, Garlic, Ginger, Honey and Thyme, but some of studies shown these herbals have not significant statistical anti-HP activity. We concluded that Cinnamon, Garlic, Ginger, Honey and Thyme can be useful as natural compounds to complement and expedite therapy and prevention of HP infection.

Keywords: Helicobacter pylori; herbal; infection

Antibacterial Effect of *Kombucha* Mushroom Extract on *Pseudomonas aeruginosa* Gram-Negative Bacteria

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Background and Aim: *Pseudomonas aeruginosa* is an opportunistic human pathogen and one of the four important hospital pathogens and biofilm formation. Treatment of *P. aeruginosa* infection with antibiotics was often difficult due to the high potential of this pathogen to develop resistance. Multidrug-resistant strains have been classified by several health care organizations as a serious threat to public health. Alternative therapies may reduce drug resistance to some extent. *Kombucha* mushroom, due to its rich polyphenolic compounds, has antioxidant, antimicrobial, anti-cancer, and anti-diabetic properties that are effective in lowering cholesterol and blood pressure and in the treatment of many disorders and diseases, and therefore its consumption has been increasing. Aim this study to investigate the effect anti-bacterial of *Kombucha* mushroom extract on *P. aeruginosa*.

Methods: In this study were used, one clinical isolate *P. aeruginosa* and from the wound and *Pseudomonas aeruginosa* ATCC27853. *Kombucha* mushroom was extracted by the Soxhlet extractor. the minimum inhibitory concentration (MIC), the minimum bactericidal concentration (MBC) Were used to evaluate the inhibitory and bactericidal effect of *Kombucha* mushroom. Then, Micro-Titer Plate (MTP) biofilm assay was used to identify isolates biofilms. Determination of minimum biofilm eradication concentration on clinical and standard pseudomonas was performed by the MBEC method.

Results: The results showed MIC *Kombucha* extract on *P.aeruginosa* and ATCC27853 250 µg/ml. MBC test showed that *Kombucha* extract at 4MIC concentration had a bactericidal ability for *P. aeruginosa*. According to the result of the MBEC method *Kombucha* extract at a concentration of 8MIC in ATCC27853 and was able to inhibit biofilm but in *P. aeruginosa* was ineffective on biofilm removal. The Results show of the MTP method, clinical isolated moderate biofilm, and ATCC25923 produced a strong biofilm.

Conclusion: The *Kombucha* mushroom was able to inhibit the growth of *P. aeruginosa* but was not able to inhibit the production of biofilm virulence factors. Therefore, the *Kombucha* mushroom was a weak anti-bacterial.

Keywords: *Kombucha* Mushroom extract; Biofilm; *Pseudomonas aeruginosa*; anti-bacterial

Antibiotic Resistance in Diabetic Foot Infection and How to Change Its Pattern Before and During COVID-19 Epidemic

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Background and Aim: The development of diabetic foot infection (DFI) is strictly associated with an open wound, but other risk factors include peripheral neuropathy and arteriopathy, diabetes-related immune dysfunction, renal impairment and the chronic course of the wound itself. The specimens underwent gram staining and inoculation on both selective and non-selective media including MacConkey agar, blood agar, chocolate agar and 5% (v/v) BA supplemented with vitamin K1 (1 µg/ml), gentamicin (75 µg/ml) (GBA) and haemin (5 µg/ml). The inoculated plates were incubated under conducive atmospheric conditions for 24–48 h to enable bacterial isolation. Isolated organisms were identified by traditional microbiological methods. In 2020 the patients' mean age was lower significantly. More patients in 2020 had history of previous ulcers. A higher rate of antibiotic resistance was detected in 2020. According to the IDSA/IWGDF classification, the prevalence of severe infections was significantly higher in 2020. The most common microbial isolates in Gram-positive bacteria were *Staphylococcus aureus*, Gram negative bacteria was *Pseudomonas aeruginosa* and Anaerobes was *Bacteroides* species respectively. Among antibiotic resistance risk factors, there was a higher rate of domiciliary antibiotic therapies, which were often self administered in the 2020 group of patients. Antibiotics may determine adverse effects or toxic reactions.

Methods: The diagnosis of infection was performed by both clinical signs and soft tissues' and/or bone's biopsy. The University of Texas Classification System was used to classify the ulcers, whereas the IDSA/IWGDF classification was used to define the severity of the infections. The classifications of wounds were according to the severity which was mild, moderate or severe based on the size, depth and level of the tissue involved in the infection, presence of metabolic instability or any systemic manifestations of infection. The specimen cultures were obtained once the wound surface had been irrigated with copious normal saline, and we subsequently debrided the superficial tissue and separated them from the exudates to isolate the colonizing pathogenic flora. Specimens were obtained by curetting the ulcer base post debridement and by aspirating the infected skin or deep tissues. The specimens underwent gram staining and inoculation on both selective and non-selective media including MacConkey agar, blood agar (BA, Basingstoke, UK), chocolate agar and 5% (v/v) BA supplemented with vitamin K1 (1 µg/ml), gentamicin (75 µg/ml) (GBA) and haemin (5 µg/ml). The inoculated plates were incubated under conducive atmospheric conditions for 24–48 h to enable bacterial isolation. Isolated organisms were identified by traditional microbiological methods.

Results: In 2020 the patients' mean age was lower significantly. More patients in 2020 had history of previous ulcers. A higher rate of antibiotic resistance was detected in 2020. According to the

IDSA/IWGDF classification, the prevalence of severe infections was significantly higher in 2020. The most common microbial isolates in Gram-positive bacteria were *Staphylococcus aureus*, Gram negative bacteria was *Pseudomonas aeruginosa* and Anaerobes was *Bacteroides* species respectively.

Conclusion: Among antibiotic resistance risk factors, there was a higher rate of domiciliary antibiotic therapies, which were often self administered in the 2020 group of patients. This was also confirmed by the logistic regression analysis, which identified history of prior hospitalization, antibiotic self administration and antibiotic prescription by general practitioners as independent predictors of antibiotic resistance in these cohorts of patients. Moreover, antibiotic resistant infections usually require the administration of specific antibiotics and the hospitalization, which is itself a notorious risk factor for antibiotic resistance. Furthermore, antibiotics may determine adverse effects or toxic reactions, which may be responsible for the worsening of the systemic clinical conditions and a dangerous reduction of patient's compliance to the therapy .

Keywords: Diabetic foot infection; Antibiotic resistance; Amputation; Diabetic foot ulcer; Bacteriology profile; Microbial sensitivity tests

Antibiotic Resistance of *Acinetobacter* Species Isolated from Patients in Different Wards of Rouhani Hospital in Babol

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Background and Aim: The study of antibiotic resistance in bacteria is very important in the management of treatment and control of infectious diseases. The aim of this study was to determine the level of bacterial contamination and antibiotic resistance of *Acinetobacter* species isolated from patient samples in different wards of Rouhani hospital in Babol.

Methods: The information of patients from the hospital information system of Rouhani hospital in Babol in the first 6 months of 2021 was analyzed. *Acinetobacter* was detected by microbiological laboratory methods and microbial susceptibility testing was performed by disk diffusion method according to the standard of the Clinical and Laboratory Standards Institute (CLSI). For data analysis, SPSS software version 21 and frequency table were performed (P-value less than 0.05).

Results: Fifthly tow species of *Acinetobacter* were isolated from patients. Antibiotic resistance was as follows; Piperacillin / Tazobactam for 52 isolates (breakpoint: 18-20, resistant: 94.2%, intermediate: 0, sensitive: 5.8%: Confidence interval (CI): 83-98.5). Ceftazidime for 31 isolates (breakpoint: 15-17, resistant: 96.8%, intermediate: 3.2, sensitive: 0: CI: 81.5-99.8). Ceftriaxone for 40 isolates (breakpoint: 14-20, resistant: 100%, intermediate: 0, sensitive: 0: CI: 89.1-100). Meropenem for 52 isolates (breakpoint: 15-17, resistant: 100%, intermediate: 0, sensitive: 0: CI: 91.4-100). Amikacin for 51 isolates (breakpoint: 15-16, resistance: 88.2%, intermediate: 0, sensitive: 11.8: CI: 75.4-95.1). Gentamicin for 1 isolate (breakpoint: 13-14, resistant: 0, intermediate: 0, sensitive: 100: CI: 0.0-94.5). Ciprofloxacin for 52 isolates (breakpoint: 22-25, resistant: 92.3, intermediate: 3.8, sensitive: 3.8: confidence: 80.6-97.5). Trimethoprim / sulfamethoxazole for 1 isolate (breakpoint: 11-15, resistant: 0, intermediate: 0, sensitive: 100: CI: 0.0- 94.5).

Conclusion: High antibiotic resistance was observed in *Acinobacter* isolated from the samples. The importance of conducting antibiotic susceptibility testing prior to drug administration is effective in the treatment and control of drug resistance.

Keywords: Infection rate; Antibiotic resistance; *Acinetobacter*

Antibiotic Resistant Rate of *Pseudomonas* spp. Species Isolated from Patients Samples

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Background and Aim: *Pseudomonas* spp. is an opportunistic nosocomial pathogen, especially in immunocompromised hosts, which has become one of the most prominent gram-negative bacteria causing nosocomial infections. This bacterium is resistant to many antibiotics. The aim of this study was to determine the antibiotic resistance of *Pseudomonas* spp. strains isolated from patient samples.

Methods: The information of 53 *Pseudomonas* spp. isolates was gathered from microbiology laboratory of Rouhani Hospital, Babol University of Medical Sciences in Babol in The first six months of 2021. Bacteria were identified by laboratory microbiological methods; also microbial susceptibility testing was performed by disk diffusion method. For data analysis, SPSS 22 and Chi-square test were used (P-value less than 0.05).

Results: Antibiotic resistance was as follows; Piperacillin/Tazobactam for 49 isolates (resistant: 46.90%, sensitive: 49.00%; Confidence interval (CI): 32.70-61.50), Ceftazidime for 17 isolates (resistant: 35.3%, sensitive: 64.7%; CI: 15.30-64.40), ceftriaxone for 35 isolates (resistant: 85.7%, sensitive: 11.40%; CI: 64.90-94.60), meropenem for 53 isolates (resistant: 75.50%, sensitive: 22.60%; CI: 61.50-85.80), amikacin for 53 isolates (resistance: 35.80%, sensitive: 60.40%; CI: 23.40-50.20), ciprofloxacin for 53 isolates (resistance: 47.20%, sensitive: 49.10%; CI: 33.50-61.30), colistin for 32 isolate (resistant: 3.10%, sensitive: 96.90%; CI: 0.20-18.00).

Conclusion: The results of this study showed high antibiotic resistant rate of *Pseudomonas* spp. isolated from different specimen. Prescribing appropriate antibiotics in the correct choice of treatment and reduction antibiotic resistance is effective.

Keywords: Antibiotic-Resistant; *Pseudomonas* spp; Patients

Comparison the Antibacterial Effect of Gold Nanoparticle with Herbal Extracts of *Salvia Officinalis* & *Zataria Multiflora* on Prevalent Pathogenic Bacteria: *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* & *Klebsiella pneumoniae*

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Background and Aim: Ever-increasing resistance of bacterial pathogens to different classes of antibiotics, causes that researchers try to finding suitable substances instead of antibiotics and compare their efficacy to each other. One group of these substances are herbal extracts which gathered from medicinal plants. Recently, by progressing of science about nanotechnology, the efficacy of different nanoparticle are examined, too. The aim of this study is comparing the antibacterial efficacy of nano-gold particle with herbal extracts of *Salvia officinalis* & *Zataria multiflora* on some prevalent pathogenic bacteria.

Methods: In this study, ethanolic extract of *Salvia officinalis* & *Zataria multiflora* were prepared. Gold nanoparticle was prepared, too. Then effect of different concentration of each one were determined on *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* in the in vitro condition by serial dilution method and their antimicrobial activity were investigated using the micro broth dilution method by determining the minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC) separately.

Results: Based on results which gathered from performed experience, *Salvia officinalis* & *Zataria multiflora* have antimicrobial activity on all strains which we examined them. the herbal extract minimum inhibitory concentration was 10%, and the maximum concentration was 70% . Whereas gold nanoparticle had inhibitory effect on *staphylococcus aureus* but it didn't inhibit growth of gram negative bacteria.

Conclusion: A comparison of the results of inhibitory effects revealed that *Salvia officinalis* and *Z. multiflora* extracts had more potent antibacterial activity than gold- nano particle, and gram positive bacteria are more sensitive to gold particle in compare with gram negative bacteria.

Keywords: Antibacterial effect; Nano-Gold; *Salvia officinalis*; *Zataria multiflora*

Detection of TVV1 and TVV2 (*Trichomonas vaginalis* Virus) from *Trichomonas vaginalis* Isolates from Alborz Province

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Background and Aim: *Trichomonas vaginalis* can be infected with double stranded RNA (dsRNA) viruses known as *T. vaginalis* virus (TVV). This viral infection may have important implications for trichomonal virulence and disease pathogenesis. In this study we identified and genetic characterized two strains of TVVs isolated from *T. vaginalis* in Alborz province, Iran.

Methods: *Trichomonas vaginalis* can be infected with double stranded RNA (dsRNA) viruses known as *T. vaginalis* virus (TVV). This viral infection may have important implications for trichomonal virulence and disease pathogenesis. In this study we identified and genetic characterized two strains of TVVs isolated from *T. vaginalis* in Alborz province, Iran.

Results: Among 12 isolates of *T. vaginalis*, three of them were infected with TVV1(2 isolates) and TVV2(1 isolates). One of any TVVs, were sequenced. The sequences were edited and aligned and then the nucleotide sequences were entrusted at GenBank under accession numbers OL690432 (TVV1) and OL804276 (TVV2).

Conclusion: For the first time in Alborz province, TVV1 and TVV2 were detected using RT-PCR in *T. vaginalis*

Keywords: *Trichomonas vaginalis* virus; TVV1; TVV2; RT-PCR



Medical Bacteriology



Elucidating the Association Between Gut Microbiome and Covid 19 Severity, Regard to the Probable Role of TMAO Produced by the Gut Microbiome

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Background and Aim: In recent years COVID-19 infection spread rapidly across the world and has led to profound impacts on public health with almost one million mortalities. Emerging evidence has suggested an association between some metabolites produced by gut microbiota with the infection that may contribute towards the severity of infection.

Methods: Several investigations have suggested an association between TMAO levels and the risk of inflammation and thrombosis. Gut-TMAO through different molecular mechanisms such as activation of NF-KB molecular pathway and induce the expression of SRs in the surfaces of macrophages and foam cells formation increases inflammation.

Results: Several studies have shown that higher TMAO increased expression of TNF- α and IL1 β as pro-inflammatory cytokines and decreased expression of the anti-inflammatory cytokine IL-10. Furthermore, Gut-TMAO by platelet aggregation and adhesion to collagen increase the risk of thrombosis.

Conclusion: Understanding the association between gut microbiome compositions such as Gut-TMAO and their effects on Sars-Covid19 infection progression help to control disease severity. In this review, we make a hypothesis that the Gut-TMAO has the potential to increase Covid disease severity.

Keywords: gut; microbiome; Covid-19; TMAO

Evaluation of Active Constituents and Antibacterial Activity of Aqueous and Methanol Extracts of *Rumex alveollatus* Against *Klebsiella pneumoniae*

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Background and Aim: Antimicrobial resistance in *Klebsiella pneumoniae* has been increasing in recent years. Therefore, many efforts have been made to find new active plant compounds as an appropriate substitute for antibiotics. This Evaluation of active constituents and antibacterial activity of aqueous and methanol extract of *Rumex alveollatus* against *Klebsiella pneumoniae*

Methods: This study was performed on standard strain of *Klebsiella pneumoniae* and a number of clinical strains. The aqueous and methanol extracts of *Rumex alveollatus* were prepared by Soxhlet apparatus. Antibacterial activities of extracts were evaluated well diffusion method. Also minimum inhibitory concentration (MIC) and MBC were assessed by microdilution method. Active compounds were identified by gas chromatography-mass. The data obtained by new SPSS software was analyzed.

Results: Inhibitory zone of methanol (500 mg/ml) on standard and clinical strains of *Klebsiella pneumoniae* were 20.83±0.78 and 18.10± 1.04 mm, respectively and also inhibition zone of aqueous (500 mg/ml) on standard and clinical strains of *Klebsiella pneumoniae* were 16.00±1.00 and 13.10± 1.04 mm, respectively. MIC results for methanol and aqueous extracts were 125 and 250 mg/ml, respectively. The highest compound isolated from the extract (1,2-Benzenedicarboxylic Acid, Diisooctyl Ester), (Benzene, 1-Methyl-2) and (1,4-Cyclohexadiene, 1-Methyl-4) 22.25%, 18.1% and 13.8%, respectively.

Conclusion: *Rumex alveollatus* methanol and aqueous extract have good antibacterial activity against tested bacteria. Therefore, the results of this study can be a valuable report regarding their effective role in infection control.

Keywords: Methanolic Extracts; Aqueous Extracts; Antibacterial Activity; Active Constituents; *Rumex Alveollatus*

Evaluation of Antibacterial and Anti-Biofilm Activity of Thyme Essential Oil on Some Bacterial Pathogens from Cystic Fibrosis Patients

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Background and Aim: Cystic fibrosis (CF) is an autosomal recessive disease characterized by respiratory, gastrointestinal, and reproductive systems problems. Patients with CF often suffer from chronic pulmonary infections due to the *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Klebsiella pneumoniae* and some other pathogens. These bacteria produce biofilms that make them more resistant to conventional antibiotics, in addition to their innate antibiotic resistance. Consequently, to improve global health, it is vital to identify innovative antibacterial medicines to combat multidrug-resistant strains. Essential oil of some medicinal plants including thyme exhibits broad-spectrum antimicrobial activity. In this study, we studied the antibacterial and antibiofilm activity of thyme essential oil on common bacterial pathogens from CF patients' lung infections.

Methods: Minimum inhibitory concentration (MIC) and Minimum biofilm inhibitory concentrations (MBICs) of thyme essential oil were determined against 30 isolates from three different genera including *P. aeruginosa*, *S. aureus* and *K. pneumoniae* using microdilution method as described by Clinical and Laboratory Standards Institute (CLSI). TTC indicator was used for determining of MBIC test.

Results: *S. aureus* was shown to be the most sensitive strain, whereas *P. aeruginosa* was found to be the most resistant. Compared to the other two bacteria, *K. pneumoniae* showed intermediate resistance. *K. pneumoniae*, *P. aeruginosa*, and *S. aureus* had MIC values of (52.88

Conclusion: Thyme essential oil has a powerful effect on planktonic cells and bacterial biofilm, and it could be utilized as an alternative to antibiotics in the treatment of CF patients' lung infections. Thymol and carvacrol, two key components of thyme, are significant in the antibacterial properties of the medicinal plant.

Keywords: Biofilm; Cystic fibrosis; MIC; MBIC; Thyme

Evaluation of Antibiotic Resistance of Bacterial Strains Isolated from Different Specimens of COVID-19 Patients Hospitalized in Shahid Sadoughi Hospital in Yazd (1399)

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Background and Aim: Antibiotic resistance to infections caused by multidrug-resistant bacteria in people with COVID-19 disease is a major threat to the health worldwide. The aim of this research is evaluation of antibiotic resistance in COVID-19 patients admitted to Shahid Sadoughi Hospital in Yazd in 1399.

Methods: In this descriptive from April 2020 to March 2021, antimicrobial resistance of 1549 gram (+) and gram (-) bacteria isolated from various specimens of hospitalized COVID-19 patients in Shahid Sadoughi Hospital in Yazd city was determined by disk diffusion method. Etest method was used for MIC detection of vancomycin and colistin according to CLSI 2020 protocols and analyzed by SPSS 25.

Results: Among 1549 bacterial isolates, the most isolates were identified as *Escherichia coli* (33.05%), *Staphylococcus aureus* (20.53%), *Pseudomonas aeruginosa* (14.59%), *Klebsiella pneumonia* (9.94%) and *Acinetobacter* spp. (7.68%), respectively. The highest antibiotic resistance of 512 *Escherichia coli* isolates was against ampicillin (85.2%), trimethoprim/sulfamethoxazole (68.7%) and ceftazoline (67.9%) and the lowest resistance was for nitrofurantoin (5.3%) and imipenem (6.2%), respectively. The highest resistance of 318 isolates of *S. aureus* were seen for penicillin G (96.8%), erythromycin (74.3%) and clindamycin (57.6%) and lowest resistance to vancomycin (0%) and nitrofurantoin (10.6%), respectively. Also 55.3% of isolate were MRSA. Among 226 *Pseudomonas aeruginosa* isolates, the highest resistance was seen for ampicillin (97.3%), amikacin (53.1%) and meropenem (52.7%), and the lowest resistance to colistin (0%) and piperacillin (36.2%). 119 *Acinetobacter* spp. Isolates were shown resistance to all antibiotics (85-100%) except colistin (0%).

Conclusion: Increasing in antibiotic resistance due to improper use of antibiotics and the emergence of infections caused by multidrug-resistant bacteria increases the mortality rate of COVID-19 patients.

Keywords: Antimicrobial resistance; COVID-19; Bacterial infections.

Evaluation of Antibiotic Resistance Pattern of *Acinetobacter baumannii* Clinical Isolates in Khorramabad Hospitals

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Background and Aim: Today, drug-resistant strains of *Acinetobacter baumannii* are one of the opportunistic pathogens in the world. This study aimed to determine the antibiotic resistance pattern of clinical isolates of *A. baumannii* in Khorramabad hospitals (i.e., Shohaday Ashayer-Shahid Rahimi), Khorramabad, Iran.

Methods: This cross-sectional study was conducted on the clinical samples collected from patients hospitalized in the different wards of Khorramabad hospitals in 2015-16 and 2017-18. The clinical samples were identified as *A. baumannii* by microbiological culture and biochemical tests then confirmed by polymerase chain reaction. The susceptibility test of bacterial isolates to antibiotics was performed and the obtained data were analyzed by SPSS software (version 22) using the Chi-square test.

Results: According to the results of the antibiogram, among the 94 isolates of *A. baumannii* collected from the patients admitted to Khorramabad hospitals, 50%, 41.49%, and 8.51% of isolates were multiple-drug resistant, extensively drug-resistant, and non-multidrug resistance, respectively.

Conclusion: Due to the sensitivity of *A. baumannii* to polymyxin B and minocycline antibiotics, these antibiotics can be used, especially as a combination therapy, in the treatment of infections caused by this bacterium. Since the rate of multidrug resistance in Khorramabad hospitals was found to be 91.49%, it is necessary to pay attention to the criteria for controlling nosocomial infections.

Keywords: *Acinetobacter baumannii*; multiple-drug resistant; extensively drug-resistant

Evaluation of the Effect of Biosynthetic Nanoparticles of Copper Oxide on AmpC Gene Expression in Clinical Strains of *Pseudomonas aeruginosa*

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Background and Aim: Nowadays, the need for new strategies to control the activities of resistant bacteria is felt more than ever. In this regard, a very promising approach is the use of nanomaterials science and the knowledge that is expanding in the field of nano synthesis biosynthesis should be considered as a global strategy. The aim of this study was to evaluate the antibacterial properties of biosynthetic copper oxide nanoparticles on the expression of the AmpC gene in *Pseudomonas aeruginosa*.

Methods: Ten samples of *P. aeruginosa* were collected from clinical centers of Khorramabad city in 1399 and identified based on biochemical tests. After treatment of MDR strains with the minimum inhibitory concentration of copper oxide nanoparticles, the expression level of the AmpC gene was evaluated using real-time PCR.

Results: After applying the effect of biosynthetic copper oxide nanoparticles on AmpC gene expression and calculations performed by MIC and ANOVA one-Tukey statistical analysis test, it was found that copper oxide nanoparticles have an inhibitory effect on the expression of AmpC genes of *P. aeruginosa* samples.

Conclusion: The objectives of this study focused on the use of nanotechnology as a new tool to address current challenges in the treatment of infectious diseases and antibiotic resistance. Therefore, for safer use of this nascent technology, the need for clinical trials and further research at the level of molecular biology is recommended.

Keywords: Nanoparticle; Multidrug Resistance; Real-time PCR; *Pseudomonas aeruginosa*

Evaluation of the Inhibitory Effect of Biosynthetic Zinc Oxide Nanoparticles on the Expression of Efflux Pumps Genes (AdeA, AdeC) in Clinical Strains of *Acinetobacter Baumannii*

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Background and Aim: Introducing an effective antibacterial agent is very important in controlling pathogenic and antibiotic-resistant bacterial strains. Recent advances in nanotechnology, especially the ability to fabricate biological nanoparticles, have led to the development of a wide range of antimicrobial agents. Therefore, the aim of this study was to investigate the antibacterial properties of ZnO NPs in inhibiting resistant strains of *Acinetobacter baumannii*.

Methods: Ten samples of *Acinetobacter* were collected from clinical centers of Khorramabad city in 1399 and were identified based on biochemical tests. After treatment of MDR strains with sub-minimum inhibitory concentration (MIC) of ZnO NPs, the expression level of efflux pump genes (AdeA, AdeC) was evaluated using real-time PCR.

Results: After applying the effect of ZnO NPs on AdeA gene, 80% of the samples showed reduced expression. 20% of the sample also had incremental expression. Also in AdeC gene, 40% of the samples showed reduced expression after the effect of ZnO NPs. 60% of the samples also had incremental expression.

Conclusion: According to the results, the optimal inhibitory effect of biosynthesized ZnO NPs on the expression of AdeA and AdeC genes, which play an important role in drug resistance of the *Acinetobacter* species, was evident. On the other hand, due to the increased expression in 60% of the samples for the AdeC gene, the possibility of mutations in the regulatory genes was raised. It seems that ZnO NPs can be used as a pharmaceutical candidate in the pharmaceutical industry in the future.

Keywords: Efflux pumps; Multiple drug resistance; Nanoparticle; Zinc Oxide

Frequency and Antimicrobial Resistance Patterns of Urinary Tract Bacteria Isolated in a Zanjan Educational Hospital in 2016-2018

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Background and Aim: Bacteria play a major role in UTIs; therefore, it is necessary to be aware of their regional prevalence and the causative pathogens for better prognosis, rapid treatment and elimination of the infectious agents in clinical settings. This study aims to evaluate the prevalence of bacterial isolates involved in UTI samples and their antibiotics resistance pattern in two educational hospitals in North West of Iran.

Methods: A cross-sectional study was designed to analyze 4214 urine samples were collected from hospitalized patients in Valie-Asr hospital in Zanjan, Iran. Samples were cultured in two media including MacConkey agar and blood agar. The standard media such as oxidase and coagulase were prepared for all of the culture isolates. Considering the results and the Bergey's microbiology book guidelines, bacterial strain was identified. Afterwards, disk diffusion method was utilized for positive cultures. Finally, data were entered to SPSS 26.

Results: The study population consisted of a total number of 3582 patients with positive urine cultures, including 56% females and 44% males. *E. coli* and *Staphylococcus epidermidis* were the most frequently isolated gram negative and gram-positive bacteria in the urine samples with isolation rates of 53.43% and 15.99%, respectively. In gram negative bacteria, nitrofurantoin and vancomycin represented the smallest resistance rates at 25.27% and 26.74%. Piperacillin showed the least efficacy with a resistance rate of 76.04%. In gram positive bacteria, vancomycin and gentamicin showed more promise with respective resistance rates of 19.34% and 27.34%. The highest resistance was associated with ampicillin (68.61%).

Conclusion: Gram-negative bacteria were significantly more prevalent than gram positive-bacteria in this study and *E. coli* was the pathogen behind more than half of the infections. while Vancomycin had the lowest resistance rate for both gram-positive and gram-negative bacteria, it is considered as a last-resort antibiotic, therefore alternatives have priority for treatment procedure.

Keywords: Nosocomial infections (NIs); urinary tract infections (UTIs); antibiotics resistance pattern

Frequency Distribution and Prevalent Sequence Types of Mcr-producing *Escherichia coli* among Clinical Isolates Around the World: A Systematic Review

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Background and Aim: The continuing rise in infections caused by multi-drug resistant (MDR) bacteria is one of the most serious public health issues in today's societies. Colistin is a last-resort antimicrobial medication used to treat infections caused by MDR gram-negative bacteria; therefore resistance to this antibiotic is extremely hazardous. The current study aimed to evaluate the global prevalence and distribution of colistin resistance genes among human clinical isolates of *Escherichia coli* (*E. coli*) as a systematic review.

Methods: PubMed, Embase, and Web of Science databases were systematically searched. For further evaluation, all original English-language articles that demonstrated colistin resistance in *E. coli* clinical isolates published between 2000 and 2020 were examined.

Results: Out of 4857 initial articles, after various stages of review and evaluation, 190 related articles were selected. More than 79 % of the publications selected in this research were published from 2014 to 2020. In Asia, Europe, America, Africa, and Oceania, the prevalence of mobilized colistin resistance (mcr) producing colistin-resistant *E. coli* was 66.72%, 25.48%, 5.19%, 2.27%, and 0.32 %, respectively.

Conclusion: The recent widespread spreading of *E. coli* strains harboring mcr conferring colistin resistance, especially in Asia and Europe, is concerning and needs more attention.

Keywords: *Escherichia coli*; mcr; colistin; antibiotic resistance; systematic review

Frequency of Mutations in *gyrA* and *parC* Genes Among Fluoroquinolone Resistant *Klebsiella pneumoniae* Isolated from Urine Specimens of Hospitalized Patients in Yazd, Iran

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Background and Aim: *Klebsiella pneumoniae* is an important cause of nosocomial infections, especially urinary tract infections. Considering the antimicrobial resistance due to the increasing use of fluoroquinolones and the importance of chromosomal genes in creating high level resistance to these drugs, the aim of this study was to evaluate the frequency of mutations in *gyrA* and *parC* chromosomal genes in *Klebsiella pneumoniae* isolated from urine specimens of hospitalized patients in Yazd city.

Methods: In this descriptive cross-sectional study, 100 *K. pneumoniae* were isolated from urine specimens and identified by biochemical tests. The fluoroquinolones susceptibility test was performed by disk diffusion method and the Minimum Inhibitory Concentration (MIC) was investigated by Etest method. The amplification of *gyrA* and *parC* genes was performed by PCR method using specific primers and after sequencing, the presence of possible mutations was investigated.

Results: The results showed that the highest and lowest resistance were observed against nalidixic acid (28%) and levofloxacin (19%), respectively. Based on MIC tests, resistance to nalidixic acid and ciprofloxacin were 32% and 27%, respectively. The point mutations in *gyrA* and *parC* genes were observed in 17 resistant isolates. The most frequent mutations in *gyrA* were S83F,I and D87A,G and in *parC* were S80I,L. Increased antimicrobial resistance was observed in the *gyrA* and *parC* mutant isolates.

Conclusion: The results of the present study showed high resistance against fluoroquinolones, which is a warning sign for treatment. The prescription of fluoroquinolones should be controlled as much as possible to prevent the spread of mutant carrying clones.

Keywords: *Klebsiella pneumoniae*; Fluoroquinolones; Antimicrobial Resistance; *gyrA*; *parC*

Frequency of Virulence Factors and *stx1* and *stx2* Genes in Fecal and Raw Food Isolates of Shigatoxigenic *Escherichia coli* in Tehran

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Background and Aim: Infectious diarrheal diseases are a major problem and cause significant mortality in the world. *Escherichia coli*, which produces Shiga toxin, is a pathotype of intestinal *Escherichia coli* that causes diarrhea in humans. This pathotype can also cause hemolytic uremic syndrome, especially in children. The present study was conducted to investigate the frequency of contamination of raw foods with this bacterium and its role in the incidence of acute diarrhea in society.

Methods: In this study, 97 samples of fresh vegetables and 100 samples of fresh chicken meat from 22 areas of authorized fruit and vegetable centers of Tehran Municipality and also 322 samples of feces of outpatients referred to hospitals in Tehran before taking any antibiotics, along with demographic information were examined for STEC. Preenrichment and culture of the samples were performed according to ISO protocols and the screened colonies were characterized after DNA purification with a special kit to identify *stx1*, *stx2*, *eae* and *ExhA* genes. To determine antibiotic resistance by disk diffusion method, all STEC isolates were subjected to antibiogram according to CLSI 2018 procedures.

Results: According to the results of our study, out of 100 samples of chicken meat 13%, out of 97 samples of fresh vegetables 14% and out of 322 samples of human feces 5% of the samples contained Shigatoxigenic *Escherichia coli*. Among chicken STEC samples 1% *eae* and 2% *ExhA*, among STEC samples vegetables 3% *eae* and 4% *ExhA* and among STEC samples feces 0.6% *eae* and 1.2% *ExhA* were identified. Among stool samples, the highest incidence of STEC was between the ages of 10 and 19 years. In all samples, resistance above 80% to the antibiotic sulfamethoxazole was observed and the lowest resistance to the boutique was meropenem. Among all STEC samples, 34.33% of the samples were AmpC and 9.30% of the ESBL samples.

Conclusion: The results of this study confirmed the presence of STEC strains in raw foods and their role in the development of acute diarrhea. The presence of *eae* and *stx2* coding strains among these isolates poses a risk of HUS among consumers. This study demonstrates the need for quality control of raw food products for STEC contamination before distribution.

Keywords: STEC; *stx1*; *stx2*; *eae*; *ExhA*; AmpC; ESBL

Frequency of Virulence-Associated Genotypes of *Helicobacter pylori* and Their Correlation with Clinical Outcome and Histological Parameters in Infected Patients

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Background and Aim: *Helicobacter pylori* (*H. pylori*) is a gram-negative which can cause several gastroduodenal diseases, including gastritis and peptic ulcer disease (PUD). *H. pylori* specific genotypes have been related to increased occurrence of gastritis and PUD. The aim of this study was to investigate the clinical relevance of the major virulence factors of *H. pylori* with clinical outcomes and histological parameters in Iranian patients.

Methods: Totally, 200 subjects with PUD and gastritis disease who underwent gastroduodenal endoscopy were enrolled in this study. The presence of the *cagA*, *vacA*, *oipA*, *babA2*, and *iceA* genes in antral gastric biopsy specimens were determined by polymerase chain reaction (PCR) and the results were compared with clinical outcomes and histological parameters.

Results: The frequency of *babA2*⁺, *oipA*⁺, *vacA* s1/m2, and *vacA* m2 genes was significantly higher in infected subjects with PUD compared with infected subjects with gastritis. In contrast, the frequency of *vacA* s1/m1 gene was significantly higher in gastritis subjects than PUD subjects. The high-density scores of *H. pylori* were strongly associated with *iceA1*⁺, *babA2*⁺, and *oipA*⁺ genes. Additionally, the high polymorphonuclear cell infiltration and high mononuclear cell infiltration scores were strongly associated with the *cagA*⁺, *iceA1*⁺, *oipA*⁺ genes and *cagA*⁺, *babA2*⁺, *oipA*⁺ genes, respectively.

Conclusion: Our study indicated that the *vacA*, *babA2*, and *oipA* virulence factors are related to a higher risk of PUD in subjects with *H. pylori*-infection. Infection with these strains was associated with a more severe gastropathy.

Keywords: *Helicobacter pylori*; Virulence factors; Gastritis; Peptic ulcer disease; Inflammation

Global prevalence of blaNDM-producing *Acinetobacter baumannii* strains isolated from clinical samples: a systematic review

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Background and Aim: The increasing number of nosocomial infections associated with *Acinetobacter baumannii* (*A. baumannii*) is causing this bacterium to become a more clinically significant organism. There is great medical concern about the emergence of NDM-type carbapenemases in *A. baumannii* strains that impede the effectiveness of practically all antimicrobial agents. The present systematic review aimed to evaluate the worldwide prevalence of NDM variants among carbapenem-resistant *A. baumannii* strains.

Methods: Using PubMed, Web of Science, and Embase databases, we identified all studies addressing the prevalence and variants of NDM in *A. baumannii* isolates. Publications selected for this review were published between 2000 and 2020.

Results: After selecting and reviewing the publications, finally, 67 articles were qualified to be reviewed. The results indicated that in Europe, NDM variants exhibit the greatest diversity. The prevalence of NDM-1 in Asia, Africa, and Europe was 64.02%, 28.05, and 5.56%, respectively. Studies that examined the genotypes of NDM-producing *A. baumannii* strains most commonly used PCR.

Conclusion: We have seen a rise in reports of strains of *A. baumannii* producing NDM on all continents over the past few years, so health care providers and physicians, as well as microbiologists, must pay more attention to prevent the spread of these strains.

Keywords: *Acinetobacter baumannii*; New Delhi Metallo- β -Lactamase; Carbapenemas; Systematic Review

Investigation of Antibacterial Effect of Ferula Macrocolea Extract and Quantity Determination of Inhibitory Effect on 4 Standard Strains of Gram-Positive and Gram-Negative Bacteria.

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Background and Aim: Today Antibiotic resistance is one of the global concerns in medicine. due to the increasing emergence of antibiotic-resistant bacteria and the problems with their treatment, the use of alternative methods to increase effectiveness and reduce drug resistance is important. In traditional medicine, the use of ferula plants due to it is various active chemical compounds with antibacterial, antioxidant, and cytotoxic activity, has attracted the attention of researchers, Because many studies have been performed on other species of Ferula. the aim of this study was to investigate the antimicrobial effect of Ferula macrocolea extract on 4 standard strains of gram positive and gram-negative bacteria.

Methods: the study was performed on 4 standard bacterial strains of *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Escherichia coli*, and *Bacillus subtilis*. First, Ferula essential oil was extracted. Microbial suspension with specific and standard concentrations was prepared in Müller-Hinton Broth medium. antimicrobial properties of the essential oil on the desired bacteria were performed using two methods of disk diffusion and micro broth dilution according to the CLSI standards.

Results: The results of this study showed that Ferula extract has a different antimicrobial effect on all strains. The highest and lowest antimicrobial and inhibitory effects of this extract were reported on *Bacillus subtilis* and *Staphylococcus epidermidis*, respectively.

Conclusion: In our study, ferula extract had different inhibitory and lethal effects on all standard gram-positive and gram-negative strains in this experiment. it was shown that a very small concentration of extract would be effective in inhibiting and killing the *Bacillus subtilis* strain, while the *Staphylococcus aureus* strain would be inhibited in higher concentrations of this extract.

Keywords: Ferula macrocolea; antibiotic resistance; antibacterial effect; gram-positive; gram-negative bacteria

Investigation of Antibiotic Profile and Biofilmogenic Ability in *Klebsiella pneumonia* Isolates with Carbapenem Resistance.

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Background and Aim: *Klebsiella pneumoniae*, a gram-negative bacillus, is a member of the enterobacteriaceae family and that is one of the most important causes of nosocomial infections capable. Infections caused by *Klebsiella pneumoniae* with carbapenem resistance as an important treatment problem are increasing worldwide. The aim of the present study was to investigate the antibiotic profile and the ability of biofilm production of *Klebsiella pneumoniae* isolates to have carbapenemase resistance.

Methods: Number of 100 isolated *Klebsiella pneumonia* were collected from patients in hospitals and laboratories in KhorramAbad city, and after confirmation of the samples with standard biochemical experiments, the samples with the enzyme method of carbapenemase enzyme were screened with the help of the phenotypic method. Then, the antibiotic profile of the obtained samples, along with the ability to form biofilm by microplate method in laboratory conditions were examined.

Results: Of the 100 samples, 39 (39%) had carbapenemase enzyme. The highest antibiotic resistance was observed with ampicillin, chloramphenicol and ciprofloxacin antibiotics, and the lowest resistance was related to gentamicin, amikacin and cotrimoxazole. Most samples were able to form biofilm.

Conclusion: The present study suggests that, unfortunately, the insulin resistance of *Klebsiella pneumonia* is increased compared to carbapenem antibiotics, and that the samples studied have a higher resistance to antibiotics such as phosphomycin compared to previous studies. It is recommended that these isolates be evaluated for the presence of resistance genes to Colistin and phosphomycin.

Keywords: *Klebsiella pneumoniae*; carbapenemase; Biofilm; Antibiogram

Investigation of Environmental Contamination with Staphylococci in the Intensive Care Unit of Sanandaj hospitals

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Background and Aim: Nosocomial infections with staphylococci, especially in the ICU, are one of the main factors increasing the cost of treatment and transmission of the infection to the community. The aim of this study was to determine the level of environmental contamination of the ICUs of teaching hospitals in Sanandaj with three important species of *Staphylococcus* (*Staphylococcus aureus*, *Staphylococcus epidermidis* and *Staphylococcus saprophyticus*) and their antibiotic resistance.

Methods: In this cross-sectional study, samples were collected by swab for 3 months at two-week intervals from different parts of the ICU and after culture, were incubated at 37° C for 24 to 48 hours. Suspicious colonies were identified using standard methods such as catalase, coagulase, mannitol fermentation, etc. Identification of heat-resistant nuclease gene (nuc) by polymerase chain reaction (PCR) was used to confirm *S. aureus*. Antibiotic susceptibility was determined by disk diffusion method for 7 antibiotics and methicillin resistance was determined using cefoxitin disk according to CLSI 2020 standard. Methicillin resistance gene *mecA* and class 1 integron were also identified by PCR.

Results: In this study, out of 90 samples contaminated with the bacteria; 29 samples (32.2%) were contaminated with *S. aureus* and 25 samples (27.8%) with *S. epidermidis*. *S. saprophyticus* was not identified. *S. aureus* showed the highest sensitivity to linezolid (100%), followed by cotrimoxazole (96.5%) and ciprofloxacin and gentamicin (93.1%, each). Seventeen isolates were resistant to cefoxitin (58.6%) (MRSA), of them 16 isolates carried *mecA*. *S. epidermidis* also showed the highest susceptibility to linezolid (100%), followed by cotrimoxazole and gentamicin (52% each) and ciprofloxacin (44%). Sixteen isolates were resistant to cefoxitin (64%) (MRSE), of them 14 isolates harbored *mecA* gene. Also, 4 *S. aureus* (13.8%) and 3 *S. epidermidis* isolates (12%) contained class 1 integron.

Conclusion: Our study showed a high prevalence of *S. aureus* and *S. epidermidis* isolates in ICU. Also, a high percentage of strains were resistant to methicillin. Given the risk of transmitting nosocomial infections, detecting contamination of environmental surfaces in hospitals can help reduce the risk of these types of infections.

Keywords: Methicillin-resistance; Nosocomial infections; Environmental surfaces; ICU

Investigation the Effect of COVID-19 on *Mycobacterium tuberculosis* Challenges and Opportunities (Systematic Review)

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Background and Aim: Coronavirus disease 19 (COVID-19) is a global threat that caused enormous damage. *Mycobacterium tuberculosis* (TB) is also one of the world's top ten causes of death. SARS-CoV-2 and *M. tuberculosis* diseases have vital parallels with respect to cell entry, resistant reaction, and safe avoidance. So the aim of this study is to investigate the effect of COVID-19 on *Mycobacterium tuberculosis* challenges and opportunities.

Methods: This review article was performed within articles published at PubMed, Science Direct, Google Scholar, SID, and Cochrane until December 2021. The keywords were SARS-CoV-2, *Mycobacterium tuberculosis*, Confection. By searching this database; 84 articles were found, 51 of them by Reading titles and abstracts were removed. 33 articles were selected under the inclusion criteria. All articles were chosen from English and Persian articles.

Results: Finally, 33 articles were included in the study. Co-infections of SARS-CoV-2 with *M. tuberculosis* were likely to result within the generation of cytokines like TNF α and IL-6 that led to exacerbated pathogenicity. Patients with latent/active TB were at a more noteworthy hazard for severe COVID-19 and those analyzed with SARS-Cov-2, ought to be taken up for the improvement of TB. COVID-19 could be related with tuberculosis reactivation in patients with different predispositions, such as diabetes mellitus, failure renal, and steroid utilization. Mask wearing and the avoidance of unnecessary physical contact helped to avoid releasing infected droplets for both COVID-19 and TB.

Conclusion: TB-endemic districts may have exacerbated pathologies within the setting of SARS-CoV-2 diseases; something that the scientific community will ought to pay attention to. However, need to be more research done at this topic.

Keywords: *Mycobacterium tuberculosis*; SARS-CoV-2; COVID-19; co-infection

Isolation and Molecular Identification of *Acanthamoeba* from Contact Lens Users in Iran

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Background and Aim: Free-living amoebae (FLA) such as *Acanthamoeba* spp., are considered as opportunistic and pathogenic protozoans. *Acanthamoeba* keratitis is a serious threat for contact lens users. This study was carried out to identify the presence of *Acanthamoeba* among contact lens users in Qazvin, Iran.

Methods: Overall, 27 soft contact lenses were collected from keratitis patients. Following DNA extraction, PCR and sequencing were conducted to identify the genotypes of the amoeba. Phylogenetic analysis of the identified sequences was performed using MEGA 7 software.

Results: Two specimens (7.4%) of *Acanthamoeba* were isolated from keratitis patients. Both isolates of *Acanthamoeba* belonging to the T3 genotype.

Conclusion: Since the amoeba is present in environmental resources, the use of contact lenses should be accompanied by considering proper hygiene.

Keywords: *Acanthamoeba*; Genotypes; Contact lens; Iran

Molecular Identification of Intestinal Microsporidiosis in HIV+ Patients

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Background and Aim: Microsporidia small single-celled eukaryotic organisms infections that occur in virtually all invertebrate and vertebrate hosts as opportunistic pathogens have been recognized that associated with a wide range of clinical syndromes in persons with HIV/AIDS, travelers, children, and organ transplant recipients. The most common microsporidia infecting humans are *Enterocytoozon bienersi* and members of the *Encephalitozoon*. The purpose of this study was to characterization and genetical comparison isolate of *Enterocytoozon* spp. and *Encephalitozoon* spp. from persons HIV+ using ribosomal RNA gene.

Methods: In this study, a total of 150 HIV+ patients' fecal samples were collected from referring to behavioral disease counseling center of Karaj County and were screened for the existence of human-pathogenic microsporidia by staining method (modified trichrome) and PCR technique.

Results: No positive specimens were identified by microscopic examination and 5 (3.33%) specimens were identified with specific primers. In the molecular study, two isolates of *Enterocytoozon bienersi* (1.33%), 2 cases of *Encephalitozoon intestinalis* (1.33%), and 1 case of co-infections (0.7%) were identified. The ITS regions of the ribosomal RNA gene are used for genotyping of *Enterocytoozon bienersi*. The 3 sequences of *Enterocytoozon bienersi* were classified into 2 genotypes including D, J genotypes previously reported. Genotype D was identified in 2 HIV + patients. The J genotype was found from 1 sample. The 3 isolates of *Encephalitozoon intestinalis* identified in this study were similar to those found in other parts of Iran and the world and showed the least amount of genetic difference among these isolates.

Conclusion: Therefore, based on previous studies and animal isolates, the zoonotic nature of this species of *Encephalitozoon* has also been established. This study is hygienic importance because microsporidian pathogens have the potential to infect humans and, in turn, contaminate the environment around humans. This information is evidence of the importance of control, identification and prevention in medical centers.

Keywords: *Encephalitozoon intestinalis*; *Enterocytoozon bienersi*; ribosomal RNA gene; genotyping

New Methods of Laboratory Diagnosis of Malaria

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Background and Aim: Malaria is the most important human parasitic infection and causes high mortality in many tropical regions of the world. Malaria infection cannot be diagnosed by clinical examination due to the similarity of its clinical symptoms with other tropical infectious diseases, so it must be diagnosed and confirmed through a laboratory.

Methods: Laboratory tests for malaria include: 1- Preparation of thin and thick blood smear from the patient's fingertip and staining of blood spread and observation of malaria parasite with electron microscopy 2- Preparing an intradermal smear by making narrow holes on the forearm with a 25-gauge needle and placing its blood on a glass slide and staining and observing leukocytes containing malaria pigment with a light microscope. 3 - Molecular methods include polymerase chain reaction (PCR) to amplify a specific part of a selected region of the malaria genome and determine its genotype, as well as the use of PCR (SNP) to detect drug-resistant malaria and mixed infections. 4- Quantitative buffy coat method involves collecting blood by piercing a finger in a capillary tube containing orange acridine and anticoagulant and centrifuging the tube and observing the nucleus of the parasite in light green fluorescence and its cytoplasm in yellow-orange with Microscope equipped with a source of ultraviolet light 5 - Serological methods including indirect immunofluorescence (IFA) based on the use of specially prepared antigen attached on a slide that is kept at -30 ° C until the test and quantitatively both types of IgG and IgM antibodies in Examine the patient's.

Results: Malaria diagnosis by conventional microscopy is the gold standard method for malaria diagnosis that is cheap and reliable. Diagnostic tests are fast, expensive, but fast and easy. Serology is the best method used as an epidemiological tool and is not suitable for diagnosing acute malaria.

Conclusion: Molecular techniques are best studied in appropriate research laboratories for the spread of drug resistance and recurrence, and They can be useful for identifying species when the number of parasites is very low or in some specimens that are at risk of extinction. Diagnostic tests based on HRP II provide positive results in the recovery phase of the disease due to the persistence of HRP II in the blood after the parasite is cleared.

Keywords: malaria; laboratory diagnosis; molecular; serological; culture

Phenotypic and Genotypic Characterization of Antibiotic Resistance Among Methicillin-Resistant *Staphylococcus aureus* Isolates in Patients Admitted to Sarab and Tabriz Hospitals in 2020

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Background and Aim: The most common human infection, *Staphylococcus aureus*, has been a source of concern due to its rising resistance to antimicrobials. The goal of this study was to determine the antibiotic resistance patterns of methicillin-resistant *Staphylococcus aureus* isolated from patients' clinical specimens in Sarab and Tabriz hospitals.

Methods: This cross-sectional study included 190 patients in Sarab and Tabriz hospitals' ICU, surgery, heart, paediatrics, infectious, and emergency departments. According to CLSI recommendations, the resistance pattern of isolates was assessed using disc diffusion and D-test methods. Cefoxitin, cefazolin, erythromycin, clindamycin, linezolid, and trimethoprim-sulfamethoxazole were among the antibiotics utilized. A PCR assay was used to check for the presence of the femA and mecA genes in all isolates.

Results: The PCR technique was used to confirm all methicillin-resistant *Staphylococcus aureus* isolates utilising the femA and mecA genes. Cefoxitin, cefazolin linezolid, and trimethoprim-sulfamethoxazole resistance was found in 30, 18, 7, and 15 methicillin-resistant *Staphylococcus aureus* isolates, respectively. Antibiotic resistance phenotypic and genotypic characterization of methicillin-resistant *Staphylococcus aureus* isolates from patients admitted to Sarab and Tabriz hospitals. 10 isolates have a D-shaped clear zone around the CLSI disc proximal to the erythromycin disc, 19 isolates have a D-shaped zone around the CLSI disc proximal to the erythromycin disc, and tiny colonies have grown to CLSI in the otherwise clear zone.

Conclusion: Even last-resort medicines, such as linezolid, are becoming increasingly resistant in methicillin-resistant *Staphylococcus aureus* isolates, posing a major threat to human health and society.

Keywords: *Staphylococcus aureus*; Antibiotic resistance; D- test; femA; mecA

Phenotypic and Genotypic Characterization of Carbapenem-Resistant *Acinetobacter baumannii*

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Background and Aim: Antibiotic resistant *Acinetobacter baumannii* is one of the medical problems worldwide. In recent decades, carbapenem-resistant strains have emerged. One of the most common mechanisms of resistances is the production of carbapenemase enzyme, which in the present study we investigated their phenotypic and genotypic frequency in Dezful teaching hospital.

Methods: In this cross-sectional study, all strains of *Acinetobacter baumannii* were identified through biochemical and molecular methods. Antimicrobial susceptibility testing, Metalobetalactamase (MBLs) and extended spectrum betalactamases (ESBLs) were performed according to CLSI guidelines. Molecular detection of blaIMP, blaVIM, blaSPM, blaOXA-23, and blaOXA-24, blaOXA-58, blaGES genes were performed.

Results: A total of 54 strains were isolated from respiratory tract (68.5%), urine (16.6%), wound (11.1%), and blood (3.7%). The highest resistance rate was against third-generation cephalosporins (98.1%) and ciprofloxacin (94.2%), while about minocycline was 13%. *Acinetobacter baumannii* isolates exhibited 74% meropenem resistance. The most carbapenem-resistant strains were isolated from the endotracheal tubes. In phenotypic methods, 26% and 80% were ESBL and MBL producers, respectively. All of isolates were semi susceptibility against colistin (MIC of less than 2 µg/mL). In 35 strains of carbapenem resistance *A. baumannii* (CRAB) the most prevalent gene was blaOXA-23, followed by blaOXA-24, blaVIM, blaGES, blaIMP, and blaOXA-58 genes, but blaSPM gene not detected. These CRABs were classified into 11 patterns based on molecular detection. Six strains were positive for genes of class A, B and D and 18 strains were positive for B and D class genes.

Conclusion: Our report highlighted the existence of CRAB and semi-sensitivity to colistin and also co-existence of several genes from different carbapenemase classes in this region. Therefore, resistant strains should be identified promptly and specific treatment protocols should be devised to control the dissemination of resistance genes in therapeutic settings.

Keywords: *A. baumannii*; Carbapenem resistance; Carbapenemase; Genotyping

Possibility of *Brucella abortus* Bacteremia in Cattle Following RB51 Vaccination

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Background and Aim: *Brucella abortus* is a facultative intracellular Gram-negative bacterium of the Brucellaceae family, causative agent of bovine brucellosis. The disease has been reported worldwide and is one of the widespread bacterial zoonoses in the world, so called undulant fever (Malta fever) in humans. *Brucella abortus* is a major cause of abortion in cows. Vaccination is one of methods for controlling and preventing the disease. In Iran, recommended doses of IRIBA (RB51) vaccine full dose (FDIRIBA) and reduced dose (RDIRIBA) containing $10\text{-}34 \times 10^9$ CFU/ml and $1\text{-}3.4 \times 10^9$ CFU/ml bacterial cells per 2 ml, respectively. The vaccines may result in bacteremia in the vaccinated animals. The present study was carried out to investigate the possibility and duration of *Brucella abortus* bacteremia after vaccination in the naïve cows.

Methods: Fifty cows were randomly selected from 500 heads of imported Simmental fleckvieh adult cows with no history of brucellosis vaccination and/or infection, divided into two equal random groups (n = 25), and vaccinated with the IRIBA vaccines. The first group received a FDIRIBA dose and the second one was vaccinated with a RDIRIBA dose. Blood samples were taken on days 3, 7, 14, 21, 28, 35, 42, 49, 56 and 64 after vaccination followed by DNA extraction using a commercial kit. DNA samples were then subjected to a *Brucella* genus-specific PCR assay. The statistical comparison of the two groups was made using chi-square test, and the study of each group in different periods of the experiment was done by the Cochran method using SPSS 24 software.

Results: The results shows that there is a possibility of bacteremia after the vaccinations. It is revealed that not only the presence of *Brucella* bacterial sequences in the blood samples was significantly different for the time intervals (P-Value less than 0.001) but also a considerable difference was detected between the two groups as the bacteremia period in group 1 was significantly longer than that for group 2 (P-Value less than 0.001).

Conclusion: Based on the results, it seems that bacteremia a dose dependent phenomenon which caused earlier onset of bacteremia in FD group compared to the other RD, and higher likelihood of bacteremia occurrence in animals vaccinated with the full dose IRIBA rather than those who received the reduced dose vaccine. According to the data of this study, the minimum and maximum time periods for bacteremia occurrence were 3 and 35 days post vaccination, respectively; while the majority of blood samples were bacteremic on the fourteenth day.

Keywords: Cattle; *Brucella*; RB51; IRIBA; Vaccine; Bacteremia; PCR

Prevalence of *aac6-Ib-cr* Gene in Clinical Isolates of *Klebsiella pneumoniae* in Khorramabad by PCR Method

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Background and Aim: *Klebsiella pneumoniae* is one of the most important cases of community-acquired and hospital-acquired infections. This bacterium is one of the most common nosocomial pathogens. The increasing emergence of multidrug resistance among hospital isolates of *Klebsiella pneumoniae* has limited treatment options for treating infections caused by this bacterium. The aim of this study was to determine the frequency of *aac6-Ib-cr* gene in clinical isolates of *Klebsiella pneumoniae*.

Methods: In this study, during 7 months, people referring to laboratories in Khorramabad city were collected. The samples taken in the laboratory were evaluated differentially and biochemically and antibiotic susceptibility was evaluated by disk diffusion method. DNA extraction was performed by boiling method and genes were identified by specific primers using PCR technique.

Results: Out of 100 samples, 63 samples (63%) were male and 37 samples (37%) were female. The highest resistance was related to cefotaxime, amikacin, tetracycline and the highest sensitivity was related to: levofloxacin, ceftazidime, ampicillin, respectively. Of the total isolates, 57 isolates had resistance, of which the prevalence of *aac6-Ib-cr* gene was 49.1%.

Conclusion: The results of this study show that the prevalence of *aac6-Ib-cr* gene is high and it was found that there is an increase in resistance to fluoroquinolones in the isolates.

Keywords: *Klebsiella pneumoniae*; fluoroquinolone; antibiotic resistance; *aac6-Ib-cr*

Prevalence of COVID-19 Among People Living with HIV: A Systematic Review and Meta-Analysis

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Background and Aim: Whether human immunodeficiency virus (HIV) infection is associated with a higher risk of morbidity and mortality due to corona virus disease 2019 (COVID-19) is unclear. In the present study, we aimed to provide a systematic review and meta-analysis of HIV among patients with COVID-19 through case reports/case series and prevalence studies.

Methods: A systematic literature search strategy was conducted via reviewing original research articles published in Medline, Web of Science, and Embase databases in 2019 and 2020. Statistical analysis was performed using STATA software, version 14.0 (Stata Corporation, College Station, Texas, USA) to report the prevalence of HIV among patients with COVID-19. Case reports/case series were also evaluated as a systematic review.

Results: Sixty-three studies (53 case reports/case series and 10 prevalence studies) were included in our study. A meta-analysis of prevalence studies showed that HIV infection among patients with COVID-19 were reported in 6 countries (Uganda, China, Iran, USA, Italy and Spain) with an overall frequency of 1.2 % [(95% CI) 0.8-1.7] among 14424 COVID-19 patients. Conforming to the results of the case reports/case series, 111 patients with HIV have been reported among 113 patients with COVID-19 from 19 countries. Most of the cases were in the USA, China, Italy, and Spain.

Conclusion: People with HIV are more likely to develop more severe complications from COVID-19. Targeted policies should be considered to address this raised risk in the current pandemic. Our findings highlight the importance of identifying underlying diseases, co-infections, co-morbidities, laboratory findings, and beneficial treatment strategies for HIV patients during the COVID-19 pandemic.

Keywords: HIV; COVID-19; Coronavirus; meta-analysis

Prevalence of *Enterococcus faecalis* Among Women with UTI in Reproductive Ages

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Background and Aim: Bacteriuria is frequent among pregnant women and is associated with significant fetal and maternal risks. *Enterococcus faecalis* is one of the opportunistic pathogens of urinary tract infections (UTIs) among women causing life threatening infections in preterm infants. Therefore, we aimed to assess *E. faecalis* among women with UTIs in reproductive ages.

Methods: Five hundred unknown bacterial isolates were collected from women with UTIs in reproductive ages (15-49 years old) from Emam-Reza hospital, Tabriz, Iran. The bacterial isolates were detected by routine biochemical test and antibiotic resistance of detected *E. faecalis* isolates were investigated by phenotypic and genotypic methods.

Results: Totally, 15 (3%) *E. faecalis* isolates were detected by phenotypic tests, which 26.7% of isolates were belonged to women in 15-30 years old and 73.4% of isolates were belonged to 31-49 years old. Antibacterial susceptibility test showed that 76.3% and 46.7% isolates were resistant to tetracycline and erythromycin and all isolates were susceptible to linezolid and teicoplanin. The highest presence of antibiotic resistance genes was related to *aadE* (80%) and *tetM* (93.3%) and the lowest presence was related to *vanB*, which was not found in any of the isolates.

Conclusion: Women in the 31–49 years old group were at a higher risk of UTIs. Most UTI isolates demonstrated phenotypes and genotypes resistant to a variety of antibiotics such as vancomycin, aminoglycosides, erythromycin, and tetracycline. In multidrug- and vancomycin-resistant isolates, linezolid may be an effective and selective drug in the treatment of enterococcal UTIs among women in reproductive ages.

Keywords: *Enterococcus faecalis*; Urinary tract infections (UTIs); reproductive ages

Prevalence of *Helicobacter pylori* Infection in Patients with Non-Alcoholic Fatty Liver Disease

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Background and Aim: The risk of *Helicobacter pylori* infection in people with fatty liver is important. Identify common factors that are associated with this disease, are different in patients due to gastrointestinal disorders. The aim of this study was to evaluate the prevalence of *Helicobacter pylori* infection in patients with non-alcoholic fatty liver disease (NAFLD) and its associated factors in NAFLD patients.

Methods: This study is based on patient records in the archives of Velayat Hospital, Qazvin. The records of patients with NAFLD with *Helicobacter pylori* infection from March 2019 to September 2020 were reviewed. Patients with suspected NAFLD who had gastrointestinal upset and abdominal obesity and required ultrasound were selected by sampling method with continuous patient referral. Patients who had a clinical diagnosis of *Helicobacter pylori* infection were evaluated by examining *Helicobacter pylori* antigens in a stool sample.

Results: One hundred and thirty patient records were reviewed with NAFLD. Among them, 40 (30.76%) patients were infected with *Helicobacter pylori*. Changes in level of aspartate aminotransferase, interleukin 6, bilirubin level, also severity of liver lesions with *Helicobacter pylori* infection were directly related to the severity of infection (p-value less than 0.05). There was no significant relationship between patient genus and disease severity and changes in laboratory parameters (p-value more than 0.05).

Conclusion: The prevalence of *Helicobacter pylori* infection was seen in patients with NAFLD. A possible link between *Helicobacter pylori* infection and NAFLD has been shown in this study. It is necessary to examine people with NAFLD for the presence of infection and to prescribe appropriate medications.

Keywords: *Helicobacter pylori*; Infection; Non-alcoholic fatty liver disease

Prevalence of *Salmonella* Diarrhea in Holstein Calves in Golestan Province and Determine Its Antibiotic Resistance Pattern

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Background and Aim: Neonatal calf diarrhea remains one of the most important problems faced by livestock, causing great economic losses. This study investigated the prevalence of *Salmonella* spp. in diarrheic calves.

Methods: Fecal and rectal samples were collected from 57 diarrheic calves up to 3 months of age from Livestock farms in Golestan province was collected in 2020. Stool samples from each calf were transferred to the microbiology laboratory in enrichment media (Selenite F Broth) and the necessary bacteriological tests were performed on them to isolate *Salmonella* spp. For data analysis, SPSS 22 and frequency distribution table were used (P-value less than 0.05).

Results: Based on bacterial culture results, the prevalence's of *Salmonella* spp. in diarrheic calves was 37%. Results showed 37% of diarrheic calves were male and 33% was female, so there was no significant difference (P-value higher than 0.05) between male and female diarrheic calves. The in-vitro antibiotic sensitivity test indicated that *Salmonella* spp. showed high sensitivity against ceftriaxone (62%), gentamicin (57%) and amoxicillin (30%). *Salmonella* spp. isolates was resistant to tetracycline (100%) and cefazolin (95%).

Conclusion: Results of the present study showed that *Salmonella* spp. isolated from diarrheic calves in farms exhibited resistance to multiple classes of antimicrobials, which may pose a public health hazard.

Keywords: Diarrhea; Neonatal calf; Fecal and rectal samples; *Salmonella* spp; Antibiotic resistance

Prevalence Survey of *Strongyloides stercoralis* in Fecal Samples of Patients

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Background and Aim: *Strongyloides stercoralis* is a globally distributed human intestinal nematode. To prevent and treat these diseases, information on the distribution and frequency of these parasites in different geographical areas is needed. Considering the importance of parasitic diseases, especially strongyloidiasis in the Northern provinces, the present study was conducted to determine the prevalence of *Strongyloides stercoralis* in the spiritual hospital of Mazandaran province in Babol city.

Methods: From April to March 2017, stool samples were collected from people referring to Rouhani Hospital. Microscopic detection was performed immediately using the wet expansion method, followed by a formal-ether concentration process for *Strongyloides stercoralis* detection. Analysis of variance (ANOVA), paired samples t-test and Chi-square were used for statistical analysis of data WITH SPSS 21 (P-value less than 0.05).

Results: The prevalence of parasitic infection in urban and rural patients was 725 (44.18%) and 916 (55.81%), respectively. The prevalence of *Strongyloides stercoralis* was observed in 39 patients, of which 20 were women and 19 men. There were also 17 infected patients from the city, and 22 patients from the rural areas. There was a significant relationship between the patient's location and the prevalence of the parasite. People in rural areas were more infected (P-value less than 0.05).

Conclusion: This study showed that *Strongyloides stercoralis* have a high prevalence in patients in rural region. Given the potential risks of these infections, it is recommended that regular screening be performed to diagnose and treat parasitic diseases in this population.

Keywords: *Strongyloides stercoralis*; fecal; patients

Productions of Solubilized Human Recombinant Interleukin 11 SHuffle T7 Strain

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Background and Aim: Human interleukin 11 (hIL-11) is a pleiotropic cytokine secreted by bone marrow stromal cells or mesenchymal cells. hIL-11 plays important roles in numerous biological regulatory processes such as the hematopoietic system, and is anti-inflammatory and anti-apoptotic. The rhIL-11 has been shown to increase platelet counts in animals and humans and is the only drug approved for application in chemotherapy-induced thrombocytopenia (CIT). Oprelvekin, the active ingredient in Neumega®, is a rhIL-11 product, which is produced in *Escherichia coli* (*E. coli*) by recombinant DNA technology. With a molecular mass of approximately 19,000 daltons for the non-glycosylated rhIL-11, the protein is composed of 177 amino acids in length in comparison to the natural hIL-11 with 178 amino acids. However, the recombinant form displays a biological activity similar to the natural form both in vitro and in vivo. Oprelvekin as a drug works by stimulating megakaryocytopoiesis and thrombopoiesis. In mice and nonhuman primate studies of animals with moderate and severe myelosuppression, in addition to compromised hematopoiesis, oprelvekin was shown to potently induce thrombopoiesis and improve the platelet nadirs, and accelerated platelet recoveries compared to the controls. In animal studies, oprelvekin was also shown to regulate intestinal epithelium growth by enhancing healing of gastrointestinal lesions.

Methods: In this study, the CDS of human IL-11 with a His-tag in the structure of pGH plasmid was digested with suitable restriction enzymes and was sub-cloned into pET15b contained the expression cassette of thioredoxin (pET15b/Trx/hIL-11). Finally, the plasmid was transformed into SHuffle bacteria, which cultured in LB medium containing ampicillin. The positive colonies that contained pET15b/Trx/hIL-11 was selected. Successful expression of the soluble rhIL-11 protein in LB medium was achieved by IPTG induction when the OD600 was approximately 0.8.

Results: Induction of recombinant protein production was performed using 0.1 mM IPTG at 30 °C for 6 h. After this time period, the cells were harvested and the expression of the recombinant protein was evaluated using 15% sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). Eventually, rhIL-11 protein was purified with Ni-affinity resin, and analyzed again with 15% SDS-PAGE.

Conclusion: In the present study, a new subspecies of Shuffle *E. coli* was obtained by transformation of an expression plasmid, pET15b/Trx/hIL-11, which was efficiently produced the soluble rhIL-11. Production of soluble recombinant hIL-11 was verified through SDS-PAGE analysis. Moreover, the efficient purification of rhIL-11 with Ni-affinity agarose was confirmed by SDS-PAGE in which a



protein band was observed with the size of about 24 kDa in comparison to the negative control contained a sample of the extracted protein from the *E. coli* cells harboring intact pET15b vector.

Keywords: Human interleukin 11 (hIL-11); *E. coli*; recombinant vector



SARS Cov-2 : Communication of Genetic, Structure & Infection

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Background and Aim: The pandemic of the novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has been posing great threats to the world in many aspects. Although some possible antiviral and antiinflammatory medications are available, thousands of people are dying daily. Now we are understand genome of SARS cov-2 is not stable and there is many mutation and diversity in that ; for this reason the characteristics and features of coronavirus are so variable. This narrative, non-systematic review provides an update on the genetic aspects of the SARS CoV-2 virus and its interactions with the human genome within the context of COVID-19.

Methods: We performed an online search on PubMed and Google Scholar and other Web of Science with the keywords: COVID-19, 2019-nCoV, SARS-CoV-2, SARS-CoV-2-diversity, SARS-cov-2-genetic, and summarized the epidemiology, virology and genetic diversity and mutation of SARS-CoV-2 and COVID-19 infection.

Results: We retrieved 23 published papers about SARS-CoV-2 from January, 2021 to June, 2021. We found that SARS CoV-2 was a kind of virus with low mortality rate and high infectivity. We summarized consequences mutations and diversity of SARS Cov-2's genome, such as peplomer or spike diversity, effect on enter in human cell and Identify & develop immune system responses.

Conclusion: SARS-CoV-2 is a novel coronavirus that has caused a global pandemic. We should pay more attention to prevent SARSCoV-2 and try to control it but we must never forget the diversity and mutations of this virus.

Keywords: SARS-Cov-2-genetic; SARS-Cov-2-diversity; severe-acute-respiratory-syndrome; coronavirus; Covid-19-infection

Sero-Epidemiological Study of *Helicobacter pylori* Infection and Its Association with Metabolic Syndrome

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Background and Aim: *Helicobacter pylori* is one of the most common bacterial infections in the world. On the other hand, Obesity and Metabolic Syndrome (MetS) are major risk factors for type 2 diabetes and cardiovascular disease. In addition, there is not enough information about the role of *Helicobacter pylori* infection on the metabolic syndrome and its relationship. In this study, the relationship between *Helicobacter pylori* infection and the incidence of metabolic syndrome is investigated.

Methods: In this study, 100 eligible clients of Farhangian Clinic in District 18 of Tehran were evaluated for age, sex, height, weight (BMI) and history of the disease. The mean age of women was 46 and men 44 years old. IgG titer of *Helicobacter pylori* antigen of ELISA technicians was evaluated in Paython ELISA fully automatic device and the indicators of metabolic syndrome, including sugar and their lipid profile were evaluated by the relevant kits in E-Selectra device and finally various information and factors by software. SPSS version 16 was statistically analyzed.

Results: The frequency of *Helicobacter pylori* anti-IgG antigen was positive in 92%, interstitial in 2% and negative in 6%. In fact, measurement of metabolic indices in metabolic syndrome shows that *Helicobacter pylori* infection does not cause any statistically significant change

Conclusion: Therefore, there is no association between *Helicobacter pylori* infection and the incidence of metabolic syndrome.

Keywords: *Helicobacter pylori*; Seroepidemiology; Metabolic syndrome; Lipid profile

Simultaneous Identification of *Mycoplasma* and *Ureaplasma* Species in Fertile and Infertile Women by Real-Time PCR Assay and Melt Curve Analysis

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Background and Aim: *Mycoplasma* species threaten reproduction and fertility worldwide. The lack of a sensitive and accurate diagnostic method has led to the neglect of the role of these bacteria in infertility. The aim of this study was to identify and compare the frequency of *Mycoplasma* isolates in fertile and infertile women using Real-Time PCR and melting curve analysis.

Methods: In this case-control analytical study, cervicovaginal swap samples were taken from 140 patients referred to two teaching hospitals in Isfahan. Using a new primer designed to amplify 16S rRNA gene and real-time PCR technique, several types of *Mycoplasma* and *Ureaplasma* in two fertile (n = 70) and infertile (n = 70) groups were examined.

Results: The frequency of *Mycoplasma* isolates in infertile and fertile groups was as follows respectively: *Mycoplasma hominis* 17.4% (n = 12), 7.1% (n = 5), *Mycoplasma genitalium*: 2.8% (n = 2), 0 % (n=0) *ureaplasma parvum* 17.4% (n = 12), 5.7% (n=4), *Ureaplasma urealiticum* 10% (n = 7), 4.2% (n = 3), coinfection 22.8% (n = 16%) and 2.8% (n = 2). The designed primer made it possible to simultaneously identify *Mycoplasma* species in cervicovaginal specimens.

Conclusion: In the present study, the frequency of *Mycoplasma* and *Ureaplasma* species in the infertile group was more than 2 times that of the fertile group. Therefore, it is important to pay attention to the role of these infectious agents in infertility.

Keywords: *Mycoplasma* spp; *Ureaplasma* spp; infertility; Real-time PC

Study of Prevalence of Bacterial Infection in Patients with Covid-19 Disease Admitted to Intensive Care Units

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Background and Aim: Although Covid-19 is a viral infection but simultaneous bacterial infection can result in a more severe infection especially in hospitalized patients. This Co-infection can complicate the treatment process and increase mortality in these patients. To date, many studies have been conducted on SARS-CoV-2 infection, but studies regarding the bacterial co-infection of SARS-CoV-2 are scarce.

Methods: Three hundred and four blood, sputum, and urine samples were collected from patients with Covid 19 disease who were hospitalized in the intensive care unit of Rasool Akram Hospital for six months. All samples were evaluated with microscopic and biochemical tests. Then they were identified by specific media. The collected data were analyzed by SPSS-26 software.

Results: Out of 304 patients with Covid 19 disease who were admitted to the intensive care unit, 178 (58.6%) were male and 126 (41.4%) were female. 138 patients had a positive culture. The highest prevalence of infection was seen with *Klebsiella pneumonia* (42) 30.45%, followed by *Acinetobacter* (30) 21.74% and *Staphylococcus aureus* (22) 15.95%. The lowest rate of infection was reported with *Citrobacter* (1) 0.7%.

Conclusion: Our study result showed that *Klebsiella pneumonia*, *Acinetobacter* and *Staphylococcus aureus* are the most common pathogens that cause secondary infections in Covid-19 patients. Although these pathogens may be different in other medical facilities but identification of these agents can be vital in reducing the Covid-19 infection mortality.

Keywords: Covid-19 disease; Bacterial infection; Hospital

Study of Prevalence of *optrA* in *Enterococcus* Isolates Collected from Hospital Infection

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Background and Aim: Enterococci are one of the opportunist bacteria which has important role in hospital infection. Nowadays antibiotic resistance is expanding. Gene transfer from one bacterium to another can be very challengeable. Most resistant in *Enterococcus* bacteria occurs through gene transfer on the plasmid. One of the resistance genes in *Enterococcus* is *optrA*. The aim of this study is to evaluate the prevalence of *optrA* in *Enterococcus* isolates collected from hospital infection.

Methods: eighty-seven isolates of *Enterococcus* were collected from different ward of Rasool Akram hospital Tehran, Iran. Microbiological and biochemical tests were confirmed to identify the isolates. Then the antibiogram test was performed for all of the isolates. The isolates which were phenotypic resistance to linezolid antibiotic were selected. Subsequently detection of the *optrA* gene was accomplished by PCR amplification.

Results: Among these isolates, 57 isolates were phenotypic resistance to Linezolid antibiotic. Molecular characterization by PCR amplification showed 55% isolates carried *optrA*.

Conclusion: Nowadays drug resistant in *Enterococcus* isolates has increased and the concerns of the medical staff in this regard are rising. Therefore evaluate and monitor of resistant genes in every ward of and every hospital is essential.

Keywords: *Enterococcus*; *optrA*; hospital infection

Study of Relationship Between Anemia and Thrombocytopenia with *Helicobacter pylori* Infection

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Background and Aim: *Helicobacter pylori* (*H. pylori*) infection is one of the most common infections worldwide that can affect children and adults. Blood disorders associated with this infection include diseases such as iron deficiency anemia, immune thrombocytopenia and vitamin B12 deficiency and maltoma. Therefore, this study aimed to determine the relationship between anemia and thrombocytopenia with *H. pylori* infection.

Methods: In this cross-sectional study, 438 people suspected of being infected with *H. pylori* infection were examined by requesting an Anti-H. Pylori IgG test referred to medical diagnostic laboratories in Gonabad. The CBC test was performed and data were analyzed by SPSS software version 26.

Results: In this study, 438 patients were included, of which 150 (34.2%) were male and 288 (65.8%) were female. The mean age of patients was 41.64 ± 16.87 years (male 40.16 ± 9.19 years and female 42.17 ± 0.23 years). The prevalence of anemia and thrombocytopenia in individuals infected with *H. pylori* in this study was 7.24% and 3.2%, respectively, and there was no significant difference with the non-infected group. There is a statistically significant association between mean age and *H. pylori* infection ($P=0.001$). Based on antibody titer analysis, 186 cases (42.5%) had a positive titer (more than 12u/ml). In addition, there was no significant difference in CBC test between the levels of *H. Pylori* antibody titers (P greater than or equal 0.05).

Conclusion: No significant differences were observed between blood groups of infected and non-infected patients. Therefore, biochemical examinations along with hematological parameters are recommended.

Keywords: *Helicobacter pylori*; Anemia; Thrombocytopenia

Superinfections Caused by *Acinetobacter baumannii* and *Pseudomonas aeruginosa* Among Hospitalized COVID-19 Patients Admitted to ICU

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Background and Aim: Hospitalization due to the invasive manifestation of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) may lead to the emergence of superinfections. We aimed to describe the prevalence of hospital-acquired *Acinetobacter baumannii* and *Pseudomonas aeruginosa* and their pattern of antibiotic resistance among the COVID-19 inpatients admitted to ICU during the summer of 2021 in Ghaem hospital, Mashhad, Iran.

Methods: This retrospective cohort study was performed in all inpatients who had positive SARS-CoV-2 reverse transcription PCR (RT-PCR) in nasal or respiratory tract samples during July, August, and September 2021. Therefore, data of eligible inpatients were investigated by the virtue of electronic health records. The reported bacteria which were diagnosed ≥ 48 hours after admission were included in our study. The obtained outcomes were analyzed by SPSS software version 22.

Results: Eight hundred and ninety-four Hospitalized patients were qualified for our study of which 45 (5.03%) *Acinetobacter baumannii* and 24 (2.68%) *Pseudomonas aeruginosa* were isolated as superinfection. The mean (SD) time from admission to diagnosis of superinfection was 12 (16.2) days. The most common sources of collected bacteria were respiratory samples (22 (48.8%) *Acinetobacter baumannii* and 14 (58.3%) *Pseudomonas aeruginosa*) and blood culture (17 (37.7%) *Acinetobacter baumannii* and 8 (33.3%) *Pseudomonas aeruginosa*). Imipenem (21, 87.5%), meropenem (15, 62.5%) and amikacin (12, 50%) were the antibiotics displaying the highest resistance frequency for *Pseudomonas aeruginosa*. besides, all isolates of *Acinetobacter baumannii* were resistant against imipenem, meropenem and piperacillin-tazobactam. Doxycycline (26, 57.7%) and piperacillin-tazobactam (12, 50%) showed the lowest frequency of resistance among *Acinetobacter baumannii* and *Pseudomonas aeruginosa*, respectively. Crude mortality rate in *Acinetobacter baumannii* and *Pseudomonas aeruginosa* superinfected patients concluded as 13 (28.8%) and 6 (25%), respectively.

Conclusion: Our findings emphasized the prevalence of two important hospital-acquired superinfections. Promoting appropriate antibiotic usage as a consequence of antimicrobial stewardship strategies has a critical role in reviving inpatients.

Keywords: COVID-19; Superinfection; *Acinetobacter baumannii*; *Pseudomonas aeruginosa*

The Effect of Polysaccharide Capsule Genes in Bacterial Colonization in Patients with Colorectal Cancer

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Background and Aim: The third main cancer in Iran following the breast and prostate cancer is colorectal cancer (CRC), which is the fourth common cancer in the world. One of the most important risk factors of this cancer is the bacteria infection and the corresponding toxins. It has been acknowledged that several virulence capsule genes have great influence on the pathogenicity of the bacteria. The aim of this study is the evaluation of effective role played of polysaccharide capsule genes in *E.coli* isolated from patients with Colorectal cancer.

Methods: In this study, the biopsies obtained from patients with CRC and inflammatory bowel disease and normal individuals who referred to the Imam khomeini hospital in Tehran. All of participants filled the consent form and questionnaire. This project approved by ethical committee in Imam Khomeini Hospital. Then we cultured the biopsies in LB buffer to grow bacteria. By using the microbial and biochemical methods as a control group, 70 *E.coli* were isolated from the cultured biopsies from 70 out of 98 patients. In the next step, the existence of virulence genes like polysaccharide capsule synthesis Genes was assessed applying PCR.

Results: The results showed that among of isolated *E.coli* bacteria, 85.7% was associated with the colibactin polyketide synthesis system (PKS island), 100% had the Kps MTI and 71.4% had the Kps MTIII.

Conclusion: In summary, we have acknowledged the prevalence of the bacteria (*E.coli* containing Kps MTI and III) has a major role in the inflammation and the incidence of the CRC.

Keywords: *Escherichia coli*; polysaccharide capsule genes; colorectal cancer

The Effect of Probiotic Bacteria and Prebiotics on the Growth of *E.coli* and *Enterococcus faecalis*

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Background and Aim: A large and diversified community of microorganisms, including a variety of anaerobic bacteria, live in the intestine. These bacteria participate in a wide spectrum of compound catabolism and produce secondary hazardous substances. The host is frequently poisoned by these secondary chemicals. The amount and type of gut flora can be balanced by consuming probiotic bacteria and prebiotics in the diet.

Methods: In this study, 100 microliters of 0.5 McFarland concentration overnight probiotic bacteria grown in MRS broth (*Lactobacillus acidophilus* ATCC 4356, *Lactobacillus casei* ATCC 39392, *Lactobacillus fermentum* ATCC 9338, *Lactobacillus plantarum* ATCC 8014, *Lactobacillus reuteri* ATCC 2372, *Lactobacillus rhamnosus* ATCC 7469) were incubated in three. After 48 hours, all cultures were centrifuged at 6000 rpm and -4 degrees Celsius for 20 minutes to extract supernatant of probiotic bacteria, and the supernatant was passed through a filter 0/22 micron to determine its effect on the growth of *E.coli* ATCC23922 and *Enterococcus faecalis* ATCC 29212. (minimum bactericidal concentration).

Results: The MIC of free supernatant of *L. casei* grown in MRS broth containing 2 percent w/v trehalose on *E. coli* and *L. casei* grown in MRS broth containing 2 percent w/v sorbitol and *L. casei* grown in control culture medium on *Enterococcus faecalis* was 14/7 percent and 14/7 percent, respectively.

Conclusion: To modulate intestinal flora, we can utilize *L. casei* supernatant with Trehalose and Sorbitol as a dietary supplement

Keywords: Probiotic; Prebiotic; MIC-MBC; Intestinal flora

The Effects of Microencapsulation of *Lactobacillus acidophilus* and *Bifidobacterium bifidum* by Alginate and Arabic Gum in Canned Carrot Jam

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Background and Aim: Microencapsulation technique with various hydrocolloidal coatings is used to improve viability potential of probiotics in processed food. As a probiotic product, canned jam should contain a sufficient number of viable probiotic bacteria.

Methods: In this study, we evaluated the protective effects of Arabic gum and alginate on the survival of two important probiotic bacteria, *Lactobacillus acidophilus* DSM 1643 and *Bifidobacterium bifidum* 1644. Microencapsulation of these two probiotic bacteria was carried out by alginate and Arabic gum, and their impacts on the survivability and the sensory characteristics of canned carrot jam were examined for one month.

Results: According to our results, although the reduction in the number of probiotics in canned jam at different time was significant, this product was able to retain a good number of probiotic bacteria. There was a significant difference between the dropped number of coated and free bacteria. However, No significant difference was observed in the survival rate of probiotics coated by Arabic Gum and alginate. Moreover, the addition of free and encapsulated probiotics had no significant impact on texture, color and flavour of the final product during the maintenance time. The probiotic survival can be enhanced by cell protection through encapsulation.

Conclusion: We compared the survivability of free bacteria with those encapsulated by Arabic gum and alginate, and showed that microencapsulation significantly improved the survival of *Lactobacillus acidophilus* and *Bifidobacterium bifidum* in canned jam for a month. However, there was no significant difference between survivability of probiotic bacteria encapsulated by Arabic gum and alginate. Also, the results of sensory evaluation indicated that microencapsulation does not affect the sensory properties of probiotic jam. Hence, it may be possible to apply these results to other products and safely use the benefits of probiotic bacteria.

Keywords: probiotic bacteria; microencapsulation; Arabic gum; alginate; canned carrot jam

The Genotyping of *Escherichia coli* O157:H7 Isolated from Slaughterhouses and Water in Different Regions of Fars Province Using Random Amplified Polymorphic DNA (RAPD) Analysis and Their Determining of Antibiotic Resistance.

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Background and Aim: *Escherichia coli* O157:H7 one of the enterohemorrhagic *E.coli* serotypes is main food-borne pathogen that can cause severe gastrointestinal and death in human. Cattle are the major reservoir for *E. coli* O157:H7. During the slaughtering and production process *E. coli* O157:H7 enter to food chain. Identifying the source of contamination plays an important role in the control of infections. The aim of this study was to determine antibiotic susceptibility and fingerprinting of *Escherichia coli* O157:H7 isolated from slaughterhouses in different region of Fars province.

Methods: Twenty-three *Escherichia coli* O157:H7 are isolated. Random amplified polymorphic DNA (RAPD) analysis with the short primer of 1283 was used to evaluate genetic relationship among the isolates and the results was analyzed by NTSYSpc software. Antibiotic susceptibility of the isolates was examined through 7 antibiotics using the Kirby-Bauer disc diffusion method.

Results: most isolates revealed sensitivity to seven the antibiotics and resistance rates were observed in only 3 strains with patterns ToB 10 (R)LP(R)AZM15(R) in Strain ECoRsed21, IPM10(R) in ECoRsed18 and ECoRsed22 and ToB 10 (R)LP(R) AZM 15(R) in strain ECoRsed20. DNA fingerprinting showed 9 genetic patterns for 23 strains by RAPD-PCR technique. The isolates from different sources were clustered from 31% to 100% based on similarity in RAPD pattern. The strains contain different RAPD pattern or source and had high genetic distance to others, showed different patterns of antibiotic susceptibility.

Conclusion: Isolates from different sources had the same genetic pattern and were located in the same cluster. Therefore, genotyping of isolates using RAPD technique is a fast, cost-effective and efficient method to determine the source of infection and its control.

Keywords: Genotyping; Antibiotic Resistance; *Escherichia coli* O157:H7; RAPD PCR

The Highly Conserved Domain of RND Multidrug Efflux Pumps in Pathogenic Gram-Negative Bacteria

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Background and Aim: RND (Resistance-Nodulation-Division) family transporters have a vital role in both intrinsic and acquired multi-drug resistance in Gram-negative bacteria. It is important to find a conserved domain in the RND family between different pathogenic bacteria for diagnostic and therapeutic purpose.

Methods: Total sequences of three-component system RND efflux pumps were retrieved from NCBI nucleotide and protein database and were subjected to conservation and variation analysis using the multiple sequence alignment feature of the CLC workbench. The phylogenetic tree for main transporters was drawn and the three-dimensional structure was also evaluated.

Results: From the sequence conservation analysis, highly conserved residues with 282 base pair (94 amino acid) long were identified. The location of the highly conserved domain is positioned in the domain 1 crystallographic structure of AcrB *Escherichia coli* and MexB *Pseudomonas aeruginosa*. The main transporter component phylogenetic tree shows the clusters of different genotypes and their evolutionary association. Each of three components of RND proteins is crucial for drug efflux, and the absence of even one component makes the entire complex totally nonfunctional.

Conclusion: Therefore, this highly conserved region can be used to disable the RND multidrug efflux pumps. In addition, this highly conserved can also be used for diagnostic aspects.

Keywords: RND multidrug efflux pumps; Conserved domain; Gram-negative bacteria

The Relationship Between *Helicobacter pylori* Infection and Immunological Indicators of TNF- α , CRP, and Fibrinogen of Patients in the West of Tehran

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Background and Aim: *Helicobacter pylori* infection is the most prevalent infection of the world. There is little information about the possible role of *Helicobacter pylori* infection on serum levels Tumor necrosis factor- alpha (TNF- α), a cellular cytokine that is classified as a cell-signaling protein, Reactive protein C (CRP), an inflammatory marker, and fibrinogen, a acute phase reactant. This study evaluated the seroprevalence of *H. pylori* infection on serum levels of TNF- α , CRP and fibrinogen in patients referred to a laboratory in the west of Tehran province.

Methods: In this cross-sectional study of the blood of 50 people who referred to clinical lab in the West of Tehran were collected. The titer of total IgG of *Helicobacter pylori* was evaluated by anti-*Helicobacter pylori* kit (IgG). Then people with consistently positive or negative results were selected to participate age, sex were recorded. In addition, the determination of serum concentration of total TNF- α was done by ELISA technique. In order to determine the quality of CRP in the serum of the subjects, agglutination method was used. Finally, In order to quantify the amount of fibrinogen in the plasma of the subject, the coagulation method (GLAUSS kit) was used. Data and different results were analyzed by SPSS statistical software.

Results: The Results showed that among the three conditions studied for *Helicobacter pylori* infection; 32% of the subjects were positive, 54% of the subjects were negative and 14% of the subjects were in the intermediate position and among them, the number of patients was higher in women than men. The serum levels of TNF- α , of CRP and fibrinogen were normal level in volunteers

Conclusion: The results also showed that there is no significant relationship between TNF- α , CRP, Fibrinogen levels in patients and *Helicobacter pylori* infection. Further studies are necessary for the detection of action mechanism of *H. pylori* on proinflammatory factors

Keywords: *Helicobacter pylori*; proinflammatory factors; prevalence

The Role of the Lung-Gut Axis Microbiota in Covid 19

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Background and Aim: Respiratory viral infections disrupt the gut microbiota. Basal enrichment of *Coprobacillus*, *Clostridium ramusum* and *Clostridium hati* was associated with Covid-19 intensity. There was an inverse correlation between the frequency of *Faecalibacterium prausnitzii* and the severity of the disease. Therefore, improving the intestinal microbiota profile may be a preventive measure to minimize the effect of COVID-19 in elderly and immunocompromised patients.

Methods: We used reputable scientific sites such as PubMed, sciencedirect to obtain update information on the role of the gut-lung axis microbiome in treatment on covid 19.

Results: SARS-CoV-2 affects the respiratory and gastrointestinal tracts, and symptoms such as watery diarrhea are associated with long-term illness and virus transmission in COVID-19. There is a possible association between gastrointestinal symptoms and the severity of COVID-19. The main meanings for predicting the course of the disease and establishing targeted gastrointestinal therapies that may affect the severity of the disease.

Conclusion: Global research on intestinal microbiota has increased our knowledge of chronic and infectious diseases and is useful in meeting the challenges of managing COVID-19 patients with related gastrointestinal diseases. Therefore, different modulation approaches to deform the intestinal microbiota may minimize severity. It is an infection and may be a treatment for COVID-19 comorbidities.

Keywords: SARS-CoV-2; gut-lung axis microbiome

***Thymus daenensis* and *Satureja hortensis* Essential Oils as New Antimicrobial Agent Against Food-Borne Pathogen *E. coli* O157:H7**

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Background and Aim: Enterohemorrhagic *Escherichia coli* O157:H7 (EHEC) causes foodborne outbreaks worldwide and the bacteria form antimicrobial-tolerant biofilm. We investigated the abilities of *Thymus daenensis* and *Satureja hortensis* essential oils (EOs) to inhibit bacterial growth, biofilm formation and quorum sensing (QS) by *E. coli* O157:H7.

Methods: The tested EOs were isolated from plant material by hydrodistillation and analyzed under chromatography-mass spectrometry. The antibacterial and anti-biofilm activities of the EOs were determined by microdilution broth and microtiter-plate (MtP) tests, respectively. The QS inhibitory (QSI) potential was examined by inhibition of swimming and swarming motility at sub-MIC levels. Real-time quantitative reverse transcription PCR (RT-qPCR) was used to determine the expression of QS-system-related genes.

Results: The MICs for *T. daenensis* and *S. hortensis* EOs against EHEC were 3.12 and 6.25 µg/mL, respectively and the MBCs were 6.25 and 12.5 µg/mL, respectively. The MtP test showed a significant ($p < 0.05$) inhibitory and disruptive effect on both EOs for EHEC biofilm formation at MIC/2 (1.56 µg/mL for *T. daenensis*; 3.12 µg/mL for *S. hortensis*) and MIC/4 (0.78 µg/mL for *T. daenensis*; 1.56 µg/mL for *S. hortensis*) concentrations. Gene expression analysis revealed significant down-regulation of *luxS* and *pfs* following treatment with MIC/2 concentrations.

Conclusion: The results of the present research point to the promising antibacterial, anti-biofilm and anti-QS potential of *T. daenensis* and *S. hortensis* EOs against *E. coli* O157:H7.

Keywords: *Escherichia coli* O157:H7; Biofilm; Quorum sensing; *Thymus daenensis*; *Satureja hortensis*

Utilizing Probiotic Lactic Acid Bacteria Isolated From Traditional Iranian Yogurt for Antimicrobial Activity Against Pathogenic Bacteria

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Background and Aim: Lactobacilli are among the most important known probiotic species, and efforts are underway to isolate them from various sources. Probiotics beneficial effects on health, is the main reason for its wide use in dairy products including yogurt. This research was carried out to isolate and identify *Lactobacillus* species present in traditional Iranian yogurt.

Methods: Thirty samples of traditional yogurt were collected from different regions of Gachsaran district south of Iran. Bacterial isolations were carried out according to the standard international protocols. Following successive culturing on selective media, and obtaining typical colonies, and upon microscopic examination, five isolates were selected, and analyzed for anti-microbial activity against three bacteria and two pathogenic fungi. In order to confirm the biochemical results, three bacterial isolates were selected for molecular analysis, and their identification was performed by means of 16S rRNA sequencing.

Results: We isolated *Lactobacillus paracasei* JCM1171 with 1384 nucleotides and *Lactobacillus plantarum* subsp. *Plantarum*.A1 1380 nucleotides. The most anti-microbial activities were related to strain A and the least effects were related to strains X3. Moreover, among the pathogenic microorganisms used, *Staphylococcus aureus* and the fungus, *Aspergillus niger* had the highest sensitivity.

Conclusion: Different probiotic lactic acid bacteria were isolated from traditional Iranian yogurt samples collected from various regions, and their anti-microbial activity against a few pathogenic bacteria and fungi were evaluated. Two yogurt samples containing X3 and X4 strains were collected from remote destinations. The environmental conditions effecting the growth of these organisms were completely different, so their biochemical properties, like sugar fermentation and production of bio-active compounds, were unique. Because of their antimicrobial characteristics, these probiotic bacteria can be useful as bio-protective agents in controlling oral and gastro-intestinal infections.

Keywords: Probiotic bacteria; Anti-microbial activity; Molecular identification



Biochemistry



Investigation of the Effects of Hydroalcoholic Extract of *Citrullus colocynthis* and Pioglitazone on Insulin, TNF- α , Resistin, and Total Antioxidant Capacity in Type II Diabetic Male Rats.

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Background and Aim: Type 2 diabetes mellitus (T2DM) is the largest growing metabolic disorder of the endocrine system in the world. In addition to diet and physical activity, T2DM is also treated with medications. These medications include sulfonylureas, biguanidines, and thiazolidinediones, which have side effects such as hypoglycemia, nausea, vomiting, flatulence, diarrhea or constipation, and unusual fatigue. Side Effects of Long-term use of these drugs necessitates the production of effective drugs with less side effects, especially of plant origin. One of the anti-diabetic herbal remedies is *Citrullus colocynthis*. In this study, we aimed to compare the extract of *Citrullus colocynthis* with pioglitazone in the control of type 2 diabetes and some biochemical and inflammatory factors

Methods: In this study 60 male Wistar rats were divided into 6 groups. Two months after the high-fat diet, rats were diabetic using streptozotocin (except for the control group). After confirmation of diabetes mellitus and blood glucose measurement, animals were treated for three consecutive weeks with doses of 100 mg / kg, 200 mg / kg, and 400 mg / kg, of hydroalcoholic extract of *Citrullus colocynthis* and 10 mg / kg of pioglitazone. Then insulin, TNF- α , resistin, glucose, HbA1c, total antioxidant capacity, blood lipid profile were measured. Finally, the collected data were analyzed using SPSS software

Results: Overall, it seems that *Citrullus colocynthis* extract by decreasing blood glucose levels, lipid profile, inflammatory factors (TNF- α), predisposing adipokines to T2DM and Increasing the total antioxidant capacity could be an effective treatment for T2DM and prevention of its complications such as cardiovascular disorders.

Conclusion: Overall, it seems that *Citrullus colocynthis* extract by decreasing blood glucose levels, lipid profile, inflammatory factors (TNF- α), predisposing adipokines to T2DM and Increasing the total antioxidant capacity could be an effective treatment for T2DM and prevention of its complications such as cardiovascular disorders.

Keywords: *Citrullus colocynthis*; Pioglitazone; Type 2 Diabetes; Resistin; Insulin

Serum C-reactive Protein Level in Patients with Chronic Obstructive Pulmonary Disease and Its Correlation with Disease Severity

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Background and Aim: Background: It has been proven that patients with chronic obstructive pulmonary disease (COPD) are at risk for increased atherothrombotic events. Studies have demonstrated that C-reactive protein (CRP) and COPD are risk factors for heart disease. The present study aimed to evaluate the serum level of CRP in patients with COPD and compare it with those of the control group.

Methods: The present case-control study was conducted on 40 patients with COPD and 51 asymptomatic individuals (control subjects). High-sensitivity C-reactive protein (hsCRP), blood gases, and spirometry were evaluated in patients with COPD and compared with those of the control group.

Results: Patients with COPD had a higher hsCRP than the control group, and the difference was statistically significant (7.6 mg/L compared to 4.2 mg/L, P value less than 0.001). A negative correlation was found between hsCRP and the predicted percentage of forced expiratory volume in one second (FEV1%). There was also a positive correlation between hsCRP and the severity of COPD based on the Global Initiative for Chronic Obstructive Lung Disease (GOLD) criteria.

Conclusion: Serum hsCRP is higher in the COPD group than the control group matched based on age, and hsCRP is correlated with the severity of COPD. Further studies with larger sample sizes are required for evaluating this correlation in patients with COPD.

Keywords: CRP; Iran; Pulmonary disease; Smoking

Supplementation with Biotin and Brewer's Yeast Ameliorated the Neurological Manifestations in Covid-19 Patients

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Background and Aim: Besides the respiratory manifestation of Coronavirus disease 2019 (COVID-19), understanding the neurological complications following COVID-19 illness will be crucial. A scoping review on neurological manifestations in COVID-19 patients revealed that headache and olfactory and gustatory disorders were common neurological manifestations of SARS-CoV-2. Since malnutrition directly or indirectly affects a variety of organ systems including the central nervous system and peripheral nervous system, the aim of this study was to investigate the possible beneficial effects of biotin and brewer's yeast supplements in improving the neurological symptoms associated with Covid-19.

Methods: Patients with laboratory-confirmed COVID-19 infection were recruited from Shariati Hospital. A total of 60 patients were randomly assigned to two groups. The intervention group (n=30) received 5000 mcg twice daily of biotin and one tablet of brewer's yeast for 10 days in addition to the drugs used in the treatment of COVID-19. The control group receive only the routine medication in the treatment of COVID-19 and did not receive the experimental treatment. The patient's symptoms were recorded in the relevant checklist from the day of referral and the patients were followed up by telephone for 10 days.

Results: Compared with the control group, the neurological manifestations including headache (p=0.017) and olfactory (p=0.002) and taste function (p=0.004) significantly improved in the intervention group.

Conclusion: Our study demonstrated that 10 days of supplementation with biotin and brewer's yeast significantly ameliorated neurological manifestations of SARS-CoV-2.

Keywords: COVID-19; Neurological manifestations; Biotin; Brewer's yeast

The Effects of Multi-Wall Carbon Nanotubes on Cell Viability and Induction of Bax in Mice Skin Fibroblasts

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Background and Aim: Carbon nanotubes are one of the greatest discoveries in nanotechnology and they currently have various applications. However, health risks of long-term exposure to these materials remain to be elucidated. Since humans are widely exposed to nanoparticles and the mechanism of their toxicity is not yet known, we investigated nanoparticles effect on cell viability and the involved mechanisms, in mouse skin fibroblasts.

Methods: Cytotoxicity of carbon nanotubes on cell viability was examined by 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) assay, intracellular generation of reactive oxygen species (ROS) by fluorometric and lactate secretion determined by photometric method. Western blot analysis was done to evaluate the expression of BCL2-associated X protein (Bax) as a key factor in apoptosis pathway.

Results: MTT results showed a significant decrease in cell viability but increased levels of ROS in fibroblasts treated with carbon nanotubes. Western blot analysis revealed a significant increase in BAX expression, reflecting the effects of carbon nanotubes on cell death. Cellular lactate secretion, necessary for fibroblast proliferation and collagen synthesis, showed a significant decrease following exposure to carbon nanotubes.

Conclusion: Consequently, both carbon nanotubes should be considered potentially toxic for skin fibroblasts which are the body's first line of defense. Carbon nanotubes by increasing ROS and Bax levels may induce apoptosis in fibroblasts via Bax apoptotic-pathways. Also, suppression of lactate generation could reduce collagen production which may speed up aging process especially in the skin.

Keywords: Carbon nanotubes; fibroblasts; Cytotoxicity; ROS; Lactate.

TNF- α Plays a Crucial Role in The Development of Gestational Diabetes Mellitus

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Background and Aim: GDM is defined as glucose intolerance that is first recognized during gestation. (GDM) poses increased risk of short- and long-term complications for mother and offspring. Women with GDM are predisposed to many co-morbidities including operative delivery and type 2 diabetes mellitus (DM). Offspring of mothers with GDM are prone to adverse health outcomes including fetal macrosomia, respiratory difficulties and metabolic complications in the neonatal period, and carry a higher risk for future obesity and alteration in glucose metabolism. Tumor necrosis factor (TNF) often called tumor necrosis factor alpha or (TNF- α) is an adipokine and a cytokine. (TNF- α) is a proinflammatory cytokine that has recently been shown to be involved in the development of several chronic inflammatory and autoimmune diseases.. The aim of this study was to evaluate whether TNF- α participates in the pathogenesis of GDM.

Methods: A total of 60 women were enrolled in this study, which included 60 GDM patients and age- and sex-matched healthy pregnant women .Serum TNF- α and insulin levels were analyzed by ELISA and glucose levels by enzymatic method. insulin resistance was calculated according to the formula.

Results: We found that serum TNF- α levels in patients with GDM increased significantly and serum insulin levels decreased. In addition, there was a positive correlation between serum TNF- α levels and insulin resistance and glucose concentration.

Conclusion: TNF- α , on the one hand, plays an important role in the development of GDM by affecting the pancreas and the function of β cells, thereby reducing insulin production, and on the other hand, by creating insulin resistance in tissues.

Keywords: TNF- α ; GDM ; insulin

4-Hydroxyisulosin, A Lipid-Lowering Agent Similar to Orlistat, Improves Obesity in SW480 Cell Line

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Background and Aim: Obesity is a chronic disease caused by excessive accumulation of fat in the body. Obesity and overweight lead to weight-related diseases and a decrease in quality of life. In synthetic drugs, only orlistat and sibutramine have been approved for long-term use in the treatment of obesity and overweight, which, in addition to the high cost and significant side effects, have very limited efficacy in the treatment of obesity. 4-Hydroxy-L-isoleucine is bioactive compound in the fenugreek plant. The 4-Hydroxy-L-isoleucine plays a significant role in the improvement of dyslipidemia, diabetes, obesity and metabolic syndrome.

Methods: SW480 cells were cultured in RPMI1640 medium. The cells were treated for 24 and 48 hours with the 4-Hydroxy-L-isoleucine and Orlistat. IC₅₀ was calculated using MTT method. The cells were then pre-treated with the IC₂₀ concentrations for 24 and 48 hours before extracting RNA and synthesis cDNA. The gene expression of ACC and FAS was evaluated by Real Time PCR method.

Results: The results showed that the mRNA expression level of ACC (P=0/001) and FAS (P=0/024) in the SW480 cell line decreased significantly after 24 and 48 hours of treatment with 4-Hydroxy-L-isoleucine and orlistat.

Conclusion: The results of this study showed that the 4-Hydroxy-L-isoleucine, up-/down-regulated significantly the expression of ACC and FAS, similar to orlistat. So it can be suggested that it can be used as new suggestion with less side effect in the treatment of obesity.

Keywords: 4-Hydroxy-L-isoleucine; Fenugreek; Orlistat; Obesit

A Simple Procedure for Fractionation of Human Plasma for Production of Plasma Derived Albumin and Intravenous Injectable Immunoglobulins (IVIG)

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Background and Aim: Human plasma is the major source of several life-saving therapeutic proteins. Among these proteins plasma derived albumin and intravenous injectable immunoglobulins (IVIG) have been listed as the strategic therapeutics by the FDA. Plenty of chemical and chromatographic procedures have been introduced for purification of plasma proteins amongst which, cohn's cold ethanol precipitation procedure remains as the main reference approach. To ensure optimal use of the plasma resource which are not infinite the manufacture of plasma products has evolved dramatically in recent years, and regulations have been established to introduce dedicated treatments to increase the safety margin and avoid transmission of pathogens. In the present study we introduce a simple method for production of two strategic plasma-derived proteins albumin and IVIG. This method is a combination of chemical and chromatographic procedures with acceptable yield and purity.

Methods: The ionic strength of human plasma was adjusted and it was subjected to 40% cold ethanol precipitation at -20. After 2h incubation the mixture was centrifuged. The supernatant was treated as albumin fraction while the pellet was considered as albumin-depleted plasma proteins. The supernatant was subjected to 45% ammonium sulphate precipitation and then it was further purified by anion exchange chromatography. Cold ethanol pellet was dissolved in citrate buffer and was precipitated by octanoic acid. The octanoic acid supernatant is immunoglobulin rich fraction which was further purified by negative-phase anion exchange chromatography. The fractions were analyzed using UV spectrophotometry, SDS-PAGE and densitometry.

Results: UV spectrophotometry gave a rough estimation of protein content of each fraction. the purification yield for albumin and IG was more than 45% and more than 60%, respectively. Densitometry of SDS-PAGE image showed a rough estimation of purity for each fraction. This was more than 90% for both purified proteins.

Conclusion: Our designed procedure is capable of acceptable purity and yield for production of plasma derived albumin and total Immunoglobulins. This is indicative of the importance and operability of development of a large-scale fractionation procedure to provide mentioned strategic therapeutics to fulfill national requirements.

Keywords: Plasma Fractionation; protein purification; therapeutic proteins; Albumin; immunoglobulin

A Systematic Review and Meta-Analysis on Blood Lead Level in Opium Addicts: An Emerging Health Threat

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Background and Aim: This meta-analysis was conducted aiming to summarize the results obtained from the previous studies so that the effect of opium on blood lead levels (BLLs) can be investigated.

Methods: Scopus, Embase, PubMed, and Web of Science (ISI) were systematically searched up to June 2020. The heterogeneity of the included studies was evaluated using Cochrane's Q test and the I² statistic. A random-effects model was used to pool the weighted mean differences (WMDs) and their 95% confidence intervals (CIs).

Results: Out of a total of 2372 citations, eleven articles with 916 participants (487 opium addicts and 429 controls) were included in the study. The meta-analysis results showed that there were higher lead levels (WMD = 14.59 µg/dL, 95% CI = 11.59 to 17.92, Z = 8.60, P < 0.001) in opium addicts than in the control group. The degree of heterogeneity observed (P < 0.001, I² = 98.1%) might be mainly the result of the type of sampling and of consumption. Moreover, the findings of meta-regression analyses indicated that publication year (β = 1.23, P = 0.287), total sample size (β = 0.05, P = 0.728), and quality scores (β = - 2.91, P = 0.546) had no effects on lead levels in opium addicts. In the sensitivity analysis, it was found that the pooled WMDs remained stable after excluding one by one study.

Conclusion: Oral opium consumption increased the amount of lead in the bloodstream, and the measurement of lead levels in opium addicts' blood may be regarded as a useful test to diagnose and prognosis of disorders that may lead poisoning causes.

Keywords: Opium; Lead; Meta-analysis

A Systematic Review of the Effects of Vitamin C in the Control of Covid-19

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Background and Aim: Coronavirus COVID-19 infection caused by SARS-CoV-2 is a complex disease in which activation of the immune system and secreted cytokines causes oxidative stress. Oxidative stress plays an important role in the pathogenesis of viral infections and the death of infected cells. Vitamin C (ascorbic acid), a water-soluble vitamin, affects cellular immunity by eliminating free radicals and preventing oxidative stress and inflammation.

Methods: In this systematic review, we reviewed the findings of studies published in various databases, including Springer, PubMed, Science Direct, Wiley, and Scopus. This study was conducted in the period 2019 to 2021 using the keywords Covid-19 and Vitamin C.

Results: Studies have shown that high-dose intravenous vitamin C therapy has moderate to high efficacy, reducing infection by up to 8% in adults and up to 14% in children. Vitamin C strengthens the immune system by promoting phagocytosis and leukocyte chemotaxis, as well as the development and maturation of T lymphocytes during infection. Animal studies have shown that vitamin C increases infection resistance in chick embryo tracheal organ cultures and protects broiler chicks against avian coronavirus. The antioxidant and anti-inflammatory effects of Vitamin C are made by a significant decrease in serum IL-1 β and TNF α levels and increased in catalase, superoxide dismutase, and glutathione levels in a rat ARDS (The acute respiratory distress syndrome) model. Additionally, vitamin C also reinforces the function of the lung epithelial barrier by promoting the epigenetic and gene expression of protein channels in the alveolar-capillary membrane. These channels which regulate the clearance of the alveolar fluid include cystic fibrosis transmembrane conductance regulator, aquaporin-5, epithelial sodium channel, and the Na⁺/K⁺-ATPase pump. Several trials have shown that vitamin C reduces upper respiratory tract infection (URTI) and shortens the duration of mechanical ventilation in patients who need the longest ventilation.

Conclusion: Vitamin C is a safe and inexpensive essential nutrient with therapeutic effects against Covid-19 infection that can be used in combination with other potential therapies.

Keywords: Covid-19; Vitamin C; diet therapy; SARS-CoV-2; Coronavirus

Acute Liver Failure in a Pediatrician with COVID-19: A Case Report

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Background and Aim: There is ample evidence of liver failure in COVID-19 patients. The presence of increased liver enzymes in SARS-CoV-2 patients was discovered in 75 of 148 patients. Aspartate aminotransferase (AST) increased in 62 percent of intensive care unit (ICU) patients compared to 25 percent of patients who did not receive ICU treatment, suggesting that more severe illness is associated with deterioration of liver enzymes in this population. A retrospective cohort study has indicated that acute liver damage is frequent in SARS-CoV-2 positive patients, although usually mild. However, a severe disease course should be expected in the 6.4 percent of individuals with severe liver damage. 67% of patients who tested positive for SARS-COV-2 had higher alanine aminotransferase (ALT) levels than those who tested negative. Here, we report a case of acute liver failure in a pediatrician with COVID-19 infection without a history of preexisting liver disease. Unfortunately, the patient passed away several days after ICU admission. We desire to highlight the knowledge that requires further studies related to liver failure in COVID-19 patients.

Methods: On July 12, 2020, a 56-year-old man with fever, severe shortness of breath, cough, and A+ blood group was admitted to M-ICU at the Ghaem Hospital in Karaj, Iran. He was a pediatrician. A polymerase chain reaction (PCR) test had been performed before he was admitted. The PCR findings indicated that the patient was positive in terms of the presence of SARS-COV-2 at the 24.5 cycle threshold (CT) value. On the first day of admission, primary laboratory findings revealed the elevated level of white blood cells (WBCs, 20900/microliter) with a high count of neutrophil (91% of WBCs), impaired liver function; (SGPT or ALT, 127 U/L), acute inflammation; (ESR, 30 mm/hr), negative D-Dimer, and increased fibrinogen. On the 2nd post-admission day, lactate dehydrogenase (LDH, 477 U/L) test was performed. Also, a drastic decrease was seen in WBCs count. On the 5th post-admission day, the patient's partial pressure of oxygen (PO₂) and oxygen saturation (SO₂) were lower than normal, and his pulmonary capacity had reduced. Finally, the patient passed away due to respiratory failure, impaired liver function; (SGPT/ALT, 2500 U/L, SGOT/AST, 4200 U/L). The ground-glass opacity was apparent in peripheral areas of both lung sides.

Conclusion: The liver biochemistry abnormalities in COVID-19 patients are moderate (1-2 times the upper limit of normal). Increased serum ALT and AST values, observed in an estimated 29-39% and 38-63% of patients, respectively. There is no consensus on the association between liver enzyme level elevations and mortality in COVID-19 patients. In this regard, some studies have reported that there is no apparent association. On the contrary, several studies have revealed that elevations in the AST and ALT levels that are more than five times the upper limit of normal are linked with an increased risk of mortality. Here, our patient on day 11 had a higher level of AST (105 times the



upper limit of normal) and ALT (more than 62 times the upper limit of normal). A tentative explanation is that patients might have a robust immune response. Another reason is that aggressive therapies and anti-viral medication may lead to liver damage. It is noteworthy that the possibility of drug-induced liver damage due to hepatotoxicity is associated with drugs used in the treatment of COVID-19 such as lopinavir, ritonavir, and hydroxychloroquine should be taken into consideration by clinicians.

Keywords: SARS-COV-2; Aspartate aminotransferase; Alanine aminotransferase; COVID-19



Anti-Inflammatory Effects of Resveratrol on Cardiovascular Diseases: A Systematic Review and Meta-Analysis of Randomized Controlled Trials

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Background and Aim: Cardiovascular diseases (CVDs) include various disorders of the heart or/blood vessels. Inflammation is one of the factors that assumes a significant role in atherosclerotic plaque formation and the pathogenesis of CVDs. As a result, targeting inflammation can reduce the risk of developing CVDs. Resveratrol is a natural polyphenolic compound produced by plants and used as a dietary supplement to improve the metabolic profile. The effects of resveratrol on biomarkers of inflammation have been previously investigated, although the current results are controversial. Here we performed a systematic review and meta-analysis to evaluate the effect of resveratrol on CRP and TNF- α levels in CVDs for published Randomized Controlled Trials (RCTs).

Methods: Online research was conducted in the following database: MEDLINE, EMBASE, Cochrane Library, Web of Science databases, and Scopus. RCTs were included that investigated the potential effects of resveratrol supplements on biomarkers of inflammation among patients with cardiovascular diseases. The meta-analysis was performed using Comprehensive Meta-Analysis (CMA) V2 software.

Results: Seven clinical trials met the inclusion criteria and were selected for the current meta-analysis. The result of our study demonstrated that resveratrol significantly decreased levels of TNF- α (WMD= -0.325, 95% CI: -0.546, -0.104; $p = 0.004$), and CRP (WMD= -0.315, 95% CI: -0.519, -0.111; $p = 0.002$) in patients with cardiovascular diseases.

Conclusion: Our results recommend that resveratrol can be used as a potential therapy in patients with cardiovascular diseases by reducing inflammatory conditions.

Keywords: Cardiovascular disease; Inflammation; Resveratrol; CRP; TNF α

Anticancer Effects of Cinnamaldehyde through Inhibition of ErbB2/HSF1/LDHA Pathway in 5637 Cell Line of Bladder Cancer

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Background and Aim: The growing prevalence of bladder cancer worldwide has become a major concern for the researchers, and the side effects of chemotherapy drugs have always been a major problem in cancer treatment. Cinnamaldehyde, the active ingredient in the Cinnamon plant, has long been considered with anti-oxidant and anti-inflammatory effects.

Methods: Bladder cancer 5637 cell lines were treated with the different concentrations of Cinnamaldehyde. MTT assay was performed to evaluate cell viability at 24, 48, and 72 hours. The concentration of 0.02, 0.04, and 0.08 mg/ml of Cinnamaldehyde were selected. Apoptosis was assessed with Annexin V-FITC/PI and Hoechst33258 staining. Cell migration was performed by the scratch test. To evaluate Cinnamaldehyde effect on glycolysis, the gene expression of epidermal growth factor receptor 2 (ErbB2), heat shock protein transcription factor-1 (HSF1) and lactate dehydrogenase A (LDHA), as well as the protein levels of HSF1 and LDHA, LDH activity and finally glucose consumption and lactate production, were measured.

Results: Cinnamaldehyde significantly increased the rate of apoptosis in the 5637 cells. Also it significantly reduced the gene expression of ErbB2, HSF1 and LDHA, protein level of HSF1 and LDHA, LDH activity, as well as cell migration, glucose consumption and lactate production. These changes were dose-dependent.

Conclusion: Based on our findings, it is clear that Cinnamaldehyde has caused down-regulation of oncogene ErbB2 and HSF1. Given the association between ErbB2 and LDHA and HSF1 and LDHA, it can be argued that Cinnamaldehyde is a potent inhibitor of upstream glycolysis regulators. Although the exact mechanism of this association is not yet clear, it is likely that Cinnamaldehyde, by inhibiting transcription and translocation levels, actually blocks all pathways that help regulate cancer-related cell survival. Therefore, with the help of more research and the findings of in vivo, it is possible to place Cinnamaldehyde as a natural compound that does not have the side effects and consequences of chemotherapy drugs in the treatment of cancer and use it orally or by injection.

Keywords: Cinnamaldehyde; ErbB2; HSF1; LDHA; bladder cancer; cancer cell metabolism.

Antioxidant Effects of *Humulus lupulus* Extract on Carbon Tetrachloride-Induced Liver Damage in Rats

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Background and Aim: In this study antioxidant effects of aqueous extract of *Humulus lupulus* was investigated using carbon tetrachloride (CCl₄)-damaged rat liver as the experimental model.

Methods: Thirty male Wistar rats were divided into 5 groups (n=6). The two first groups including normal and CCl₄ control groups, administered with distilled water orally for 14 days. The three later groups received 50, 100, and 200 mg/kg of *Humulus lupulus* extract for 14 days respectively. All of the groups except normal control group received 1 ml CCl₄ (1:1 v/v dissolved in olive oil) on 14th day. All animals were sacrificed 12 h after post-treatment. The liver slices were collected and homogenized to determine the total antioxidant capacity (TAC) and malondialdehyde (MDA) parameters.

Results: Administration of carbon tetrachloride decreased the amount of TAC and increased the level of MDA compared to the normal control group. Pretreatment with a *Humulus lupulus* extract significantly increased TAC capacity and decreased the MDA level (P < 0.05).

Conclusion: The results of this study demonstrated that the pretreatment with *Humulus lupulus* extract significantly ameliorates CCl₄-induced liver damage in rats, which may be attributable to its potential antioxidant propensity.

Keywords: Antioxidant; Carbon tetrachloride; *Humulus lupulus*

Arsenic Trioxide Induction Autophagy in Human Acute Promyelocytic Leukemia

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Background and Aim: Autophagy is a survival pathway required for cellular viability during starvation through catabolic self digestion of damaged proteins and organelles; however, autophagy may result in cell death if it proceeds to completion. Although the exact mechanism of this process is not clear, it seems that proper regulation of autophagy can potentially contribute to the therapeutics of cancers.

Methods: In this study, the role of autophagy in the As₂O₃-induced death of NB4 cells was assessed using electron microscopy. Also; Quantitative real-time PCR method was used for expression of autophagy related genes in dose and Time dependent manner.

Results: Our results showed that autophagy induces death in NB4 cells treated with arsenic trioxide. It was also found that the Atg7, Bcln1 and LC3 genes, which are essential genes for induction of autophagy, are the target of arsenic trioxide invasion

Conclusion: This study showed that in addition to known mechanisms such as induction of apoptosis, epigenetic changes, induction of differentiation and angiogenesis changes in As₂O₃-treated leukemia cells, induction of autophagy also plays a key role in this therapeutic pathway.

Keywords: Acute Promyelocytic Leukemia; Autophagy; Gene Expression; Arsenic Trioxide

Association of Asprosin Concentration and Atherogenic Lipid Profile in Umbilical Cord Blood: A Cross-Sectional Study

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Background and Aim: Elevated lipids in umbilical cord blood affect fetal programming, which can lead to a higher risk of developing cardiovascular disease in later life. However, the causes of changes in lipid profile of umbilical cord blood are not clear yet. this study aimed to determine the association of asprosin concentration with TAG, TC, HDL-C, LDL-C concentrations and TAG/HDL-C, TC/HDL-C, LDL-C/HDL-C and non-HDL-C/HDL-C ratio in umbilical cord blood.

Methods: This cross-sectional study was based on 150 mother-pair of birth cohort study in Sabzevar, Iran. Multiple linear regression was used to estimate the association of lipid concentration and lipid ratios with asprosin in cord blood samples controlled for the relevant covariates.

Results: In fully adjusted models, each 1 ng/ml increase in asprosin was associated with 0.19 (95% CI: 0.06, 0.31, P < 0.01), 0.19 (95% CI: 0.10, 0.29, P < 0.01), 0.17 (95% CI: 0.09, 0.25, P < 0.01), 0.17 (95% CI: 0.09, 0.25, P < 0.01), 0.01 (95% CI: 0.00, 0.013, P < 0.01), 0.01 (95% CI: 0.01, 0.01, P < 0.01), 0.01 (95% CI: 0.01, 0.01, P < 0.01) and 0.01 (95% CI: 0.01, 0.01, P < 0.01) increase in TAG, TC, LDL-C, TAG/HDL-C, TC/HDL-C, LDL-C/HDL-C and non-HDL-C/HDL-C ratio respectively.

Conclusion: Overall, our findings support the link between cord asprosin concentration and development of atherogenic lipid profile in newborns. Further studies are needed to confirm the findings of this study in other populations.

Keywords: Asprosin; Lipid profile; Umbilical cord blood; Triglyceride; Cholesterol

Association of Oxidative Stress with Female Infertility

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Background and Aim: Oxidative stress (OS) results when the level of reactive oxygen species exceeds the capacity of antioxidants to neutralize them. In females, OS is hypothesized to negatively impact a number of processes involved in reproduction, including folliculogenesis, oocyte maturation, endometriosis, and embryo development. In this study, we measured some oxidative markers in infertile and normal individuals.

Methods: 80 infertile women and 73 normal were chosen according to certain including and excluding criteria. Malondialdehyde (MDA) as a lipid peroxidation marker, total antioxidant capacity (TCA), and the activity of antioxidant enzymes (superoxide dismutase (SOD) and catalase) were assessed in fertile and infertile women

Results: MDA levels in plasma were found to be higher significantly ($p < .001$) in patients and TAC was higher infertile individuals. SOD and catalase activities in plasma were significantly ($p < .05$) lower in infertile women.

Conclusion: All these results suggested that the examination of blood oxidative profiles might be helpful in the administration of antioxidants as adjuvant therapy in infertile patients and taking an antioxidant may provide benefits for subfertile women.

Keywords: Oxidative stress; superoxide dismutase; total antioxidant capacity; infertility

Association of PDL-1 Expression Pattern with Inflammatory Cytokines (TGF β and IFN γ) in Patients with Malignant Bone Tumor

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Background and Aim: Primary bone cancers include osteosarcoma, Ewing sarcoma, and chondrosarcoma that impose a burden of morbidity and mortality annually. Different clinical manifestations and complex histology remain as major therapeutic challenges for bone tumors that can be refined by early diagnosis and detection. To this aim, the gene and protein expression pattern of PDL-1 and its relevance to the inflammatory cytokines (IFN γ , TGF β) and patient's clinic pathophysiology were assessed in malignant bone tumors in the current study.

Methods: The tumor and normal bone tissue and serum of 60 patients with osteosarcoma and Ewing sarcoma bone tumors and the matched healthy subjects were enrolled in the study. The protein and gene expression level of PDL-1 were assessed via RT-PCR and immunohistochemistry and the level of IFN γ and TGF β were assessed using ELISA. The side-to-side comparisons were conducted using the parametric unpaired t-test and nonparametric Mann–Whitney U test. The chi-square test was used to analyze the statistical differences between bone tumors and the variables (age, gender, metastasis, chemotherapy status, tumor size, tumor grade, and tumor recurrence).

Results: The gene and protein expression level of PDL-1 was increased significantly in tumor tissue of patients compared to normal tissues. The increased level of IFN γ and TGF β in the serum of the patients with malignant bone tumor were also observed compared to healthy controls which was associated with both increased levels of PDL-1 protein and gene expression. It should be noted that the increase in PDL-1, IFN γ and TGF β was also associated with metastasis and recurrence and the chemotherapy status of patients.

Conclusion: Our data show a more efficient and accessible picture of PDL-1 expression pattern and serum levels of IFN γ and TGF β in bone cancer, which can be used as a possible marker for diagnosis in malignant bone tumor patients.

Keywords: PDL-1; IFN γ ; TGF β ; Malignant bone tumors; Cancer

Biotin and Brewer's Yeast Significantly Reduces Anorexia in COVID-19 Patients

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Background and Aim: Infected patients with coronavirus disease 2019 (COVID-19) can present anorexia and reduced nutrient intake due to their symptoms such as fever, headache, myalgia, and malaise. Eating disorder can lead to immune system weakness and increase the duration of recovery time. On the other hand, receiving nutrients such as biotin and brewer's yeast which is rich in B vitamins and minerals like selenium and chromium, is effective in preventing and improving the clinical symptoms of viral diseases such as Covid-19. The aim of present investigation was evaluating the possible beneficial effects of biotin and brewer's yeast supplements in improving the anorexia associated with Covid-19.

Methods: We identified 60 COVID-19 patients (older than 18 years) who were admitted to the Shariati Hospital, Tehran, Iran from January to March 2021. Patients who met our inclusion criteria were allocated in control (routine management) or intervention group (routine management, biotin 5000 mcg twice a day, and one tablet of brewer's yeast for 10 days) by simple randomization after PCR test. Included participants were followed up about the severity of anorexia by telephone interview for 10 days from the day of referral and data were recorded in the relevant checklist.

Results: The results of this study shown that the anorexia of Covid-19 patients in the intervention group significantly ($P=0.004$) reduced compared to the control group during 10 days follow up.

Conclusion: According to the results of the present study, it seems that the prescription of these supplements in the diet of these patients can be effective in the reducing anorexia.

Keywords: COVID-19; Anorexia; Biotin; Brewer's yeast

Carfilzomib Induces Drug Resistance in A2780 Ovarian Cancer Cells Through p53-Dependent and Caspase-3 Independent Pathways

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Background and Aim: Resistance to selective small-molecule inhibitors has been a substantial factor for limiting the efficacy of ovarian cancer. Recent studies have revealed that proteasome inhibitors induce acquired drug resistance. The possible mechanisms underlying the resistance to carfilzomib (CFZ), a recently developed inhibitor of proteasome, has not been well studied. This experimental study has aimed to determine if CFZ induces drug resistance in A2780 ovarian cancer cells through p53- and caspase-3 dependent pathways.

Methods: The A2780CFZ cells were generated by continuous culturing of A2780S cells in the presence of CFZ for 4 months. The MTT cytotoxic assay was applied to compare the survival rates in A2780CFZ and A2780S cells. Also, the relative expression of p53 and caspase-3 genes were evaluated using quantitative real-time polymerase chain reaction (qRT-PCR). The nonparametric ANOVA and Friedman tests were used for data analysis.

Results: A significant difference was observed between the viability of resistant- and sensitive- A2780 cells exposed to various concentrations of CFZ, indicating that A2780S cells have become resistant to this drug under long-term culture. Compared with A2780CFZ cells, the mRNA levels of p53 gene in A2780S cells were significantly increased after 12 ($P = 0.008$), and 24 hours ($P = 0.034$). Also, no significant differences were observed regarding caspase-3 mRNA levels between both cell lines ($P > 0.05$).

Conclusion: The findings of this study suggest that regulation of p53 gene expression in A2780CFZ cells might be the possible primary mechanism for gaining resistance against CFZ, but this might be independent of caspase cascades activation. Further studies are required to find strategies for overcoming CFZ resistance.

Keywords: Drug Resistance; Proteasome Inhibitor; Carfilzomib; Ovarian Cancer

Carvacrol Exerts Nephroprotective Effect in Rat Model of Diclofenac-Induced Renal Injury through Regulation of Oxidative Stress and Suppression of Inflammatory Response

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Background and Aim: Diclofenac (DIC) is an NSAID that can cause toxic effects in animals and humans and carvacrol (CAR) is a monoterpene compound that displays effective pharmacological and biological actions. The purpose of this work was to assess the influences of CAR on DIC-induced renal injury and oxidative stress in male rats.

Methods: The rats were segregated into four groups. Group 1, control group; Group 2 received DIC-only; Groups 3, received CAR-only and group 4 received DIC plus CAR. Changes in biochemical indexes, pathological changes, molecular biological indexes, and genes related to the inflammation of main organs were evaluated.

Results: The results of this work indicated that the amounts of the serum protein carbonyl, sGOT, sGPT, urea, creatinine, uric acid, nitrite content, MDA, serum TNF- α , and renal TNF- α gene expression were remarkably increased and the levels of the GPx, GSH, CAT, and SOD were significantly reduced in DIC-only treated animals compared to the control group. On the other hand, treatment with CAR after exposure to DIC led to significant improvements in abnormalities of DIC-induced renal injury and serum biochemical factors.

Conclusion: The data approve that CAR diminished the deleterious effects of DIC exposure. In this regard, the findings of this study indicated that the administration of CAR could alleviate the noxious effects of DIC on the antioxidant defense system and renal tissue.

Keywords: Carvacrol; Diclofenac; Oxidative stress; DIC-induced renal injury

Chicoric Acid Attenuates Palmitate-Induced TGF- β 1 Gene Expression in Human Peripheral Blood Mononuclear Cells

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Background and Aim: Type 2 diabetes (T2D) is an endocrine disorder around the world. The prevalence of T2D has increased markedly worldwide and is one of the most important global public healthcare problems. Several studies have demonstrated that transforming growth factor β 1 (TGF- β 1) signaling pathway has an important role in the pathogenesis of T2D. So, identification of agents that regulate this pathway could introduce novel therapies for T2D. Chicoric acid (CA), a phenolic compound extracted from chicory (*Cichorium intybus* L.), has been shown to exert anti-diabetic effects, although underlying the exact molecular mechanism of CA effects on T2D remains unexplored. In this study, we aimed to evaluate the effects of palmitate and CA on TGF- β 1 signaling pathway.

Methods: Forty subjects (20 newly diagnosed patients with T2D and 20 healthy subjects), were enrolled in this research. PBMCs were isolated from blood samples and then treated with palmitate and CA as follows: control groups (untreated, treated with bovine serum albumin (BSA) 1 % for 12 h), CA groups (treated with 50 μ M CA for 6 h), palmitate groups (treated with 500 μ M palmitate for 12 h), palmitate + CA groups (pretreated with 500 μ M palmitate for 12 h and then treated with 50 μ M CA for 6 h). Finally, TGF- β 1 gene expression was assessed using real-time polymerase chain reaction (PCR).

Results: Palmitate increased TGF- β 1 gene expression in PBMCs from healthy subjects and patients with diabetes ($p < 0.001$). However, palmitate-increased TGF- β 1 gene expression is reversely regulated by CA in healthy subjects from 2.21 fold to 1.34 and in patients with diabetes from 2.66 fold to 1.78 ($p < 0.001$).

Conclusion: Our results provide evidence that CA could be as a novel TGF- β 1 signaling pathway inhibitor through down regulation of TGF- β 1 gene expression. Although further investigation is needed to better understand the molecular mechanism of CA effects on diabetes.

Keywords: Type 2 diabetes (T2D); Palmitate; Chicoric acid (CA); Transforming growth factor β 1 (TGF- β 1)

***Chlorella vulgaris* is an Effective Supplement in Counteracting Non-Alcoholic Fatty Liver Disease-Related Complications through Modulation of Dyslipidemia, Insulin Resistance, and Inflammatory Pathways**

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Background and Aim: This study was aimed to investigate the effect of microalgae *Chlorella vulgaris* (*C. vulgaris*) on nonalcoholic fatty liver disease (NAFLD)-related complications induced by high-fat diet (HFD). Fifty adult male rats were divided into six groups. Control group and HFD group treated with or without *C. vulgaris* 5% and 10%. Biochemical parameters in serum were measured by spectrophotometric and enzyme-linked immunosorbent assay (ELISA) methods. The relative gene expression levels of Tumor Necrosis Factor- α (TNF- α), NF-kappa B (NF- κ B), and p38 Mitogen-Activated Protein Kinases (p38 MAPK) in the liver were assessed by using quantitative real-time PCR, while the protein levels of NF- κ B and TNF- α in the liver homogenate were determined by ELISA. The effects of HFD significantly were reversed by *C. vulgaris*, especially at a 10% dose. Therefore, it can be concluded that *C. vulgaris* therapeutically could be useful to improve NAFLD and its complications.

Methods: Fifty adult male Wistar rats (214–244 g) were used in the present study. Animals were maintained in standard conditions (12-hr dark/ light cycle at $22 \pm 2^\circ\text{C}$) for one week. Afterward, animals were randomly divided into two groups including normal control received standard chow (N) and rats who received a high-fat diet (HFD) to induce NAFLD. The treatment period by standard chow and HFD lasted for 12 weeks and after the fifth week, both N and HFD groups were divided into six groups. The groups were; group N: normal control received standard chow (n = 8), group NCv5: normal control received standard chow containing 5% (w/w) *C. vulgaris* powder (n = 8), group NCv10: normal control received standard chow containing 10% (w/w) *C. vulgaris* powder (n = 8), group H; received high-fat diet (n = 10), group HCv5: received high fat diet containing 5% (w/w) *C. vulgaris* powder (n = 8) and HCv10: received high-fat diet containing 10% (w/w) *C. vulgaris* powder (n = 8; Lee et al., 2008).

Results: The analysis results of biochemical and general parameters are presented in Table 1. There were no statistical differences in body weight between groups at the beginning of study, but, compared with the control group, treatment with HFD significantly increased the final body weight ($p < .001$). Administration of *C. vulgaris* considerably modulated these parameters in the HFD group ($p < .001$). However, analysis of glycemic control indices revealed that feeding animals with an HFD regimen caused a significant increase in concentrations of FBS and HOMA compared to the controls ($p < .001$), conversely, *C. vulgaris* administration significantly reversed these effects ($p < .001$). There was no significant difference in the insulin concentration between the studied groups ($p > .05$). NAFLD induced by a high-fat diet promotes the accumulation of TG and TC in liver tissue in comparison to control subjects ($p < .001$). The presence of powdered *C. vulgaris*, especially at 10% dose, significantly ameliorated the effects of HFD on liver TG and TC ($p < .05$).

Conclusion: In the present study, the effect of *C. vulgaris* 5% and 10% on the HFD-induced NAFLD-related complications were investigated. Our results revealed that NAFLD not only induces dyslipidemia and insulin resistance phenotypes, but also up-regulates the TNF- α , NF- κ B and p38 MAPK genes. Furthermore, liver histopathology of animals fed HFD regimen, revealed severe steatosis, ballooning degeneration as well as lobular and sinusoidal inflammation. However, *C. vulgaris* especially with a 10% dose, significantly reversed NAFLD-related complications.

Keywords: *Chlorella vulgaris*; high fat diet; NF-kappa B; Nonalcoholic fatty liver disease; P38 mitogen-activated protein kinases; TNF- α

Chronic Exposure to BPA Induce Diabetes Mellitus Type II by Alleviation of Nesfatin-1 in Mice

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Background and Aim: One of the emerging organic pollutants is bisphenol A [BPA, 2,2-bis-(4hydroxyphenyl) propane]. BPA is a profusely manufactured organic chemical, extensively used in the production of numerous household and every-day products. As a result, BPA is bulkily discharged in the environment, being traced in municipal wastewaters, sewage sludge, inland and marine waters. An emerging and rapidly growing area in the study of BPA's toxicity is its impact on the cardiovascular (CV) system. Recent epidemiological studies have shown that higher urinary BPA concentration in humans is associated with various types of CV diseases, including angina, hypertension, heart attack and coronary and peripheral arterial disease. Extensive studies have suggested potential links between BPA exposure and diseases including obesity and diabetes. Statistically significant association were found between higher BPA concentrations and CV diseases and diabetes. Association between urinary concentrations of BPA and type 2 diabetes in Korean adults also was reported. Nesfatin-1 is a new peptide that controls appetite and food intake. It was reported that nesfatin-1 has anti-hyperglycemic effects and administration of Nesfatin-1 significantly reduced blood glucose and represents a new class of insulin helpers. The objective of this research was to study the changes of serum nesfatin-1 in mice with chronic exposure.

Methods: Thirty mice randomly divided into two group (BPA+ and BPA-) and BPA administered as i.p. injection at dose 100 µg/kg BW for 45 days in BPA+ group. At the end of the study, blood samples were taken from heart and glucose, insulin, TG, cholesterol, LDL, HDL and Nesfatin-1 concentrations were measured and the relationship between nesfatin-1 and metabolic parameters were investigated.

Results: Serum nesfatin-1 concentrations in BPA exposure mice were lower than healthy controls (0.18 ± 0.04 ng/ml, 0.35 ± 0.08 ng/ml, respectively, and $p = 0.011$). We found a negative correlation between nesfatin-1 and fasting glucose and cholesterol, TG and LDL. In obese mice, serum nesfatin-1 concentrations were significantly lower when compared with non-obese subjects (0.16 ± 0.03 ng/ml, 0.37 ± 0.05 ng/ml, respectively; $p = 0.002$). In addition, we found that atherogenic index in mice received BPA were significantly higher in comparison with healthy ones (0.93 ± 0.11 ng/ml, 0.32 ± 0.13 ng/ml, respectively; $p = 0.005$).

Conclusion: Our study has shown that chronic exposure to BPA induce diabetes mellitus type II and nesfatin-1 concentrations were significantly decreased. Also the results of this study indicate that nesfatin-1 may have a significant role in diabetes mellitus type II.

Keywords: Bisphenol a; Nesfatin-1; Diabetes Mellitus; Mice; chronic exposure

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Comparison of the Expression Regulation of Three Genes in Cisplatin-Induced Nephrotoxicity

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Background and Aim: Cisplatin is a powerful chemotherapeutic drug. Main side effect of cisplatin is nephrotoxicity due to reactive oxygen species (ROS) production. ROS cause apoptosis by inducing the Caspase-3 activity. Studies showed Heme Oxygenase and Clusterin gene expression were induced in nephrotoxicity due to cisplatin administration. Our purpose was to comparison of Gallic acid effect on gene expression of Heme Oxygenase, Clusterin and Caspase-3 in Cisplatin nephrotoxicity in rats.

Methods: group1:(Control)(distilled water,10 days, gavage); group2:(Gallic acid 50 mg/kg,10 days, gavage); group3:(Cisplatin 6 mg/kg,I.P. on 5th day); group4:(Gallic acid:50 mg/kg,10 days and Cisplatin,6 mg/kg,I.P., on 5th day). Rats were anaesthetized. The gene expression levels were assessed by RT-qPCR in four groups on kidneys.

Results: Cisplatin group showed significant Up-regulation in Caspase-3 and Heme Oxygenase mRNA expression. The Gallic acid + Cisplatin group showed significant down-regulation in Caspase-3 and Heme Oxygenase expression compared to the cisplatin group alone (P less than 0.05). The expression level of Clusterin gene was not showed significant Up-regulation or downregulation.

Conclusion: Caspase-3 and Heme Oxygenase genes generally induced by drugs that produce nephrotoxicity. Gallic acid attenuated the Cisplatin-induced increase in Caspase-3 and Heme Oxygenase gene expression but not in Clusterin gene expression. Therefore, Gallic acid can have considered a useful compound in improve outcomes in patients with cisplatin induced nephrotoxicity.

Keywords: cisplatin; gene expression; Caspase-3; Heme Oxygenase; Clusterin.

Correlation Between Insulin Resistance and Polycystic Ovary Syndrome

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Background and Aim: Polycystic Ovary Syndrome (PCOS) is a heterogeneous disorder presenting with several complaints including ovarian dysfunction, hyperandrogenism, menstrual irregularity, insulin resistance (IR), metabolic disorder and obesity. This syndrome is associated with features of the metabolic syndrome including type 2 diabetes (T2D), hypertension, and dyslipidemia. Insulin resistance is associated with consequent hyperinsulinemia, which activates excessive ovarian androgen production, leading to polycystic ovary syndrome development.

Results: Articles show between 44% and 70% of women with polycystic ovary syndrome are insulin-resistant. Recent studies suggest that hyperinsulinemia, due to increased B-cell secretory capacity, is a prominent feature in polycystic ovary syndrome subjects independent from body weight, which drives levels of androgen excess in polycystic ovary syndrome. Clinical trials had also verified abnormal glucose tolerance leads to type 2 diabetes in polycystic ovary syndrome women. Articles demonstrated many women with polycystic ovary syndrome have insulin resistance beyond that predicted by their body mass index, with 50–70% of these women demonstrating insulin resistance by various measures. Other articles show insulin resistance and hyperinsulinemia play a role in the reproductive, and endocrine features of polycystic ovary syndrome by contributing to hyperandrogenism and disruption of gonadotropin secretion.

Conclusion: Insulin resistance is part of the pathogenesis of polycystic ovary syndrome and is associated with increased risk of type 2 diabetes in polycystic ovary syndrome also the individual family histories of type 2 diabetes and obesity will increase the prevalence of both diseases in women with polycystic ovary syndrome.

Keywords: Polycystic Ovary Syndrome; Insulin Resistance; Type 2 Diabetes

Correlation Between Serum Manganese Level and LDL Oxidation-Induced Malondialdehyde Levels in Cardiovascular Patients

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Background and Aim: In recent years, a number of studies have pointed to the Mn and MDA levels as a risk factor for cardiovascular diseases; the aim of this study was to study the relationship between serum Mn level and Malondialdehyde LDL-induced in coronary artery patients.

Methods: In this case-control study, 138 people who referred to Bushehr Heart Center for routine examination participated. Demographic information of the participants was registered by using a standard questionnaire. In this data article, we measured the level of Mn and malondialdehyde (MDA) in the blood of patients undergoing coronary angiography. The samples were taken from patients with cardiovascular disease in Bushehr's university hospital, Iran. Patients were divided in two groups: normal angiography and abnormal angiography. After the chemical digestion of samples, the concentration levels of Mn and MDA in both groups by using ELISA kits. Before undergoing the angiographic processes, venous blood samples were obtained via venipuncture from the antecubital vein into gel and clot activator tubes and stored at -80°C until the analysis.

Results: Serum levels of Mn in the case group were not significant as compared with the control ($p=0.575$). There was significant in MDA level between the two groups ($p=0.001$). By separating the four factors like age, sex, and type 2 diabetes in the two groups, still, there was significant difference between the two groups ($p=0.033$, $p=0.011$, $p=0.002$ respectively). The factors like BMI, hyperlipidemia and blood pressure in the two groups, there was no significant difference between the two groups ($p=0.577$, $p=0.307$ and $p=0.726$ respectively). Further, Mn level and MDA in the serum of the two groups were not associated ($p=0.780$).

Conclusion: The results of this study suggest that serum MDA level is associated with coronary artery disease. But serum Mn level is not associated. In addition, no relation was found between serum Mn and MDA levels in the two groups.

Keywords: Manganese; LDL; Malondialdehyde; Cardiovascular

Correlation of Lipid Ratios and TyG Index As Insulin Resistance Indicators with the Severity of COVID-19: A Cross-Sectional Study

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Background and Aim: Data coming from different sources show that loss of metabolic health and insulin resistance (IR) are correlated with the poor prognosis of COVID-19. Since the direct (hyperinsulinaemic-euglycaemic clamp technique) and indirect methods (HOMA-IR, QUICKI, and FG-IR) for IR assessment are technically demanding, time-consuming and expensive. Therefore, using simple and inexpensive biochemical indices for IR estimation can be helpful for determining COVID-19 prognosis. So, this study aims to evaluate triglyceride and glucose index (TyG) and lipid ratios such as TC/HDL-C, TG/HDL-C, LDL-C/HDL-C indices, as markers of IR, with the severity of COVID-19

Methods: In the current study, we collected demographic, clinical, and biochemical data to conduct a cross-sectional study in 1200 adult patients with confirmed COVID-19 diagnosis by quantitative polymerase chain reaction (qPCR). Biochemical parameters including fasting blood glucose (FBG), total cholesterol (TC), triglyceride (TG), low-density lipoprotein-cholesterol (LDL-C), and high-density lipoprotein-cholesterol (HDL-C) were evaluated using the commercially available kits (Pars Azmoon, Tehran, Iran). TC/HDL-C, TG/HDL-C, LDL-C/HDL-C, and TyG indices were calculated. Regression models were used to investigate the association between TyG with the severity of COVID-19

Results: The results showed the TyG index and TG/HDL-C levels were significantly higher in the severe group (severe vs. mild 8.63 ± 0.023 , 8.78 ± 0.05 , $p = 0.008$ and 3.61 ± 0.11 , 4.08 ± 0.19 , $p = 0.039$ respectively) Logistic regression model revealed that an increase in TyG and TG/HDL-C indices at the time of COVID-19 diagnosis positively correlated with disease severity ($OR = 1.33$, $p = 0.029$ and $OR = 1.08$, $p = 0.01$ respectively) among Iranian diagnosed people with coronavirus infection after adjusting for BMI, sex and age. TC/HDL-C, LDL-C/HDL-C ratios did not show a significant correlation with disease severity.

Conclusion: The surrogate indicators of IR including TG/HDL-C ratio and TyG index can be used as an early biochemical marker of COVID-19 severe prognosis.

Keywords: Insulin resistance; Lipid ratios; TyG index; COVID-19; Severity

Covid-19 and Male Infertility

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Background and Aim: It has shown that Covid-19, which affects almost all human organs also affects male genital system and sperms and impairs male fertility. virus temporarily reduces number of sperms for weeks to months, where it may cause infertility. Studies revealed that ACE2 enzyme activity is increased in men with Covid-19 which express highly in male genital system.

Methods: This study was performed on 25 fertile men without Covid, 25 fertile men with Covid-19 and genital fluids were analyzed. In this case-control study, sampling was performed within 4 months after obtaining consent from patients. parameters, such as semen volume , cell number, morphology and pH were evaluated.

Results: Andrological and morphological studies of sperms showed that sperms number, motility and morphology decreased by 34%, 26% and 41%, respectively. And 37% of cells decreased in all three parameters

Conclusion: This study unravelled that virus affects sperms, which is associated with reduced sperm quality and infertility.

Keywords: Covid-19; male infertility

COVID-19 and Renin Angiotensin Aldosterone System: Pathogenesis and Therapy

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Background and Aim: The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) binds to the ACE2 component of the renin-angiotensin-aldosterone system (RAAS) and infects the human cells. The aims of the present review were to look at the role and alteration of the RAAS components in SARS-CoV-2 infection, therapeutic approaches, and clinical trials in this field.

Results: ACE inhibitors, angiotensin receptor blockers, and mineralocorticoid receptor inhibitors are agents that significantly enhance the ACE2 and Ang-(1-7) levels, which can be suggestive for their role as therapeutics against SARS-CoV-2 infection. Beta-adrenergic blockers, which negatively regulate renin release from juxtaglomerular cells, and vitamin D, as a regulator of the RAAS and renin expression, are proposed therapeutics in the treatment of COVID-19. Some antihyperglycemic agents could be potentially protective against COVID-19-induced lung injury. Also, the inhibition of the Janus kinase/signal transducer and activator of the transcription pathway as a potential treatment for COVID-19 has been suggested. Finally, resveratrol, an antioxidant that can suppress Ang II, has been suggested as an adjunct to other therapies.

Conclusion: Regarding the suggested potential therapies for COVID-19, there are many clinical trials whose results might change the treatment strategies of SARS-CoV-2 infection. So, the results of well-organized clinical trials on the efficacy and safety of the mentioned agents in the treatment of COVID-19 will be useful in the management and therapy of the disease.

Keywords: ACE2; antihyperglycemic drugs; COVID-19; Janus kinase inhibitors; RAAS inhibitors; SARS-CoV-2

COVID-19 in Schizophrenic Patients: A Focus on Dysregulated Immune Response

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Background and Aim: Considering that the mortality rate of COVID-19 continues to rise, looking for the people being at higher risk of developing this illness is of utmost importance. There is growing evidence showing the increased susceptibility to COVID-19 infection and poor prognosis in schizophrenic patients. One of the most important mechanisms that link schizophrenia to more severe COVID-19 infection is the dysregulated immune system. As of today, there is a huge number of studies reporting the significant association between the mortality rate in schizophrenic patients and the elevated level of inflammatory markers including, interleukin-6 (IL-6), tumor necrosis factor- α (TNF- α), C-reactive protein (CRP), growth differentiation factor 15 (GDF-15), and D-dimer. Interestingly, both chronic schizophrenia and drug-naïve first-episode psychosis (FEP) are demonstrated to exhibit a systemic pro-inflammatory state, as observed by higher pro-inflammatory biomarkers such as IL-1b, IL-6, TNF- α , GDF-15, and CRP. These findings raise the possibility that the systemic hyper-inflammation triggered by SARS-CoV-2 infection may be more pronounced in patients with schizophrenia, which ultimately leads to the damage of tissues and multiorgan failure. we provide a narrative of the current understanding of COVID-19 in patients with schizophrenia and the potential of the dysregulated immune system that may be linked to increased sensitivity.

Results: High concentrations of inflammatory biomarkers such as CRP (>200 mg/L) and ferritin ($>2,500$ ng/mL) at admission are strong predictors of COVID-19 severity. As such, patients with SMD (severe mental disorders), including schizophrenia that hospitalized for severe COVID-19, had high levels of inflammatory biological markers such as CRP, IL-6, D-dimer, and ferritin. Furthermore, there is a relationship between lymphocyte subpopulations such as natural killer (NK) cells that contribute to early antiviral immunity and schizophrenia. Interestingly, the low number of NK cells is a common finding in individuals with schizophrenia and severe COVID-19. Therefore, due to the decreased NK activity in schizophrenic patients, higher viral loads can reduce the immune response, exposing them to more severe COVID-19. Added to this, there is evidence that exposure to antipsychotic therapy is associated with the increased risk of respiratory infections, which are a deadly clinical demonstration of COVID-19. These findings, at least in part, explain why schizophrenic patients are more susceptible to COVID-19 and experience worse clinical outcomes compared to the general population.

Conclusion: The current evidence shows that schizophrenia may increase the risk of mortality and the complications arising from the COVID-19 due to the dysregulated immune system. Those with



schizophrenia are a highly vulnerable population, whose immune response may decline than the general population.

Keywords: Schizophrenia; COVID19; Dysregulated immune system; Inflammation; Psychosis



CTRP1 and Adiponectin Can be Considered as Emerging non-Invasive Parameters of Liver Enlargement in the Context of Non-Alcoholic Fatty Liver Disease

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Background and Aim: Non-alcoholic fatty liver disease (NAFLD) leads to hepatocyte fatty infiltration and liver enlargement. More importantly, this condition is often complicated by clinically significant portal hypertension, reflected by measuring portal vein diameter. Nowadays, tremendous efforts have been made to develop new biomarkers to help achieve this goal. Adipokines, especially the members of CTRPs are promising candidates for novel pharmacological treatment strategies and as diagnostic tools. The adiponectin and its paralogue; adiponectin CTRP1 have captured attention in recent years. CTRP1 and adiponectin are involved in the regulation of inflammatory pathways and other signaling pathways pertinent to the pathogenesis of NAFLD. Here, we aimed to evaluate the possible association of adiponectin and CTRP1 with the portal vein and liver enlargement in the context of type 2 diabetes (T2D) and NAFLD.

Methods: The circulating level of CTRP1 and adiponectin was evaluated in patients with NAFLD (n=22), NAFLD+T2D (N=20), T2D(N=20), and controls (N=20). diameters of the liver and portal vein were evaluated with abdominal sonography.

Results: The serum levels of adiponectin and CTRP1 in patients with NAFLD, NAFLD+T2D, T2D were significantly lower and higher, respectively, compared with those in controls. CTRP1 levels showed a positive correlation with liver diameter. However, adiponectin level was inversely correlated with this index. Neither adipokines nor CTRP1 was correlated with portal vein diameters.

Conclusion: It seems that CTRP1 and adiponectin can be considered as emerging non-invasive parameters of liver enlargement. However, studies with a larger sample size are needed to establish this concept

Keywords: CTRP1; adiponectin; NAFLD; T2DM; liver diameter axis

Dexamethasone Suppresses the Proliferation and Migration of VSMCs by FAK in High Glucose Conditions

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Background and Aim: High glucose conditions cause some changes in the vessels of diabetes through the signal transduction pathways. Dexamethasone and other corticosteroids have a wide range of biological effects in immunological events. In the present study, the effects of dexamethasone were investigated on the VSMC (vascular smooth muscle cell) proliferation, and migration based on the FAK gene and protein expression changes in high glucose conditions.

Methods: The vascular smooth muscle cells were cultured in DMEM and were treated with dexamethasone (10^{-7} M, 10^{-6} M, and 10^{-5} M) for 24, and 48 hours in high glucose conditions (25 mM). The cell viability was estimated by MTT method. The FAK gene and protein expression levels were determined by RT-qPCR and western blotting techniques, respectively. A scratch assay was used to evaluate cellular migration.

Results: The viability of VSMCs was less than 50% with dexamethasone values higher than 10^{-4} M. The FAK gene expression levels decreased significantly dependent on dexamethasone at 24 and 48 hours. However, the decreases of FAK protein expression levels as compared with gene expression levels were significantly observed with a time lag at 24 and 48 hour periods.

Conclusion: The results showed that inhibition of VSMC proliferation and migration by dexamethasone in high glucose conditions may be related to the changes of FAK.

Keywords: VSMC; FAK; High-glucose; Migration; Dexamethasone

Diagnosing COVID-19 Using Novel Biomarkers, Artificial Intelligence, and Deep-Learning Technology

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Background and Aim: The coronavirus disease 2019 (COVID-19) is still spreading worldwide causing infections and deaths. To find a better course of treatment, the need for prompt identification and diagnosis is vital and our need for effective biomarkers is obvious. Moreover, using new technologies such as Artificial Intelligence (AI) based diagnosis could be a game changer in fighting this pandemic or the potential future ones.

Methods and results: Studies have shown that IL-6 which is activated by macrophages is sharply elevated in COVID-19 patients. Lactate Dehydrogenase could be another biomarker as its secretion is triggered from necrosis cell membrane which could be a sign of viral infection. D-Dimer which is the measure of coagulation has been higher in non-survivors than in survivors. Moreover, Thrombocytopenia could be used as a marker since patients had significantly lower platelet count. Because of the growing evidence of higher mortality in patients with cardiovascular disease, using cardiac troponin I (hs-TnI) as a marker seem reasonable. On the other hand, coughing has been one of the symptoms of COVID-19 in many patients. Using AI on a smartphone app, a complex diagnosis device was made which records the sounds of the cough and compares it to the database. The results showed that the app was able to distinguish between COVID cough and non-COVID cough. Furthermore, AI which uses deep-learning algorithm may help in increasing diagnosing accuracy of COVID-19 using X-ray images as an alternative to Reverse transcription polymerase chain reaction (RT-PCR) which is the main clinical tool in COVID-19 diagnosis.

Conclusion: AI has shown a promising future in diagnosis of COVID-19 because of its accurate and sensitive recognition ability. Moreover, using AI-based technology will reduce the need for medical personnel. Using better biomarkers together with AI and deep-learning technology may bring new horizons in medical treatment and better management of pandemic.

Keywords: COVID-19; SARS-CoV-2; Biomarkers; Artificial Intelligence; Deep-Learning

Diagnostic Performance of Beta2-Microglobulin in Discriminating Between Good and Poor Early Graft Function: A Cut-Off Value Determination

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Background and Aim: Poor early graft function (PEGF) is a common complication occurring during the first-week post kidney transplantation (post-KT). PEGF is strongly associated with increased morbidity and mortality in kidney recipients (KRs). Timely diagnosis of PEGF leads to better managements and proper decisions. The aim of the present investigation is to evaluate the diagnostic and prognostic capability of beta-2 microglobulin (B2M) in discriminating between Good early graft function and PEGF.

Methods: In this cross-sectional study, KRs (n=39) were recruited. Inclusion criteria were: 1) recipients of the first kidney either from living donors (LD)- or brain death donors (BDD); 2) Age >14 years and 3) inhabitant of West Azerbaijan province, Iran. Exclusion criteria were: 1) autoimmune disease and 2) acute phase and/or chronic inflammation. PEGF was defined as requirement for dialysis within 7 days after operation and/or serum creatinine >1.70 mg/dL on fifth day after transplantation. Blood (4 mL) was drawn by venipuncture in a Vacutainer® Tube. Sampling was performed at day -1 (preoperative period) and at 2, 16, 24, 36, 48, 60 and 72 hours. Serum B2M (AESKU) was measured using ELISA method.

Results: This study compromised kidney recipients (males: n=21; Age: 38.3±14.5 and females: n=18; Age: 45.6±10.6; P=0.121). The frequency of PEGF was 12 (30.8%). No significant difference was noted in B2M levels between GEGF and PEGF groups at preoperative period and at 2, 16, 24 and 36 hours. Area under curves (AUCs [95% confidence intervals]) for B2M at 48-, 60- and 72 hours were 0.7 [0.50-0.89], 0.78 [0.63-0.93] and 0.78 [0.63-0.93], respectively. The Cut-off value, sensitivity and specificity for B2M at 72 hours were 3.55 µg/ml, 75% and 65%, respectively.

Conclusion: In renal transplant patients, B2M offers a good diagnostic and prognostic capability in discriminating between PEGF and GEGF. Future and multicenter studies are deemed necessary to establish our findings on large scales.

Keywords: Poor early graft function; Beta-2 microglobulin; Renal recipients; Transplantation

Does Opium Affects the Malondialdehyde in Coronary Artery Disease?

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Background and Aim: Studies displayed that consuming opium has a direct association with the incidence of cardiovascular diseases (CVD), and increases the risk of inflammation and oxidative stress. Oxidative stress affects lipids and leads to generating secondary compounds such as malondialdehyde (MDA). Therefore, the aim of the present study was to explore the role of opium on the plasma levels of MDA in the coronary artery disease (CAD) opium-addicted and non-opium-addicted patients.

Methods: This case-control study was conducted on 77 CAD-suspected men. Subjects were divided into three groups: 1) CAD opium-addicted (CAD + addict, n=30); 2) CAD non-opium-addicted (CAD, n=30); and 3) non opium-addicted with no CAD as a control group (Ctrl, n=17). The blood level of MDA was determined by the Thiobarbituric acid assay method.

Results: The plasma level of MDA was significantly increased in both CAD and CAD + addict patients in comparison with the Ctrl group ($P = 0.01$ and $P = 0.002$, respectively).

Conclusion: There were no significant differences between CAD and CAD + addict patients, therefore, it appears that opium addiction did not the synergistic effects with CAD to increase the levels of MDA.

Keywords: Opium; Coronary artery disease; Malondialdehyde

Does Sour Tea Have an Effect on Cardiometabolic Risk Factors Compared to Other Herbal Teas and Antihypertensive Drugs? Results of A Systematic Review and Meta-Analysis of Randomized Controlled Trials

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Background and Aim: The sour tea plant has been linked to several properties including antihypertensive and antioxidant properties. Our goal with this meta-analysis was to synthesize the evidence concerning the effect of sour tea compared to other herbal teas and antihypertensive medications on cardiometabolic risk factors.

Methods: In this study searches were conducted in PubMed, Web of Science (ISI), Embase, and Scopus (Elsevier) databases to identify articles related to the subject that have been published as of December 2019. Throughout this review, all clinical trials were included if they evaluated the effects of sour tea consumption on systolic blood pressure (SBP), diastolic blood pressure (DBP), lipid profile including LDL-C (low-density lipoprotein cholesterol), HDL-C (high-density lipoprotein cholesterol), TC (total cholesterol), TG (triglyceride); and FBS (fasting blood sugar) for more than one week. The weighted mean difference (WMD) and 95% confidence intervals (CI) were applied for explanation of the effect sizes.

Results: There were 22 studies with 24 effect sizes included in the analysis. Our results revealed that sour tea consumption had a significant reduction effect on the SBP levels (WMD: -7.14, 95 % CI: -11.16, -3.12, p less than 0.001), and DBP levels (WMD: -3.54, 95 % CI: -5.02, -2.06, p less than 0.001), while statistically significant changes was not seen in terms of lipid profiles, FBS, body mass index (BMI) and body weight (BW). Moreover, for SBP and DBP a significant level of reduction was observed in the subgroup with dose of greater than or equal 2 gr and duration of intervention (less than 4 and greater than or equal 4 week).

Conclusion: Based on the results of this meta-analysis, *Hibiscus sabdariffa* consumption has been shown to effectively decrease both SBP and DBP and the reduction in SBP was obviously greater in dose of more than 2 g but the changes in lipid profiles, FBS, BMI and BW were not significant.

Keywords: *Hibiscus*; blood pressure; lipid profiles, body mass index; blood glucose.

Effect of Camphor on Biochemical Factors and Antioxidant Enzymes Against Gentamicin-Induced Nephrotoxicity in Rats

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Background and Aim: Camphor is a natural antioxidant with anti-inflammatory and tissue repair properties. Nephrotoxicity is considered as an important side effect of gentamicin use. Therefore, evaluating the effect of natural antioxidants can resolve this complication. We aimed to assay the effect of camphor on biochemical factors and antioxidant enzymes such as Catalase (CAT), Glutathione peroxidase (GPX), Glutathione (GSH), Malondialdehyde (MDA), Nitric oxide (NO), urea, creatinine and urine protein against gentamicin- induced nephrotoxicity in rats.

Methods: Thirty adult male Wistar rats were allocated to 5 groups. Positive control and treatment groups were given gentamicin to induce nephrotoxicity. Animal treatment groups were treated with camphor in olive oil for 12 days. Serum, renal tissue and urine of rats were taken after the twelfth day. Desired factors and enzymes were investigated by suitable methods.

Results: Our study revealed that nephrotoxicity caused by gentamicin showed a marked decrease in the catalase, GPX, MDA and GSH activity levels of renal tissue and the GSH and GPX activity of serum, however, presented an increase in MDA, urea and NO activity in the serum, and did not show changes in creatinine concentration of serum and urine. The insignificant levels of factors and enzymes in the serum and renal tissue of camphor treated groups, as compared with the gentamicin group, indicated that the camphor had no effect on biochemical factors and antioxidant enzymes.

Conclusion: Camphor can be useful in the attenuation of gentamicin-induced nephrotoxicity based on the levels of examined biochemical factors and antioxidant enzymes.

Keywords: Gentamicin; Nephrotoxicity; Camphor; Biochemical factors; Antioxidant enzymes

Effect of COVID-19 in Patients with Age-Related Diseases: A Review

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Background and Aim: The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) worldwide pandemic has taken the world by storm since December 2019. Many risk factors are considered to be linked with the development of COVID-19. COVID-19 mortality rates increase with patient age and underlying diseases, making the sick and elderly most vulnerable to severe infection. Patients with preexisting chronic conditions such as diabetes, cardiovascular disease, and chronic kidney disease are highly at risk of respiratory illness due to coronavirus infection. In this study we investigated the potential association of these conditions with SARS-CoV-2.

Results: Age and comorbidities are strong predictors of hospital admission, critical illness, and death rate in COVID-19 patients. Patients suffering from cardiovascular disease, hypertension, diabetes, and chronic kidney disease, are more likely to experience severe symptoms. Hyperinflammatory response induced by COVID-19 is a major indicator of the severity and death rates in infected patients. Chronic inflammation is associated with cardiovascular disease, and type 2 diabetes, and also aging itself.

Conclusion: The novel COVID-19 pandemic has caused Many challenges in health care. As the coronavirus pandemic persists, the medical community will need to continue investigating COVID-19 therapies. Thus far, we know there is a certain relation between age-related diseases and COVID-19. This study was a review of age-related diseases and predisposition to inflammation. Understanding the mechanisms of the relationship between COVID-19, inflammation, and immunity could be helpful in developing appropriate drug therapies.

Keywords: SARS-CoV-2; COVID-19; Age-related disease; Diabetes; Cardiovascular disease.

Effect of Flaxseed on Lipid Profile and Liver Enzymes in An Animal Model of NAFLD

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Background and Aim: Nonalcoholic fatty liver disease is considered the hepatic manifestation of metabolic syndrome and is the most common cause of liver dysfunction worldwide, risen substantially in recent decades, affecting about 25% of the adult population.

Methods: Twenty-four male Wistar rats were divided into three groups (N=8): 1) control, 2) HFD and 3) HFD+ flaxseed. HFD groups received HFD for 15 weeks in order to induce NAFLD. Flaxseed (40g/kg/day) was administered for 8 weeks. After treatment, the animals were anesthetized and the serum level of liver enzymes, lipid profiles, and kidney indices was determined.

Results: TG and CHO in HFD + flaxseed had a significantly decrease compared to the HFD group. The levels of AST, ALT, ALP, and GGT were significantly decreased in the HFD + flaxseed. In contrast, the level of Urea and Creatinine doesn't have significant changes ($P \leq 0.05$).

Conclusion: The data demonstrate the favorable effects of flaxseed in improving liver and functions in NAFLD condition without any side effects of the kidney.

Keywords: Flaxseed; High-Fat Diet; Nonalcoholic Fatty Liver Disease

Effect of Gilbert's Syndrome Associated Polymorphic Alleles (rs8175347 and rs4148323) of UDP-Glucuronyl Transferase on Serum Bilirubin Level

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Background and Aim: The most common mutations in Gilbert's syndrome are associated with the promoter region of the gene (rs8175347) and the codon region in exon1 (rs4148323). This is the comparative study of the bilirubin level with rs8175347 (UGT1A1*28) and rs4148323 (UGT1A1*6) polymorphisms in patients with Gilbert's syndrome.

Methods: The study subjects included 63 individuals with suspected Gilbert's syndrome and 61 healthy individuals. After clinical screening test, rs8175347 and rs4148323 polymorphisms were evaluated using PCR-SSCP, sequencing analysis and PCR-RFLP methods, respectively.

Results: Genotype distribution of rs4148323 polymorphism in the exon1 region was more than that of rs8175347 polymorphism in the population with Gilbert's syndrome ($p < 0.001$), and AA allele in rs4148323 polymorphism had more effect on Gilbert's syndrome in the study population (OR = 9.40, CI = 3.4–25.94). Analysis of serum total bilirubin between two polymorphisms did not find a significant difference between rs8175347 and rs4148323 ($p = 0.93$). Combination of two polymorphisms showed a synergic effect on increasing the serum total bilirubin in this study.

Conclusion: Rs8175347 and rs4148323 had a significant effect on increasing the bilirubin level, and their effect increased when they were combined with each other.

Keywords: Rs8175347; Rs4148323; Polymorphic alleles; UDP-Glucuronyl transferase; Gilbert's syndrome; Bilirubin level

Effect of Hydroxytyrosol on Antioxidant Enzyme Activity in HCT-116 Colorectal Cancer Cell Line

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Background and Aim: Hydroxytyrosol is a powerful antioxidant that has various activities such as controlling oxidative stress, inhibiting cell proliferation and inducing apoptosis. The aim of this study was to investigate the effect of hydroxytyrosol on Nrf2 gene expression and the activity of enzymes catalase, superoxide dismutase and glutathione peroxidase in HCT-116 cell line of human colorectal cancer.

Methods: In this study, For 24 hours HCT-116 cells were treated with different concentrations of hydroxytyrosol, after that the expression level of Nrf2 gene was determined by Real time-PCR and calorimetric methods were used to measure the activity of enzymes catalase, superoxide dismutase and glutathione peroxidase.

Results: The results showed that Hydroxytyrosol can change Nrf2 gene expression also analysis of antioxidant enzyme activity showed that Hydroxytyrosol significantly increased the activity of enzymes of catalase, superoxide dismutase and glutathione peroxidase in the treatment groups compared to the control group.

Conclusion: It seems that Hydroxytyrosol can increase of the activity of antioxidant enzymes through changes in Nrf2 gene expression in HCT-116 cells.

Keywords: HCT-116; Hydroxytyrosol; Antioxidant enzymes

Effect of Saffron Carotenoids on Differentiated-PC12 Cells Treated with Amyloid Beta Oligomers as a Cell Model of Alzheimer's Disease

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Background and Aim: The accumulation of misfolded and aggregated amyloid-beta ($A\beta$) peptides is the main feature of AD that induce some alterations in signaling pathways, including the endoplasmic reticulum stress, which lead to apoptosis in nerve cells and autophagy. In this study, we examined the effect of saffron and its active components (Crocins and Crocetin) on the interlink between these three signaling paths.

Methods: By using $A\beta$ oligomers ($A\beta$ Os), we induced an AD-like condition in dPC12 cells. After treating these cells with either Crocin (Cro) or Crocetin (Crt), the mentioned signaling pathways were studied and compared with the untreated dPC12 cells. Both preventive and therapeutic modalities of saffron carotenoids were considered in this study by applying various methods.

Results: MTT assay indicated the protective effect of both carotenoids on dPC12 cells against $A\beta$ Os. Flow cytometry and Western blotting of PARP protein confirmed that $A\beta$ Os induce apoptosis in dPC12 cells, and both carotenoids suppress this effect. Finally, RT-PCR and Western blotting data indicate that $A\beta$ Os induce XBP1 splicing in both mRNA and protein levels, and Cro/ Crt augmented its splicing, too. The $A\beta$ O-induced eIF2 α phosphorylation was reduced in the presence of Cro/ Crt in a time-dependent manner. Beclin1 and LC3II levels were increased by either Cro or Crt, but LC3II accumulation in the presence of CQ showed that $A\beta$ Os impairs autophagic flux. Interestingly, either Cro or Crt resolves this autophagic defect and increases autophagosome degradation. Application of IRE1 α -XBP1s inhibitor indicated that Cro/Crt-induced autophagy was linked with activation of this branch of UPR in preventive and therapeutic modalities.

Conclusion: In summary, we demonstrate that the protective effect of either Cro or Crt against $A\beta$ Os is dependent on the activation of XBP1, inducing autophagic flux, and reducing the amount of broken PARP. Therefore, the IRE1 α -XBP1s-autophagy axis has been suggested as a novel protective mechanism for either Cro or Crt.

Keywords: Alzheimer's disease (AD); PC12; $A\beta$ oligomer ($A\beta$ O); unfolded protein response (UPR); autophagy; Crocin (Cro); Crocetin (Crt).

Effect of Trehalose Administration on Kidney Function and Renal Histopathological Scores in Aged Rats

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Background and Aim: Aging is a natural phenomenon in which a progressive decrease occurs in the function of various organs, such as kidneys. With age, the glomerular filtration rate (GFR) gradually reduces due to a decrease in the number of functional nephrons. Trehalose is a non-reducing disaccharide which has been shown to protect cells against various cellular damage. In this study, we investigated the effect of oral administration of trehalose on the function and pathological indicators of kidneys in aged rats.

Methods: In this study, 23 male Wistar rats were used which were divided into three groups including the young control group, the old control group, and the old trehalose group. After one month oral administration of 2% trehalose, blood and urine samples were collected and kidneys were removed. Then serum urea and creatinine, as well as urine creatinine and albumin levels, were measured and glomerular filtration rate (GFR) was calculated. Finally, to evaluate pathological indices, the left kidney of mice was used for cross-sectional preparation of tissue, eosin and hematoxylin, and trichrome staining.

Results: According to our data, A significant decrease was observed in GFR, albuminuria and serum urea and creatinine level in old rats treated with trehalose compared to old control animals (pvalue was less than 0.05). Interestingly, the study of pathological indices also demonstrated that administration of trehalose led to a significant reduction in renal inflammation and tubular necrosis in aged rats treated with trehalose compared to control group (pvalue was less than 0.05).

Conclusion: Collectively, these findings showed that treatment with trehalose could ameliorate renal function and inflammation, thus reducing tubular necrosis in aged kidneys.

Keywords: Aging; Trehalose; Kidney function; Pathological indices

Effect of β -Amyloid on the Expression Level of ABCA1 Protein and mir-106b Involved in Cholesterol Homeostasis in Astrocytes Isolated from C57BL/6J Mice Brain

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Background and Aim: Alzheimer's disease is a neurodegenerative disorder with an aberrant accumulation of amyloid-beta and cholesterol in the CNS. During development of CNS, cholesterol is synthesized in astrocytes and transported out of the cell through ApoE-HDL complex and the ABCA1. A β affect cholesterol homeostasis and may lead to changes in the expression levels of ABCA1 and its regulatory microRNAs. In this study, we investigated the effect of A β on this protein and mir-106b in cultured astrocyte cells.

Methods: Astrocyte cells were isolated from the brain of newborn C57BL/6J mice and cultured in a DMEM low glucose medium with 10% FBS. The cells were then treated with 1 μ M beta-amyloid and the expression of ABCA1 and mir-106b were measured by real-time PCR. Online software IRNdb, TargetScan, and RNAhybrid were used for InSilico Assessment.

Results: Amyloid-beta increases ABCA1 expression within astrocytes and the expression level of mir-106b as a negative regulator of ABCA1 showed a significant decrease (**P< .01). The interaction of ABCA1 mRNA with mir-106b was performed by RNAhybrid.

Conclusion: Cholesterol synthesis is altered in the brains of Alzheimer's patients, and the expression of genes and their regulatory miRNAs are also changed. According to the results of this study increased expression of ABCA1, followed by decreased expression of its regulatory mir-106b, may be due to disruption of cholesterol homeostasis in astrocytes which is treated whit amyloid-beta. It can be hypothesized that these cells may have increased the expression of the main cholesterol transporter to remove excess cholesterol, and further research is needed to investigate this hypothesis.

Keywords: Alzheimer's disease; Amyloid-beta; ABCA1; Mir-106b

Effects of *Capparis spinosa* L. Extract on the Serum Levels of Liver Enzymes in Mice

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Background and Aim: Based on the previous investigations on animal models, the liver is one of the main organs in the evaluation of drug toxicity; so, evaluating the function of this organ can be considered as a great method to study the toxicity of some new drug agents. The aim of this study is to evaluate the hepatoprotective effect of methanolic and aqueous extract of *Capparis spinosa* L. fruit in mice.

Methods: The hepatoprotective effects of methanolic extract of *C. spinosa* was assessed by measuring the serum levels of enzymes including bilirubin, alkaline phosphatase (ALP), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) in the treated mice after oral administration of *C. spinosa* at the doses of 250, 500, and 750 µg/kg, respectively, for 14 days.

Results: The obtained findings revealed that after orally administration of *C. spinosa* extract although the serum level of AST, ALT, ALP, and bilirubin was changed in a dose-dependent manner, no significant difference ($p > 0.05$) was observed in the concentration of serum liver enzymes between the treatment (with oral administrations of *C. spinosa* and control groups).

Conclusion: The findings revealed the hepatoprotective effects of methanolic extract of *C. spinosa* in mice. However, other supplementary tests are required to reach this conclusion.

Keywords: *Capparis spinosa*; BALB/c mice; liver; enzymes

Effects of Opium on TGF- β Circulating Levels in Coronary Artery Disease

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Background and Aim: It is suggested that opium may play a potential role in the development and progression of coronary artery disease (CAD) through inflammation and oxidative stress. The aim of the present study was to explore the role of opium on the plasma level of TGF- β in the CAD opium-addicted and non-opium-addicted patients who received routine cardiovascular-related drugs.

Methods: Seventy-seven subjects suspected of CAD were selected as candidates for coronary angiography in this case-control study. They were categorized into three groups including: 1) CAD opium-addicted (CAD+OA, N=30); 2) CAD non-opium-addicted (CAD, N=30); and 3) non-opium-addicted with no CAD individuals as a control group (Ctrl, N=17). The CAD and CAD+OA patients were under treatment with atorvastatin, clopidogrel, aspirin and glyceryl trinitrate. Plasma levels of TGF- β was evaluated by ELISA technique.

Results: Results: The mean plasma levels of TGF- β was significantly lower in the CAD+OA group than CAD patients ($P = 0.020$), whereas no significant difference was found between Ctrl group with CAD+OA and CAD groups ($P > 0.05$).

Conclusion: TGF- β decreased in the CAD+OA patients compared to the CAD, but not in Ctrl group. Since the CAD and CAD+OA patients were under treatment by anti-inflammatory drugs, it could be assumed that the drugs modulate the cytokine expression.

Keywords: Opium; Coronary artery disease; Cytokines; TGF- β .

Effects of *Securigera securidaca* Seed Extract in Combination with Glibenclamide on Antioxidant Capacity, Fibroblast Growth Factor 21 and Insulin Resistance in Hyperglycemic Rats

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Background and Aim: Undesired effects of synthetic antidiabetic agents have made researchers seek safer and healthier resources. With this aspect, herbal materials have attracted substantial research interest and are being extensively investigated. Considering that herb-drug interactions can be a double-edged sword presenting both risks and benefits, investigation of such interactions is greatly in demand. The aim of the study was to investigate possible beneficial effects of hydroalcoholic extract of *S. Securidaca* seed (HESS) on antioxidant capacity, fibroblast growth factor 21 (FGF21), and insulin resistance in Streptozotocin (STZ)-induced diabetic rats, alone and in combination with glibenclamide.

Methods: Forty male Wistar rats were randomly divided into eight equal groups including healthy and diabetic controls and six treated groups with various doses of HESS alone and in combination with glibenclamide, for 35 consecutive days. Serum samples were taken and analyzed for biochemical profile, HOMA indexes, FGF21, oxidative/nitrosative stress, and inflammatory biomarkers as compared with the controls. Moreover, the total phenolic and flavonoid contents of herbal extract were assessed.

Results: Total phenols and flavonoids contents in the hydroalcoholic extract of *S. Securidaca* seeds were 93.3 ± 1.5 mg (GAE) / g (D.W.) and 46 ± 1.7 mg (QE) / g (D.W.) , respectively . Glibenclamide is more effective than the 100 and 200 mg / kg B.W. doses of HESS in increasing insulin and lowering blood sugar (P lower than 0.05) . There were significant differences between all groups in the mean of serum FGF21 levels (P lower than 0.05) . No significant differences were observed in urea and creatinine levels among the groups (P higher than 0.05) . PON1 , SOD , and GPx activities were significantly increased (P lower than 0.05) in treatment groups as a dose - dependent manner , and reached the normalcy in combination with glibenclamide . Increase in HESS dose ameliorated antioxidant power (FRAP) , ROS and MDA levels , and synergistically improved in combination with glibenclamide . Not significantly , both hs - CRP and TNF - α markers were increased in diabetic rats and decreased by an increase in HESS dose .

Conclusion: HESS may ameliorate oxidative stress complications in diabetic cases and may be a good supplement substance in combination with blood sugar-reducing drugs.

Keywords: *Securigera Securidaca*; hydroalcoholic extract; phenolics; diabetes; oxidative stress; antioxidant

Effects of Trehalose on NFE2L2, Catalase, and Superoxide Dismutase in the Kidney of Aged Rats

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Background and Aim: Aging is associated with an increase in oxidative stress, which damages organs such as the kidney. Trehalose has abundant beneficial activities including antioxidative effects. This study aimed to investigate the effects of trehalose on several antioxidant parameters of the aged kidney.

Methods: Wistar rats were divided into three groups: young (4 months), aged (24 months), and aged-trehalose. The third group was treated with 2% trehalose for one month. The expression of target genes and enzyme activities in the kidney of the animals were evaluated by quantitative polymerase chain reaction (qPCR) and enzyme colorimetric procedures, respectively.

Results: Protein levels of NFE2L2 showed a 50% reduction in aged rats compared to young rats ($P < 0.001$), which was restored by trehalose intervention. In addition, the activity and mRNA levels of catalase (CAT) increased in aged rats while treatment with trehalose reversed this trend. On the other hand, superoxide dismutase (SOD) activity was reduced in the kidneys of aged rats but was not affected by trehalose intervention.

Conclusion: It is concluded that trehalose supplementation alleviates the antioxidant system impairments in the kidneys of aged rats. However, further investigations are needed to thoroughly describe the antioxidative impacts of trehalose on the kidneys during aging.

Keywords: Trehalose; Oxidative stress; NFE2L2; SOD; CAT

Elevated Homocysteine, as A Biomarker of Cardiac Injury, in Panic Disorder Patients Due to Oxidative Stress

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Background and Aim: Patients with panic disorder (PD) suffer from elevated oxidative stress as a consequence of serotonin metabolism disorder. These patients have elevated serotonin concentration and catabolism of serotonin via monoamine oxidase. The aim of the present study was to evaluate serum homocysteine concentration and its relationship with oxidative stress level in PD patients, regarding homocysteine as a diagnostic biomarker of heart disease.

Methods: Sixty patients with PD according to the DSM-5 diagnostic criteria for a panic attack and 60 healthy individuals were included in the present study. Peripheral venous blood samples were taken from patients. Erythrocytes and serum were separated from blood, and RBC hemolysates were prepared to investigate oxidative stress indices including glutathione and glutathione peroxidase. Serum homocysteine and carbonyl groups concentrations were measured in all samples. Data were analyzed using ANOVA, and $p < .05$ was considered significant.

Results: Results showed that serum carbonyl groups concentration was significantly higher in patients with PD than in healthy individuals ($p < .001$). The results also indicated decreased serum glutathione concentration and glutathione peroxidase activity in patients ($p < .003$). In addition, elevated homocysteine concentration in PD patients' serum was observed during the present study ($p < .003$).

Conclusion: Our findings support that patient with PD experience higher levels of oxidative stress, due to impaired serotonin metabolism, which is related to the prognosis of heart disease in these patients.

Keywords: Protein Carbonylation; glutathione; homocysteine; panic disorder

Endoplasmic Reticulum Stress in Placenta

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Background and Aim: Recent studies indicated that high levels of endoplasmic reticulum (ER) stress in placenta are related to many gestational disorders, including fetal growth retardation, preterm delivery, and intrauterine growth restriction (IUGR), preeclampsia, and gestational diabetes. It also predisposes the infant to several disorders in adulthood. We will discuss about ER stress and its role in the placenta and gestational disorders.

Results: ER is a vital intracellular organ that plays key roles in various cellular processes such as protein synthesis, folding, post-translational changes, and calcium homeostasis. Disruption of the protein folding process and accumulation of unfolded proteins, as well as disruption of calcium homeostasis in the ER, can lead to ER stress. Various factors such as hypoxia, glucose deprivation, oxidative stress, viral infection, mutations in certain proteins can cause ER stress. ER stress triggers several signaling pathways like PERK leading to phosphorylation of eIF2 α and inhibition of unnecessary protein synthesis, as well as ATF6 leading to increased expression of GRP78, GRP94 and folding capacity of proteins, and IRE1 leading to the activation of XBP-1, degradation of unfolded proteins and stimulating the inflammatory response. The imbalance of ER stress and related signaling pathways can cause various changes in the placenta. Higher levels of ER stress may alter placental morphology, function and impairs fetal growth. Low levels of ER stress can cause a growth-restricted phenotype in the placenta. These results emphasize the complex role of ER stress in placenta during the pregnancy. Explaining the role of ER stress in placenta could be useful for having a safe pregnancy and reducing the chance of disorders in adulthood.

Conclusion: These results emphasize the complex role of ER stress in placenta during the pregnancy. Explaining the role of ER stress in placenta could be useful for having a safe pregnancy and reducing the chance of disorders in adulthood.

Keywords: Endoplasmic reticulum stress; placenta; ER stress

Enhancement of Extracellular L-asparaginase Production from *Sarocladiumstrictum* by Optimization of Enzyme Production through Statistical Experimental Design

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Background and Aim: Utilization of L-asparaginase enzyme as chemotherapeutic agent has been one of the effective strategies for treatment of lymphoblastic leukemia. Since the currently used bacterial L-asparaginase cause side effects due to its antigenicity and L-glutaminase activity search for new sources has been an active field of research.

Methods: This study focuses on isolation and identification of a L-asparaginase productive yeast strain and engineering the culturing media for enzyme production. Among 130 yeast isolates, strain AG18 showed the superior enzyme producer and was identified as a *Sarocladiumstrictum* (HM016901) based on its nucleotide sequence of internal transcribed spacer (ITS) gene. For enhancement of enzyme production, use of two-level factorial design identified yeast extract (the most effective factor with 22.74% contribution), glucose, glycerol, starch and L-asparagine as significantly positive factors among different carbon and nitrogen sources. Then Response Surface methodology was employed in order to optimize the levels of the significant factors, from which 84.5% increase in enzyme production was obtained (from 0.982 to 1.812 IU/ml). Among the variables carbon and nitrogen source, L-asparagine, glucose and yeast extract had significant effects on L-asparaginase production.

Results: The K_m and V_{max} values of the produced enzyme were 9.74 mM and 8.19 $\mu\text{mol/min}$ for hydrolysis of L-asparagine and no activity was observed in the studied range for hydrolysis of L-glutamine. The toxicity of the isolated L-asparaginase was measured in-vitro on K562 cell lines and HL60 cells, and the IC_{50} values were calculated as 0.4 U/ml and 0.5 U/ml respectively.

Conclusion: The lack of significant L-glutaminase activity and promising toxicity properties in the isolated enzyme suggests *Sarocladiumstrictum* to be a new source for L-asparaginase with lower toxicity to normal cells and lower probability for antigenicity and side effect problems

Keywords: L-asparaginase; Media optimization; Response surface methodology; kinetic characterization

Evaluate the Correlation Between the Serum Levels of C1q/TNF-Related Protein-12 and the Severity of Coronary Artery Disease

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Background and Aim: Coronary artery disease (CAD) is the world's largest cause of death. The association of CAD with inflammation is well established. Recently, it has been confirmed that the C1q/TNF-related protein 12 (CTRP12) has a great anti-inflammatory effect. However, few data are available regarding the serum CTRP12 concentration levels in CAD patients. The study was performed to evaluate the correlation between the serum levels of CTRP12 and the CAD severity regarding to the number of affected vessels.

Methods: About 200 suspected CAD patients and 50 healthy ones as a control, were evaluated based on case control study. According to the results of angiography, patients were divided into CAD + (n equal 150) with any major coronary artery stenosis $\geq 50\%$ and CAD- (n equal 50) with less than 50% stenosis of the arteries. The CAD + patients were categorized into one- (1VD), two- (2VD) and three - vessel disease (3VD) based on the number of stenotic vessels. In the current study, different parameters such as CTRP12, tumor necrosis factor α (TNF - α), interleukin - 6 (IL - 6), total oxidant status (TOS), total antioxidant capacity (TAC), and malondialdehyde (MDA) levels were evaluated, and also lipid profiles, hs - CRP and demographic factors were investigated as well.

Results: Data revealed that CTRP12 and TAC levels in CAD + group were significantly lower than control subjects (P less than 0.05). CTRP12 levels were found to be significantly lower in the 3VD compared with 1VD and 2VD subgroups (p less than 0.01 and p less than 0.05, respectively).

Conclusion: Our results confirmed that serum CTRP12 level is inversely associated with CAD severity. Therefore, it may be used as a prediction marker for CAD.

Keywords: Coronary artery disease (CAD); Inflammation; Oxidative stress; Adipokine

Evaluation of Adenosine Deaminase (ADA) Level in Patients with Prostate Cancer

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Background and Aim: Prostate cancer is a developing cancer in prostate gland. It is one of the common cancers among men. This cancer usually develops slowly but sometimes its metastatic form. Screening of prostate cancer is done by measuring the PSA level, but it sometimes leads to overdiagnosis in some cases. Therefore, finding another factor except PSA, for screening the cancer seems necessary. Elevation in the level of ADA is observed in some diseases like cancers. It is considered that measuring ADA level beside PSA level could be a helpful factor for screening prostate cancer.

Methods: In this study, blood sample were collected from 40 patients and 40 control participants matched for age. PSA, free PSA and ADA levels were compared in the serum of both groups. PSA assessment was based on sandwich ELISA although ADA level was measured by Colorimetric methods. SPSS software was used for analyzing data.

Results: According to obtained data from patient and control groups the mean of PSA level was 23.9 ± 11.96 and 1.33 ± 0.61 ng/ml, respectively which showed a significant increase in patients ($P < 0.05$). ADA level was 30.99 ± 0.99 and 9.8 ± 0.8 U/L in patient and control groups. It is significantly higher in patients than controls ($P < 0.05$).

Conclusion: The study results showed a significant elevation in PSA and ADA levels in prostate cancer patients compared with control group. Therefore, serum ADA and PSA levels might be useful indexes in screening and monitoring of prostate cancer.

Keywords: Prostate cancer; Prostate specific antigen (PSA); Adenosine deaminase (ADA); ELISA method; Colorimetric test.

Evaluation of Antioxidant Effect and Cytotoxicity of Different Concentrations of Honey on MCF-7 Breast Cancer Cell Line

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Background and Aim: Background: Honey is one of the most important natural compounds used in traditional medicine to treat many diseases. It has many properties including antioxidant, anti-inflammatory, antimicrobial, anti-cancer, and immune system modulating. In this study, two different kinds of honey-based on total phenolic content (TPC) (high-TPC honey and low-TPC honey) were used to evaluate their antioxidant and cytotoxicity effects in the MCF-7 cell line.

Methods: Materials and Methods: MCF-7 cells were seeded into a 96-well plate (104 cells per well) in RPMI-1640 under 37 °C and 5% CO₂. After 24 hours, cells were treated with different concentrations of honey (0, 3, 6.25, 8, 10, 12.5, 15, 20 and 25 %) for 24 hours. Finally, cell mortality was assessed using MTT colorimetric test. Also, MCF-7 cells were seeded into a 12-well plate (105 cells per well) and treated with honey (0 and 3%) for 24 hours. 2', 7'-dichlorofluorescein diacetate (DCF-DA) was used to measure intracellular reactive oxygen species (ROS). In summary, cells were treated with DCF-DA (10 µM in RPMI) at 37 °C for 45 min. Then, cells were trypsinized and after washing twice with PBS, the fluorescence intensity was measured in the F1 channel using a flow cytometry device (CyFlowCube 6, SysmexPartec, Germany).

Results: Results: The results of the MTT assay showed that the IC₅₀ of high-TPC honey was lower than low-TPC honey on MCF-7 cells (6.25 % and 12.5 %, respectively). The results of flow cytometry also showed that high-TPC honey has more antioxidant effects than low-TPC honey. The percent of fluorescent cells in control, low-TPC, and high-TPC groups were 98%, 95%, and 90.5% respectively.

Conclusion: Discussion and Conclusion: Previous researches have shown that the effect of honey on cancer cells varies in vitro depending on the type of honey and plants used by the bee. This cytotoxicity of honey can be related to phenolic and flavonoid substances or their antioxidant properties. Honey can induce cytotoxicity in a variety of ways including increasing caspase 3, caspase 8, caspase 9, and proapoptotic protein Bax and decreasing anti-apoptotic protein Bcl2. Honey as an antioxidant helps prevent several acute and chronic disorders. The role of oxidative stress involving free radicals in the carcinogenic process is well established. Oxidative protective agents in living organisms are composed of catalase, superoxide dismutase, peroxidase, ascorbic acid, tocopherol, and polyphenols. In addition to these mechanisms, flavonoid compounds also show antioxidant properties. Phenolic acids and flavonoids are responsible for the antioxidant activity of honey. The results of the present study showed that the TPC of honey has an important role in its antioxidant and



cytotoxicity effect. Honey with high TPC has more antioxidant and cytotoxicity effects compared to low TPC honey on the MCF-7 cell line.

Keywords: Keywords; Antioxidant; Breast Cancer; Cytotoxicity; Honey



Evaluation of calcium levels in colorectal cancer.

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Background and Aim: Colorectal cancer is one of the most common cancers diagnosed in both males and females in the worldwide. Due to effective cancer screening measures, the mortality and the incidence rate of colorectal cancer has decreased. Several in vitro and in vivo studies have certified the preventive role of calcium in cancer. Calcium and bile acids protect the mucus membrane via creating insoluble calcium-soaps and inhibition cytotoxic effect of fatty acids in the intestine.

Methods: In this study, blood samples were taken from 50 patients and 50 healthy individuals. The serum was then removed and kept in the freezer until the final test. Calcium level was measured by photometric analysis. The confidence interval was 95% and a P value < 0.05 was considered statistically significant.

Results: A lower level of calcium was found in the serum of CRC patients. The decrease was significant in women over 50 years of age ($P < 0.05$), but it was not significant in women under 50 years of age and men.

Conclusion: According to the results of this study, low calcium levels, especially women in postmenopausal age, could be a pathogenic and prognostic factor in colorectal cancer.

Keywords: Colorectal Cancer; Calcium; screening

Evaluation of The Correlation Between CA15-3, CEA, Estradiol, Vaspin, and Obestatin Parameters Between Saliva and Serum of Women With Breast Cancer

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Background and Aim: To find suitable biomarkers for diagnosis of Breast cancer in serum and saliva; also, to examine the correlation between salivary and serum concentrations of suitable biomarkers.

Methods: This case-control study included 30 women with breast cancer as a case group and 30 healthy women as a matched control group. Blood and saliva specimens were collected from all participants. We evaluated serum and salivary cancer antigen 15-3 (CA15-3), carcinoembryonic antigen (CEA), estradiol, vaspin, and obestatin concentrations. Mann-Whitney U testing and Spearman correlation coefficients were used for statistical analysis.

Results: Serum and salivary concentrations of estradiol were significantly higher in patients with breast cancer (BC) than in healthy women ($P < .05$). Also, serum CEA and salivary obestatin concentrations were significantly higher in BC patients than in the control group ($P < .05$). However, there was no significant difference between other parameters in patients with BC and controls ($P > 0.05$). We observed a positive correlation between serum and salivary concentrations of CA15-3 ($P < 0.05$).

Conclusion: The results of this study demonstrated that concentrations of CEA and estradiol in serum, obestatin in serum and saliva, and estradiol in saliva were significantly different between the 2 groups. The Correlation between serum and salivary concentrations of CA15-3 were positive. This results suggests that saliva could possibly be used as a biological sample. However, more studies are needed.

Keywords: Breast cancer; Saliva; Serum; Vaspin; Obestatin; Estradiol; CA15-3; CEA

Evaluation of The Effect of Different Growth Media on Melatonin Production by *Cryptococcus Uzbekistanis*

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Background and Aim: Melatonin (N-acetyl-5-methoxytryptamine) is a neuro hormone produced in the pineal gland. As well, it is found in animals, plants, fungi and bacteria. Its biological properties are related to the circadian rhythm, reproductive physiology and as an antioxidant via non receptor process. Melatonin is known to help declining the effects of jet lag, especially in intercontinental travel by cultivating the necessary re-set of the body's circadian rhythm. Natural producing melatonin in human depends on the amount of light and also age, indeed, natural melatonin production declines with aging.

Methods: The present study aimed to detect melatonin expression in yeast species. A number of available yeast species were tested. We have found that *Cryptococcus uzbekistanis* produces 46 ppm of melatonin in 35 % Czapek and 1 % tryptophan and 64 % water. Melatonin was assayed with spectrophotometry and high-performance liquid chromatography (HPLC) was utilized in its final detection.

Results: We have clearly shown that melatonin is synthesized during growth of *C. uzbekistanis*. Box-behnken methodology was used to identify significance factors between carbon sources, temperature, time of growth and tryptophan concentration for optimization melatonin producing. Then Response Surface methodology (RSM) was employed in order to optimize the levels of these factors.

Conclusion: This is among few instances that a microorganism is found that produces melatonin and thus microorganisms can be utilized in industrial production of melatonin

Keywords: Melatonin; *C. uzbekistanis*; Czapek; Media engineering; Experimental design method; Box-behnken; RSM; Spectrophotometry; HPLC

Evaluation of The Effect of MG132 Aldehyde Peptide on Induction of Cell death, Apoptosis Pattern, Caspase 3 And 8 Activity and The Amount of Reactive Oxygen Species In Triple-Negative Breast Cancer Cell Line

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Background and Aim: Activation of apoptosis-inducing pathways and cell-mediated signaling pathways has been considered as a new molecular mechanism for controlling cancer cell growth. In this study, the effect of MG132 peptide aldehyde on induction of cell death and activation of apoptosis, the amount of reactive oxygen species in cells, the activity of caspases 3 and 8 and mitochondrial activity in triple-negative breast cell line were investigated.

Methods: The breast cancer cells (MDA-MB-231), were treated with different concentrations of MG132 at different time intervals and the rate of cell death was assessed by MTT assay. The Annexine V-FITC protocol by flow cytometry was used to measure the percentage of apoptotic, necrotic and viable cells. The activity of caspases 3 and 8 was examined by ELISA. To investigate the role of mitochondria, the ability to evaluate mitochondrial membrane potential by JC-1 probe and fluorescence intensity was used. The amount of intracellular ROS was assessed using fluorescence absorbance.

Results: The results of the present study show that treatment of MDA-MB-231 cells with concentrations of 1-10 μM of MG132 shows a significant reduction in the number of living cells. Concentrations of 5 and 10 μM also increased the number of cells in the early stage of apoptosis, which was in MDA-MB-231 cells with an increase in the production of reactive oxygen species at a concentration of 10 μM . Based on the results of the present study, the mitochondrial membrane potential decreased significantly with increasing MG132 concentration, indicating the role of mitochondria in MG132-induced apoptosis. MG132 also increased the activity of caspases 3 and 8 in MDA-MB-231 cells.

Conclusion: The peptide aldehyde MG132 can induce apoptosis in MDA-MB-231 cells in a concentration and time dependent manner by reducing the potential of mitochondrial membranes, activating caspases and increasing the production of reactive oxygen species.

Keywords: MG132; Breast cancer; Apoptosis; Caspase; MDA-MB-231

Evaluation of The Effect of OPRM1 Agonist on Induction of Cell Death, Apoptosis Pattern, Caspase 3 Activity and The Amount of Reactive Oxygen Species In Pituitary Adenoma Cells

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Background and Aim: The pituitary gland is considered as one of the most important gland in the body which control body hemostasis, growth and proliferation. Cellular homeostasis is dependent on the balance between cell death and survival and this balance is controlled by intracellular complexing mechanisms. In this study, the effect of OPRM1 agonist on induction of cell death and activation of apoptosis, cell cycle distribution, the amount of reactive oxygen species in cells and the activity of caspases 3 activity in GH3 cells

Methods: The GH3 cells were treated with different concentrations of OPRM1 agonist at different time intervals and the rate of cell death was assessed by MTT assay. The Annexine V-FITC protocol by flow cytometry was used to measure the percentage of apoptotic, necrotic and viable cells. The activity of caspases 3 was examined by ELISA. The amount of intracellular ROS was assessed using fluorescence absorbance. The cell cycle distribution was assessed using PI protocol and flow cytometry.

Results: Following activation of the OPRM1 receptor, the dose-dependent and time-dependent survival of GH3 cells showed a significant decrease, which was associated with an increase in the percentage of primary apoptosis in the cells. Also increased the percentage of cells in the Sub-G1 phase of the cell cycle, increased ROS production, and increased caspase-3 activity was observed in the GH3 cells. The results showed that simultaneous treatment with the receptor antagonist could restore the percentage of apoptosis, the percentage of cells in the Sub-G1 phase, the amount of ROS and caspase-3 activity induced by the agonist.

Conclusion: The OPRM1 agonist can induce apoptosis in GH3 cells in a concentration and time dependent manner by activating caspases and increasing the production of reactive oxygen species and showed the importance of the OPRM1 receptor in the pathogenesis of pituitary adenomas

Keywords: Pituitary adenomas, OPRM1, apoptosis, cell cycle, caspase, intracellular free radicals

Evaluation Of the Relationship Between Serum Calcium And Magnesium Levels And Coronary Artery Disease

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Background and Aim: Cardiovascular disease are most common reason morbidity and mortality in many countries. Coronary artery disease is most common cardiovascular disease. Serum magnesium is inversely associated to coronary artery syndrome and serum calcium is direct associated to coronary artery syndrome. Many factors have been implicated in the pathogenesis of atherosclerosis, including changes in intracellular concentrations of Calcium, Sodium, potassium, and magnesium. There is a significant inverse correlation between serum magnesium and there is a significant direct correlation between serum calcium incidence of cardiovascular diseases. Magnesium is a mineral with important functions in the body such as antiarrhythmic effect, actions in vascular tone, contractility, glucose metabolism, and insulin homeostasis. In addition, lower concentration of magnesium and higher concentration of calcium are associated with oxidative stress, pre-inflammatory state, endothelial dysfunction, platelet aggregation, insulin resistance, and hyperglycemia. The aim of this study was to examine the cross-sectional association of serum magnesium and calcium levels with coronary artery syndrome in patients that come to cardiac hospital in 2015 year.

Methods: We included 120 subjects (40% women) (50% patient), aged 30-75 years, symptomatic for cardiovascular disease. Coronary arteries obstructions by angiography using the method described. Cross-sectional associations of serum magnesium and calcium with cardiometabolic factors and atherosclerosis, were examined in logistic regression models adjusted for age, sex, smoking status, systolic blood pressure, past history and family history of cardiovascular diseases .We used and analyzing information by SPSS version 18 software.

Results: According to our study, systolic and diastolic blood pressure are lower in patients group .average of group is higher in patients group. 60% of remembers in both of group are men. Serum magnesium level is same in both of groups. Serum calcium level is same in both of groups.

Conclusion: Serum magnesium and calcium are not associated with coronary artery syndrome

Keywords: Calcium; Magnesium; Coronary Artery Disease; Serum

Gallic Acid Exerts Anti-Inflammatory, Anti-Oxidative Stress, And Nephroprotective Effects Against Paraquat-Induced Renal Injury in Male Rats

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Background and Aim: Paraquat (PRQ) is a toxic chemical compound that is very noxious to animals and humans. Gallic acid is a phenolic compound that has antioxidant properties. In this study, we evaluated the ameliorative effect of gallic acid against PRQ-induced renal injury and oxidative stress.

Methods: In this research, the rats were segregated into six groups. Group 1 is the control group; group 2 received paraquat only; group 3 received gallic acid only; and groups 4, 5, and 6 received paraquat plus gallic acid at doses of 25, 50, and 100 mg/kg bw respectively.

Results: Findings of this work displayed that the renal contents of the vitamin C, superoxide dismutase (SOD), and catalase (CAT) significantly reduced and the levels of the serum protein carbonyl, creatinine, serum glutamate pyruvate transaminase (sGPT), urea, serum glutamate oxaloacetate transaminase (sGOT), uric acid, MDA, serum IL-1 β , and the kidney IL-1 β gene expression were remarkably increased in the group receiving PRQ only compared with that in the control group. On the other hand, treatment with gallic acid after exposure to PRQ led to a significant elevation in renal vitamin C, SOD, and CAT levels plus a remarkable decrease in the serum protein carbonyl, creatinine, sGPT, urea, sGOT, uric acid, MDA, IL-1 β , and renal gene expression of IL-1 β in comparison with the PRQ-only-treated rats. Histological changes were also ameliorated by gallic acid administration.

Conclusion: The data approve that gallic acid diminished the deleterious effects of PRQ exposure. In this regard, our results indicated that the administration of gallic acid could alleviate the noxious effects of PRQ on the antioxidant defense system and renal tissue.

Keywords: Gallic acid; Paraquat; PRQ-induced renal injury; Oxidative stress

Genetic Association Analysis of NOS3AS Variants (rs71539868 & rs7830) With Susceptibility to Essential Hypertension

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Background and Aim: Hypertension (HTN) is the leading cause of death in the world, with 13% of annual deaths worldwide following HTN. Primary hypertension is a multifactorial disease in which both environmental and genetic factors play a role. One of the genes recently proposed in connection with HTN is the gene encoding NOS3AS, or sONE. sONE is long non-coding RNA which encodes an antisense to eNOS. The eNOS is a key enzyme in regulating vascular wall homeostasis and its key role in the pathogenesis of hypertension has been well established. The aim of this study was to investigate the relationship between rs71539868, and rs7830 variants of NOS3AS gene with susceptibility to hypertension in the population of Kermanshah province.

Methods: In this case-control study, 110 HTN cases and 110 healthy individuals were enrolled. The genotypes were determined using tetra-primer ARMS PCR method. The results were finally analyzed using SPSS software version 22.

Results: The allelic and genotypic frequency of rs71539868 variant were not significantly different between the two groups. However, in rs7830 variant, the AA genotype was significantly associated with the risk of HTN(OR = 0.42,PV= 0.036).

Conclusion: The rs7830 polymorphism in the NOS3AS gene may play a protective role in preventing hypertension.

Keywords: essential hypertension; LNCRNA; Variant

Higher Mortality Rate in Lung Cancer Patients During COVID-19 Pandemic

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Background and Aim: Coronavirus disease 2019 (COVID-19) is a respiratory tract infection caused by the severe acute respiratory syndrome (SARS-CoV-2). Lung cancer is the most common cancer among males with high mortality rate. Lung cancer Patients may have severe COVID-19 complications. Optimal cancer care during COVID-19 pandemic could be designed if patient-specific and cancer-specific features which affect the severity of COVID 19 are better understood.

Results: Reportedly, cancer patients have a higher risk of infection with SARS-CoV-2 (0.79%), in comparison with non-cancer patients (0.37%) because of their suppressed immune state caused by the malignancy and anticancer treatments. COVID-19 in lung cancer patients (7 out of 12) and patients aged 60 years or older was highly severe (62% hospitalized, 25% mortality). However, this severity only caused a small fraction of lung cancer deaths during pandemic (approximately 11%). COVID-19 severity factors to a large extent are related to patient-specific features such as smoking and chronic obstructive pulmonary disease. On the other hand, Cancer-specific features like prior thoracic surgery/radiation and systemic therapies did effect severity. Antigen super types of Human leukocyte were normally similar in mild or severe COVID-19 cases compared with non-COVID-19 controls. inflammatory monocyte and macrophage activation cause acute lung injury that results in release of pro inflammatory cytokines such as interleukin (IL)-6, IL-1 and tumor necrosis factor- α which have vital role in immune-associated pneumonitis. Thus, these cytokines may be a therapeutic target when T cell is dominant in infiltration or there are signs of increased IL-6 inflammation as in increased C-reactive protein.

Conclusion: The mortality rate in lung cancer patients are higher than normal population. The high severity in lung cancer patients is linked with COVID-19 and features that are patient specific are the most determining factors of severity. A fairly good treatment option could be hydroxychloroquine combined with azithromycin. Of note, it is better to reduce hospital visits as they could increase the risk of nosocomial transmission

Keywords: Lung Cancer; COVID-19; SARS-CoV-2

Improvement of Thermos Stability of Cholesterol Oxidase From *Streptomyces* Sp. SA-COO by Random Mutagenesis

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Background and Aim: Cholesterol oxidase (ChO) (EC 1.1.3.6; Cholesterol: Oxygen oxidoreductase) is a bacterial enzyme dependent on Flavin adenine dinucleotide (FAD) that is industrially important and valuable for the clinical determination of total or free serum cholesterol.

Methods: To enhance the thermal stability of *Streptomyces* Sp. SA-COO cholesterol oxidase, random mutagenesis was used. A random mutant library was generated using two types of error-prone PCR (single step and serial dilution) and two mutants (ChOA-M1 and ChOA-M2) with improved thermos stability were obtained.

Results: The best mutant ChOA-M1 acquired three amino acid substitutions (G49T, W52K, and F62V) and improved thermos stability (at 50°C for 5 h) by 40% and increased the kcat/Km value by 23%. The optimum pH was desirably changed to encompass a broad range from alkali to acid and circular dichroism revealed no significant secondary structure changes in mutants against wild type.

Conclusion: These findings indicated that random mutagenesis was an effective technique for optimizing cholesterol oxidase properties and make a foundation for practical applications of Cholesterol oxidase in clinical diagnosis and industrial fields.

Keywords: Cholesterol oxidase; Error-prone PCR; Thermos stability; Random mutagenesis

In Vitro Activation of Human HSCs with Cholesterol, Palmitic Acid, and Glucose (Three main and abundant ingredients in the diet) to Induce Liver Fibrosis and Compare Their Effects on The Expression of Liver Fibrosis Main Genes

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Background and Aim: Hepatic fibrosis is the result of chronic liver damage and causes by the activation of hepatic stellate cells (HSCs). In liver damage, HSCs are activated and changed to myofibroblasts. Increasing the genes expression, such as beta-converting growth factor (TGF- β), actin-alpha smooth muscle (α -SMA) and collagen1 α (Fibrogenic genes) shows the activation of HSCs which leads to hepatic fibrosis. Intaking much cholesterol, palmitic acid, and glucose have harmful effects on human health but their influence in activating human HSCs has not been studied. **Aim:** In this study, we investigated the effect of cholesterol, palmitic acid, and glucose on human HSCs activation by measuring the mRNA expression level of TGF- β , α -SMA, and collagen1 α genes involved in hepatic fibrosis.

Methods: Methods and Materials The cytotoxic effects of cholesterol, palmitic acid, and glucose on the cell viability of Human HSCs were determined by MTT assay. Then the Cells were incubated in media containing different concentrations of cholesterol, palmitic acid, and glucose for 24 h. At last, the mRNA expression of TGF- β , α -SMA, and collagen1 α genes were measured using real-time PCR

Results: Results Our data indicated that the mRNA expression of TGF- β , α -SMA, and collagen1 α genes was significantly elevated by treating with cholesterol and palmitic acid, but glucose had no effects.

Conclusion: Conclusion The progression of the disease is fatal, and no definitive cure has been found. Therefore, it is important to know the causes of this disease to prevent its development and progression. The results demonstrated that cholesterol and palmitic acid, in high concentrations, could activate HSCs and elevate the mRNA expression level of fibrogenic genes. Thus, controlling fat intaking and knowing its mechanism is crucial to treat and attenuate hepatic fibrosis.

Keywords: Keywords; hepatic fibrosis; Cholesterol; Palmitic acid; Glucose; fibrogenic genes

Increased Salivary and Serum PSA, MiR-193a-5P and MiR-323 Levels Can Be Used as A New Non-Invasive Diagnostic Markers for Early and Accurate Diagnosis of Prostate Neoplasia

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Background and Aim: Prostate cancer (PCa) (malignant type) and benign prostatic hyperplasia (BPH) (benign type) are two types of prostate disease. It is often difficult to distinguish these two diseases from each other and healthy men. At present, prostate-specific antigen (PSA) levels and invasive sampling methods such as biopsy are widely used to prostate disease in the clinic, but they are not sufficient for an accurate early diagnosis and is associated with the suffering of patients. Some miRNAs have a high potential for diagnosing prostate diseases, but no study has been done at the clinical level (in vivo) for diagnostic prostate diseases. On the other hand, saliva, as a biological sample, can be collected noninvasively, repeatedly, and without the use of healthcare personnel. Therefore, the aim of this study was to provide a non-invasive and accurate diagnostic panel based on miRNAs and saliva for patients with prostate cancer and benign prostatic hyperplasia (BPH).

Methods: This case-control study was conducted on 180 men with ages between 45-50 years: 60 PCa patients (newly diagnosed), 60 BPH patients (newly diagnosed) and 60 healthy men from the Hospital Ayatollah Khansari in Arak, Iran. Saliva and serum samples were taken from all patients. The MiR-193a-5P, MiR-323 expression levels and PSA levels in saliva and serum samples were quantified by quantitative real-time PCR and ELISA .T-test analysis was used to compare the two groups, Pearson correlation coefficient analysis was used to evaluate the relationship between saliva and serum and receiver operating characteristic (ROC) analysis was used to evaluate the sensitivity and specificity.

Results: Serum and salivary levels of PSA, MiR-193a-5P and MiR-323 were significantly increased in PCa patients and BPH patients compared to healthy men ($P < 0.001$).Also, Serum and salivary levels of PSA, MiR-193a-5P and MiR-323 were significantly increased in PCa patients compared to BPH patients ($P < 0.001$). Salivary levels of all markers were lower ($P < 0.05$) compared to serum in PCa patients and BPH patients groups. Positive correlation and significant was observed between serum and salivary levels of all markers studied ($P < 0.05$) in PCa patients and BPH patients groups. The ROC analysis of serum and salivary PSA, MiR-193a-5P and MiR-323 in our study showed a good accuracy with good sensitivity and specificity for diagnostic and differentiation of PCa patients from BPH patients.



Conclusion: According to the results of the present study, increased salivary PSA, MiR-193a-5P and MiR-323 levels can be used as a new non-invasive diagnostic markers for early and accurate diagnosis of prostate cancer.

Keywords: MiRNAs; Prostate Cancer (PCa); Benign Prostatic Hyperplasia (BPH); Saliva



Iron chelator or Iron Supplement Consumption in COVID-19? The Role of Iron With Severity Infection

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Background and Aim: Iron is a trace element that is used to replicate the virus and has a role in the vital functions of the body and the host's innate immune system. The mechanism of iron in COVID-19 severity is still not well understood. The aim of this study was to evaluate the association of the iron with COVID-19 severity.

Methods: A case-control study was performed on 147 patients with a positive PCR test result and 39 normal individuals admitted to the Persian Gulf Martyrs Hospital in Bushehr, Iran. The iron profiles and related tests were measured along with hematological analytes.

Results: Hemoglobin (Hb), Fe, and saturated transferrin decreased in all the groups compared to the controls, but ferritin increased in the patient groups. After adjusting for age and sex, we found that increased ferritin levels augmented the odds ratio (OR) of the disease in the moderate (OR = 2.95, P = 0.007), severe (OR = 6.1, P less than 0.001), and critical groups (OR = 8.34, P less than 0.001). The decreased levels of Fe reduced the OR of the disease in the mild (OR = 0.96, P < 0.001), moderate (OR = 0.96, P less than 0.001), severe (OR = 0.95, P less than 0.001), and critical (OR = 0.98, P = 0.001) groups.

Conclusion: It could be concluded that the use of iron chelators to reduce iron intake can be considered a therapeutic goal. In addition, measuring Fe and ferritin is beneficial for the diagnosis of the disease and determining its severity.

Keywords: COVID-19; Fe; ferritin; disease severity; Iron chelator

Is There any Relationship Between Serum Zinc Levels and Angiotensin-Converting Enzyme 2 Gene Expression in Patients with Coronavirus Disease 2019?

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Background and Aim: The level of angiotensin-converting enzyme 2 (ACE2) expression in different tissues is essential in the sensitivity, symptoms and consequences of COVID-19 infection. It seems that zinc is involved in the structure of the ACE2 enzyme has been identified; nonetheless, the relationship between ACE2 expression and zinc serum levels in severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2)-infected patients is still unclear. This study aimed to evaluate the expression of ACE2 in peripheral blood-derived immune cells of COVID-19 patients and its relationship with serum zinc levels.

Methods: Thirty healthy subjects and thirty patients with COVID-19 were enrolled in this study. The COVID-19 infection was confirmed by positive real-time polymerase chain reaction (RT-PCR) and radiological data. Peripheral blood samples were taken from healthy subjects and COVID-19 patients. Whole blood samples were also used to measure ACE2 gene expression by RT-PCR technique. The correlation matrix evaluated the relationship between ACE2 expression, serum zinc levels, and other related variables.

Results: The outcomes showed no considerable alteration in serum zinc levels between patients and the control group. Likewise, the ACE2 gene expression results showed a significant decrease in this receptor's expression in COVID-19 patients compared with the healthy subjects. A significant positive correlation was observed between serum zinc level and ACE2 gene expression in patients with COVID-19.

Conclusion: The immune system seems to reduce the mRNA expression of the ACE2 in the peripheral blood leukocytes following SARS-CoV-2 infection. Moreover, zinc deficiency can make patients more susceptible to SARS-CoV-2 infection.

Keywords: Zinc; ACE2; COVID-19

Isolation and Identification of Anti-Biofilm EPS Producing Isolate

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Background and Aim: Biofilms are surface-attached agglomerations and important virulence factors in microorganisms. Biofilm-forming microorganisms show significantly reduced susceptibility to antibiotics and host defenses. The treatment, inhibition or eradication of infections are complicated in presence of biofilms. Therefore, finding anti-biofilm agents is critical. Lactic acid bacteria (LAB) are promising source of various post biotics like bacteriocins, organic acids, fatty acids, and exopolysaccharides (EPSs). EPSs are widely produced by LAB in dairy products and fermented foods. They possess many bioactivities. In the current study, we aimed to isolate LAB from dairy products and screen them for anti-biofilm EPS production.

Methods: Samples of traditional dairy products were collected from different geographical locations of Damghan. Bacterial isolates were isolated by diluting samples and their culturing on MRS agars. The isolated LAB were screened for EPS production. Anti-biofilm activity of produced EPS (0.195 to 12.5 mg. mL⁻¹) was evaluated against *Staphylococcus aureus*, *Bacillus cereus*, and *Yersinia enterocolitica*. Finally, anti-biofilm EPS producing isolate was molecularly identified.

Results: Among isolated bacteria, 13 isolates were gram positive and catalase negative which considered as LAB. Among them, one isolate showed the ability to produce EPS. This EPS showed most anti-biofilm activity against *Y.enterocolitica* (51.36-24.06%), which was followed by *S.aureus* (49.6-12.33%), and *B.cereus* (27.16-11.66%). Sequencing of 16S rRNA gene showed that EPS producing isolate showed 99.21% similarity to *Enterococcus durans*.

Conclusion: According to the results, EPS derived LAB can be considered as promising anti-biofilm agents with application in food industries.

Keywords: Lactic acid bacteria; Exopolysaccharide; Anti-biofilm

Low Plasma C1q/TNF-Related Protein 13 Levels in Patients with Type 2 Diabetes and Non-Alcoholic Fatty Liver Was Associated with Increased Risk of Disease

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Background and Aim: Recent studies have shown that C1q/TNF-related proteins (CTRPs) are involved in the pathophysiology of metabolic disorders, such as non-alcoholic fatty liver disease (NAFLD) and type 2 diabetes (T2DM). C1q/TNF-related protein 13 (CTRP13), a hormone secreted by adipose tissue, can regulate energy metabolism in peripheral tissues. There is a little information concerning CTRP13 in the context of NAFLD and T2DM. We evaluated the plasma levels of CTRP13 in healthy control and patients with NAFLD, T2DM and NAFLD+T2DM, and also correlations of CTRP13 plasma levels with clinical and subclinical features.

Methods: Circulating CTRP13 was examined in 88 participants (all men) including healthy control (n=20), T2DM patients (n=22), NAFLD patients (n=22) and NAFLD+T2DM patients (n=22). CTRP13 and adiponectin plasma levels were measured by ELISA method.

Results: CTRP13 serum levels were higher in the control group than the other groups (all p less than 0.001). CTRP13 had significant negative correlation with age, SBP, DBP and metabolic factors including BMI, visceral fat, Insulin, HOMA-IR, FBG, TG, AST, ALT, ALP and γ -GT and have a positive with plasma concentration of correlation adiponectin. CTRP13 had a significant inverse correlation with CMT (r equal -0.345) and liver stiffness (LS) (r equal 0.372) (both, p less than 0.001). Lower CTRP13 in patients with T2DM, NAFLD and NAFLD + T2DM was associated with increased risk of the diseases.

Conclusion: Our results suggested that CTRP13 could be an associated factor with NAFLD in patients with and without T2DM

Keywords: CTRPs; NAFLD; Type 2 Diabetes

Mediterranean Diet Is Associated with Improved Total Antioxidant Content of Human Breast Milk and Infant Urine

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Background and Aim: Background: The composition of breast milk (BM) is dynamic and can be influenced by maternal variables like diet and nutritional status. This study examined the association of maternal adherence to the Med Diet and total antioxidant content of BM and infant urine.

Methods: Background: The composition of breast milk (BM) is dynamic and can be influenced by maternal variables like diet and nutritional status. This study examined the association of maternal adherence to the Med Diet and total antioxidant content of BM and infant urine. Methods: We collected 350 BM samples and urine samples from their infants. The dietary intakes of the subjects were recorded using a validated 65 items-food frequency questionnaire (FFQ). The total antioxidant status of the samples was assessed using the ferric reducing/antioxidant power (FRAP), the 1, 1-diphenyl-2-picrylhydrazyl (DPPH), thiobarbituric acid reactive substances (TBARS), and thiol quantification assays. Milk protein, calcium, and triglyceride (TG) were also determined using standard biochemical kits.

Results: Results: Subjects with the highest Med Diet scores are included in the highest tertile due to more consumption of dietary fiber, fruits, vegetables, nuts, legumes, and seeds, low-fat dairy, whole grains, and less consumption of red meat, sweetened beverages, and sodium than those in the first tertile with the lowest Med Diet scores. BM from mothers with the highest tertile of Med Diet contained significantly higher values of DPPH, FRAP, calcium and protein as well as lower TG concentration in BM compared to those in the first tertile.

Conclusion: Conclusion: Our findings revealed that high maternal adherence to the Med Diet can improve BM quality in breastfeeding mothers and thus promote better neonatal development.

Keywords: Mediterranean diet; Breast milk; Antioxidant content; Mothers; Infant urine

Melatonin as A Potential Drug for COVID-19 Patients

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Background and Aim: The coronavirus disease 2019 (COVID-19) has caused many health issues worldwide including death of millions. Since the beginning of the pandemic many conventional drugs have been suggested for treatment of this disease. Melatonin is a naturally occurring tryptophan derivative which has several functions as an anti-inflammatory, anti-oxidant, and anti-viral, making a potential candidate as a drug of choice. Melatonin is synthesized in the pineal gland and immune cells, produced endogenously in small quantities and declines with age. The preclinical data predict that melatonin might inhibit COVID-19 progression. In this study, out of 78 primary articles searched in PubMed and Google Scholar databases from 2017 to 2021, 20 articles with the COVID-19, melatonin, SARS-CoV-2, treatment, physiological effects keywords were selected and studied.

Results: Viruses could cause an eruption of inflammatory cytokines and reactive oxygen species. However, melatonin has been shown to diminish reactive oxygen species damage, cytokine-induced inflammation, and lymphopenia in viral infections like COVID-19 due to its antioxidant characteristic. The data indicated that melatonin impeded programmed cell death caused by COVID-19 and inflammation in the lungs. Since melatonin improves sleep condition and reduces anxiety, it may have positive effect on immune system as sleep and stress are related with the immune system function. Moreover, melatonin may have role in preventing fibrosis which is one of the most dangerous post-COVID complications. Melatonin may also have role in mitigating the potential oxidative stress caused by mechanical ventilation in patients.

Conclusion: Furthermore, there may be connections between melatonin and increased effectiveness of COVID-19 vaccination. These data show the potential use of melatonin in COVID-19 patients even in the first days of diagnosis.

Keywords: COVID-19; Melatonin; SARS-CoV-2; Treatment; Physiological effects

Mesenchymal Stem Cells (MSCs), A New Approach in The Treatment of COVID-19 Disease

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Background and Aim: Exosomes and other extracellular vesicles are recruited by the viruses during infection to deliver the viral structural proteins and genomes to different host cells. The results of the in-depth analysis revealed that the SARS-CoV-2 virus might also recruit this system to spread its essential components to the target cells such as pneumocytes, bronchial, and nasal epithelial cells. Exosomal spread of viral components could also result in COVID-19 relapse and re-infection. The exosomal pathway could also be exploited as a Trojan horse to treat COVID-19 disease. Thus far, several clinical trials were developed to evaluate the efficacy of mesenchymal stem cells (MSCs) and their exosomes (MSCs-Exo) in the management of several viral infections; the foremost COVID-19 disease.

Results: The ability to release specific cytokines, the convenience in gene manipulation and the prolonged survival of the MSCs and MSCs-Exo in the patients made these stem cells a promising tool for the treatment strategies. MSCs could be isolated from different tissues, including adipose tissue, umbilical cord blood (UCB), Wharton's jelly, and bone marrow. Despite the minimal number of COVID-19 patients who were treated with MSCs, it seems that this approach could protect alveolar epithelial cells and also promote lung regeneration. MSCs readily migrate into the lungs and by secreting the anti-inflammatory cytokines halt the SARS-CoV2-mediated tissue damage. MSCs-Exo has also been shown to be safe, improve the clinical status of the patients, and reduce the need for oxygenation.

Conclusion: Although it is early to hazard a conclusion, it seems that MSCs and their derivatives could be a valuable tool for the treatment of COVID-19 disease. Despite the success of the vaccines, it should not be forgotten that it is better to be prepared, especially against the new variant of the SARS-CoV-2 virus, at which mutations might attenuate the efficacy of the vaccines.

Keywords: Exosomes; Extracellular vesicles; SARS-CoV-2 virus; Re-infection; Treatment

Metformin in Combination with Genistein Ameliorates Skeletal Muscle Inflammation in High-Fat Diet Fed c57BL/6 Mice

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Background and Aim: Although the beneficial effects of metformin (MET) and genistein in ameliorating inflammation have been elucidated, their combined impacts on skeletal muscle inflammation have not been clearly understood. This study aimed to examine the possible preventive effect of MET in combination with genistein on skeletal muscle inflammation in a high-fat diet (HFD) fed C57BL/6 mice.

Methods: Fifty C57BL/6 male mice were fed on an HFD for 10 weeks. The mice were categorized into five groups, control, HFD, HFD + MET (0.23%), HFD+ genistein (0.2%), and HFD+ MET + genistein for 12 weeks.

Results: The results showed that treatment with MET and genistein, either alone or in combination, led to reduced weight gain, fasting blood glucose, plasma insulin, HOMA-IR levels, and Area Under the Curves (AUCs) in ipGTT. MET in combination with genistein demonstrated decreasing effect on macrophages infiltration rate compared to genistein and MET groups alone. The expression of iNOS was reduced, whereas the expression of M2 macrophage markers was increased in combined treatment of MET and genistein. Furthermore, MET in combination with genistein reduced the expression of TNF- α , IL-1 β , MCP-1, and IL-6 and increased the expression of IL-10 in comparison with genistein and MET groups alone. Plasma and skeletal muscle triglycerides and intra-myocellular lipid deposition were reversed by treatment with MET and genistein, alone or in combination.

Conclusion: These results imply that the combination therapy of MET and genistein may have therapeutic potential for decreasing obesity-induced skeletal muscle inflammation in the HFD-fed model.

Keywords: Metformin; Skeletal Muscle Inflammation; Genistein; Macrophages; Cytokines

Non-Invasive Techniques to Evaluate Hepatic De Novo Lipogenesis in Patients with Non-Alcoholic Fatty Liver Disease

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Background and Aim: With the widespread epidemic of obesity, obesity-related diseases have caused widespread global problems. Meanwhile, the incidence of hepatic metabolic syndrome, namely non-alcoholic fatty liver disease (NAFLD), has become a major cause of liver failure and liver transplantation. Currently, the standard and approved method of screening for the disease is liver biopsy, which is an invasive procedure and is not repeatable. One source of accumulated lipids in the liver is hepatic de novo lipogenesis (DNL), which appears to play a key role in the pathogenesis of NAFLD. We can use the following three techniques to evaluate hepatic DNL: Stable isotope tracer studies, Lipid profiling, and Indirect calorimetry. Stable isotope tracer studies are a safe method that is widely used to measure DNL, lipid flow, turnover rate, and lipoprotein production and secretion. In lipid profiling, the lipid content of the individual is examined. In disease conditions, lipids are converted to new biomarkers. Some fatty acids decrease or increase only in NASH, while some levels in both NAFLD and NASH change. DNL reactions are energy-intensive; therefore, the indirect calorimetry technique, which is based on measuring substrate consumption and energy to assess the body's total DNL in vivo, is widely used.

Results: The hepatic DNL is one of the key pathways in the spread of this disease. Hence, assessment of the amount and speed of this pathway using labeled isotopes, fatty acid profiling and indirect calorimetry, we can study the disease process.

Conclusion: Stable isotope tracer studies are a safe method that is widely used to measure DNL, lipid flow, turnover rate, and lipoprotein production and secretion. In lipid profiling, the lipid content of the individual is examined. In disease conditions, lipids are converted to new biomarkers. Some fatty acids decrease or increase only in NASH, while some levels in both NAFLD and NASH change. DNL reactions are energy-intensive; therefore, the indirect calorimetry technique, which is based on measuring substrate consumption and energy to assess the body's total DNL in vivo, is widely used. However, due to the few studies that have been done so far, more caution should be exercised in clinical trials. Due to the increasing progress in technology and also in the field of lipidomics, the leading horizon of this path is very clear and the need for more research in this field is felt more than before.

Keywords: DNL; NAFLD; hepatic de novo lipogenesis; Stable isotope tracer studies; Lipid profiling; Indirect calorimetry

Obesogen: From Metabolic Disruption to Obesity

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Background and Aim: Obesity is an important worldwide public health concern that contributes to the increased incidence of multiple chronic disorders such as type 2 diabetes, heart disease, and cancer. It is well-established that obesity is a multifactorial disorder and related to several factors such as metabolic factors, lifestyle, genetic factors, and exposure to various chemicals called “obesogens. We will discuss the obesogens and their mechanism in the pathogenesis of obesity.

Results: Obesogens are chemicals that disrupt the normal body's homeostasis energy. These chemicals are found in various food, baby bottles, toys, plastics, cookware, and cosmetics. They exert their effects by disrupting different metabolic pathways such as activation of PPAR γ /RXR, altering gut microbiome composition, inducing dysbiosis, an imbalance between producing beneficial and harmful metabolites via gut microbiota, dysregulation of signaling pathways pertinent to the regulation of energy homeostasis, interference with hormone pathway, indirect or direct epigenetic modifications, regulation of appetite, fat cell differentiation, and alteration in the process of converting mesenchymal cells to pre-adipocytes and eventually adipocytes.

Conclusion: Explaining the different mechanisms of obesity could be useful for finding new insights for therapeutic strategies and preventing approaches for improving obesity and metabolic abnormalities.

Keywords: Obesogen; obesity; obesogen mechanism

Opium Consumption Does not Affect the Expression of CD9 Tetraspanin in Coronary Artery Disease

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Background and Aim: The genetic deletion of CD9, as a member of the tetraspanin family leads to decreasing in CD36 and reduction in the formation of atherosclerotic plaques. Studies showed that using opium increases oxidative stress and contributes to the development of atherosclerotic plaques. The aim of the present study was to evaluate the effect of opium on the expression of CD9 in the monocytes in coronary artery disease (CAD) patients with and without opium addiction.

Methods: This case-control study was carried out on 77 CAD-suspected individuals. Subjects were divided into three groups: 1) Severe CAD (50%> stenosis) with addiction to opium (CAD plus addict, n=30); 2) Severe CAD (50%> stenosis) with no addiction to opium (CAD, n=30); 3) Normal coronary arteries, with no opium addiction as a control group (Ctrl, n=17). Flow cytometry was performed to determine the expression level of CD9 protein, and mRNA level was evaluated using the real-time PCR technique.

Results: There was no significant difference in CD9 protein and mRNA expression level among the three studied groups ($P > 0.05$ for all comparisons).

Conclusion: Opium did not show a significant effect on the expression of CD9 at the gene and protein levels, hence, the role of opium in the development of cardiovascular diseases in humans may be mediated by other mechanisms.

Keywords: Opium; Coronary artery disease; CD9

Oxidant/Antioxidant Status and PON1 (paraoxonase1) in Patients with Covid19

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Background and Aim: According the studies and evidence suggests that oxidative stress is closely related to the pathogenesis and severity of COVID-19 infection. Therefor we, suggested that to compare total oxidant status (TOS), total antioxidant status (TAS), oxidative stress index (OSI) and PON1 (antioxidant enzyme) levels between COVID-19 patients and health control.

Methods: We recruited 50 patients with COVID-19 infection and 50 health control group with any infection disease such as Influenza and colds. TAS, TOS, OSI and PON1 levels were determined with ELISA (Enzyme-linked immunosorbent assay) compared between the two groups.

Results: The concentration of the PON1 in compare between the two groups, from a statistical point of view in health people was higher than patient group and this was significant (pvalue<0.05), also in the compare of TOS, there was significant differentiation between the two groups, but the level of the TAC and OSI was not significant between the two groups.

Conclusion: In the current study our results shows that the increase in oxidative stress and decrease in PON1 levels in COVID-19-infected patients in compare with control group can be a risk factor for patients with COVID-19-infected. However, more studies are needed to clarify its clinical values.

Keywords: COVID-19; paraoxonase1; oxidative stress

Oxidative Stress as a Master Regulator During Covid-19 Pathogenesis

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Background and Aim: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) causes Corona Virus Disease-19 (COVID-19), an infectious disease that directly affects lung tissues inducing acute respiratory infection. Typical clinical manifestations are including fatigue, fever, nausea/vomiting, cough, loss of smelling sense, and in advanced stages, acute respiratory distress syndrome, severe pneumonia and septic shock are common. according to age and different pathophysiological conditions, mortality risk is estimated approximately in about 5-15%. The main objective of this study is to investigate the possible mechanisms of oxidative stress during COVID-19 pathology.

Results: Oxidative stress is one of the most known mechanisms underlying COVID-19 pathogenesis. The imbalance between oxidants-antioxidant status results in numerous disturbances in various tissues. The free radicals are produced during oxidative stress can be neutralized by antioxidant agents such as vitamin C, E, selenium, zinc, glutathione, antioxidant enzymes and etc. Some studies indicated the capability of SARS-COV-2 in the formation of free radicals. Virus receptor on host cells, angiotensin-converting enzyme 2 (ACE2) receptor, plays a critical role in oxidative stress so that ACE2 down-regulation is contributed to reactive oxygen species (ROS) generation via NADPH oxidase (NOX) activation. Another significant mechanism which can occur in COVID-19-mediated oxidative state is the iron release from red blood cells into the blood stream. During this process, virus attacks to the heme groups, and releases Fe³⁺ ions which can trigger Fenton and Haber–Weiss reactions to generate ROS. These results provide some evidence for putative mechanisms of virus-mediated oxidative stress.

Conclusion: The potential role of oxidative stress during Covid-19 infection recommends the prescription of antioxidants as useful agents to fight against Covid-19 infection.

Keywords: COVID-19; SARS-CoV-2; Oxidative Stress; Antioxidants

Predictors for The Severe Coronavirus Disease 2019 (COVID-19) Infection in Patients with Underlying Liver Disease: A Retrospective Analytical Study in Iran

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Background and Aim: Risk factors for clinical outcomes of COVID-19 pneumonia have not yet been well established in patients with underlying liver diseases. Our study aimed to describe the clinical characteristics and outcomes of COVID-19 infection among patients with underlying liver diseases and determine the risk factors for severe COVID-19 among them.

Methods: In a retrospective analytical study, 1002 patients with confirmed COVID-19 pneumonia were divided into two groups: patients with and without underlying liver diseases. The admission period was from 5 March to 14 May 2020. The prevalence of underlying conditions, Demographic data, clinical parameters, laboratory data, and participants' outcomes were evaluated. Logistic regression was used to estimate the predictive factors.

Results: Eighty-one (8%) of patients had underlying liver diseases. The frequencies of gastrointestinal symptoms such as diarrhea and vomiting were significantly higher among patients with liver diseases (48% vs. 25% and 46.1% vs. 30% respectively, both $P < 0.05$). Moreover, ALT and AST were significantly higher among patients with liver diseases (54.5 ± 45.6 vs. 37.1 ± 28.4 , $P = 0.013$ and 41.4 ± 27.2 vs. 29.2 ± 24.3 , $P = 0.028$, respectively). Additionally, the mortality rate was significantly high in patients with liver disease (12.4% vs. 7%, $P = 0.018$). We also observed that the parameters such as neutrophil to leukocyte ratio [Odds Ratio Adjusted (ORAdj) 1.81, 95% CI 1.21–3.11, $P = 0.011$] and blood group A (ORAdj 1.59, 95% CI 1.15–2.11, $P = 0.001$) were associated with progression of symptoms of COVID-19.

Conclusion: This study's findings demonstrated that the presence of any chronic liver disease is associated with poorer outcomes in a population with COVID-19.

Keywords: coronavirus disease; infection; liver disease

Prevalence and Predictors of Cardiac and Liver Iron Overload in Patients with Thalassemia

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Background and Aim: Iron overload in the heart and liver is very common in patients with thalassemia. Iron cardiomyopathy is a leading cause of death in patients with thalassemia major (TM) dependent on blood transfusions, and MRI (T2 *) can detect preclinical iron overload of the heart. The aim of this study was to evaluate other biochemical and hematological predictors as an alternative and indirect study of iron deposition in myocardial cells and hepatocytes.

Methods: All patients enrolled underwent MRI of the heart and liver to assess iron overload. A total of 62 patients were included in this study. The mean age of patients was 35.7 ± 3.1 years (56.5%) were male and 43.5% were female. 47 patients (75.8%) received regular blood transfusions.

Results: Independent predictors of iron overload from laboratory data were serum ferritin and transaminases for CIO and LIO. No correlation was observed between other biochemical and hematological factors in cardiac and hepatic cirrhosis based on T2 * MRI.

Conclusion: Serum ferritin can be used as a screening tool to rule out CIO and to diagnose LIO. Therefore, serum ferritin measurement can indicate the risk of hepatic cirrhosis in patients with beta thalassemia major.

Keywords: Cardiac hemosiderosis; thalassemia; T2* MRI; Iron overload

Prognostic Value and Utility of NT-proBNP Levels in SARS-CoV-2 Infection

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Background and Aim: Patients with congestive heart failure (CHF) are at a higher risk of being infected with SARSCoV2 an ominous sign because severe respiratory disease can itself cause CHF to decompensation. On the other hand, patients with COVID-19 can get new onset congestive heart failure as retrospective data have been shown from various studies from China and the USA. Increased levels of serum brain natriuretic peptide (BNP) may indicate the contribution of cardiac failure in pulmonary edema frequently seen in COVID-19 patients, which is mainly because of acute respiratory distress syndrome (ARDS). In contrast, some cases of COVID-19 have shown raised BNP levels without significant ventricular failure. Therefore, it is essential to evaluate cardiac contribution during the treatment of pulmonary edema, which may be overlooked. The usefulness of cardiac troponin and NT-pro BNP as prognostic markers in CHF is well recognized. Similarly, biochemical markers of myocardial injury have been associated with higher mortality in SARS-CoV-2 infection suggesting their potential role as a risk stratification tool. Since NT-pro BNP levels may also elevate in pneumonia, the prognostic importance of this biomarker has been evaluated in COVID-19 subjects in various studies.

Results: An NT-pro BNP > 2598 pg/ml had an excellent sensitivity in predicting mortality in patients with a previous diagnosis of HF. (sensitivity 91.7%, specificity 80%). Recent studies confirm that patients with low BNP values are at very low risk of events within six months after discharge. Low BNP values predicted favorable outcomes in patients with severe COVID-19 by the history of heart failure. Increased levels of serum brain natriuretic peptide (BNP) may indicate the contribution of cardiac failure in pulmonary edema frequently seen in COVID 19 patients, which is mainly because of acute respiratory distress syndrome Cardiac involvement in COVID-19 infection appears to significantly adversely impact patient prognosis and survival. The pre-existence of CHF and high cardiac biomarkers like NT-pro BNP and CK-MB levels in COVID-19 patients correlates with worse outcomes. Our study has several key findings; (I) NT-pro BNP levels are independently linked with in-hospital mortality in COVID-19 pneumonia subjects without HF, (II) NT-pro BNP can be utilized as a prognostic biomarker in subjects hospitalized with COVID-19 pneumonia who were free from HF. Moreover, a recent retrospective study suggested a possible direct cardiac injury in COVID-19 patients linking elevated levels of NT-pro BNP levels to mortality.

Conclusion: In conclusion, higher plasma concentrations of BNP or NT-pro BNP are significantly associated with higher disease severity and increased mortality in COVID-19. Additional studies are required to determine whether these cardiac biomarkers can be incorporated into robust predictive tools that further assist with early management and monitoring in this patient group. Therefore, NT-proBNP levels could be used for stratification risk purposes to guide medical decisions if larger studies confirm this finding.



Keywords: B-type natriuretic peptide; COVID-19, heart failure; Mortality



Prognostic Value of Circular RNAs in Digestive System Malignancies: Evidence From a Systematic Review and Meta-Analysis

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Background and Aim: Dysregulation of circular RNAs (circ RNAs) has been reported in many types of cancer. However, the clinico-pathological and prognostic values of in digestive system cancers are still unclear. This meta-analysis was performed to investigate the potential prognostic role of circRNAs in digestive system malignancies.

Methods: An electronic comprehensive literature search was conducted to determine eligible studies reporting the prognostic value circRNAs in digestive system malignancies. Pooled hazard ratios (HRs), odds ratios (ORs), and 95% confidence intervals (CIs) were extracted to evaluate the association between circRNAs and overall survival (OS) and clinico-pathological parameters.

Results: A total of 69 eligible studies were included in this meta-analysis. The results showed a significant association between high expression of oncogenic circRNAs and poor OS (HR=2.16; 95% CI, 1.79- 2.60).

Conclusion: Our meta-results suggest that aberrant expression of circRNAs is might be indicative of poor prognosis and advanced disease in patients with digestive system malignancies. Further investigations are needed to determine the value of circRNAs levels in clinical management of patients.

Keywords: Circular RNA; digestive system cancer; meta-analysis; prognosis; biomarker

Protective Effect of Cinnamic Acid Against Hyperglycemia in Hepg2 Cells

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Background and Aim: Cinnamic acid, a phenyl propanoid acid, has been investigated as a potential alternative therapy for diabetes and its complications in a number of experiments. The cinnamic acid (CA) mechanism as a natural substance in hyperglycemic conditions is not yet fully understood. In this study, we investigate the ability of CA in in HepG2 cells exposed to a high amount of glucose

Methods: In the first stage, the viability of HepG2 cells at different concentrations of glucose and CA was assessed by MTT assay. Oxidative stress markers (CAT, GPx) were measured spectrophotometrically. After RNA extraction, the effect of different concentrations of CA on the expression of inflammatory factors (IL-6) in HepG2 cells was assessed using real-time PCR.

Results: In HepG2 cells, CA increased catalase and glutathione peroxidase activity in a dose-dependent manner in the presence of high glucose concentrations, with the greatest effect seen at a concentration of 75 mg/ml. Furthermore, CA decreased the expression of IL-6 genes in HepG2 cells in the presence of high glucose levels.

Conclusion: The results of our study indicated that CA reduced hyperglycemia-induced complications in HepG2 cells by decreasing IL-6 gene expression and limiting oxidative stress

Keywords: Cinnamic acid; Oxidative stress; Inflammatory factors

Recombinant Highly Antigenic Nucleocapsid Based Protein as a Diagnostic Tool for ELISA of Anti Nucleocapsid Antibody Assay

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Background and Aim: The Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) have four main structural proteins includes: spike glycoprotein, Nucleo capsid phosphoprotein (NP), membrane protein and envelope protein. NP is the most abundant viral phosphoprotein that produced and shed during infection. NP exhibits high immunogenicity and can be detected in serum and other fluids samples during the first two weeks of infection with peak viral shedding around ten days after infection. So Based on these finding and other structural and antigenic studies, we designed a recombinant NP sequence based protein to be used in an immunoassay for detecting the immunoglobulins in patient recovered from the covid-19.

Methods: We aligned the NPs from SARS-CoV and SARS-CoV 2 and selected highly antigenic parts of the sars-cov2 sequence based on Insilco studies. We cloned the gene of target protein in bl21 E.coli, expressed the protein in proper media, purified the product by nickel Immobilized metal affinity chromatography (IMAC) and characterized the product recombinant protein following electrophoresis, Western blotting and ELISA. Finally we used the purified recombinant protein in enzyme-linked immunosorbent assay (ELISA) development of antibody detection in the serum sample of patients, 43 sample from the convalescent patients were compared with 25 sample of the prior to 2019 samples by this homemade ELISA and the results were analyzed .The sensitivity, specificity according to cut-off value of test were measured by ROC curve.

Results: Cloning and expression were verified by PCR and purification was verified by SDS-PAGE and Western blotting. Of 43 clinical samples confirmed to be infected by SARS-CoV-2, 30 were IgG-positive (70%), and of 25 past 2019 samples 19 were IgG-negative (76%) thus the sensitivity and specificity were determined as 70% and 76% respectively in optimum cut-off point by receiver operating characteristic curve (ROC) curve.

Conclusion: In this study an ELISA was developed to detect SARS-CoV-2-NP specific IgG antibodies by using the NP as an abundant and highly immunogenic target. The sensitivity and specificity were determined as 70% and 76% respectively seems to compare with commercial assays but not much better, an issue which may be related to other factors like multi protein coating in ELISA.

Keywords: Recombinant protein; Antigen, Antibody; Nucleocapsid; SARS-CoV 2; ELISA

Regulatory Effects of Various IFN- β for Mutations Therapy on CXC Chemokines CXCL1 and CXCL9 Gene/Protein Levels in Relapsing-Remitting Multiple Sclerosis Patients

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Background and Aim: The multiple sclerosis (MS) is a clinical complicated immune system disorder. The most frequent symptoms of MS are re-current myelin losing in parallel with central nervous system (CNS) inflammation. Chemokines as key members of immune system are involved in the immune responses. This project was aimed to examine and compare the serum levels of CXCL1 and CXCL9 in relapse remitting (RRMS) MS patients, following therapy with IFN- β formulations therapy.

Methods: Clinical specimens were obtained from 50 unrelated healthy controls along with 40 RRMS patients treated with CinnovexTM (CVX, Interferon beta-1a, made in Iran) and Avonex[®] (AVX, Interferon beta-1a, made in USA). Specimens were taken at Special Diseases Department in Ali Ibn Rafsanjan Hospital, Rafsanjan, Iran. The fold changes of CXCL1 and CXCL9 expression genes against β -actin gene were determined by real-time-PCR technique. The expression proteins and serum levels of CXCL1 and CXCL9 measured by ELISA method. Data analyses using ANOVA test.

Results: levels of CXCL1 and CXCL9 were significantly increased and it was more upregulated by AVX and with a lesser extent by CVX.

Conclusion: Results of our study proposed that AVX is much more powerful than CVX in regulation of the immune system and the dose of CVX probably should be increased to achieve an overt therapeutic response in RRMS patients.

Keywords: Avonex[®]; CinnovexTM; CXCL1; CXCL9; Multiple sclerosis

Relationship Between Zinc and Copper in Covid Patients 19

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Background and Aim: The covid-19 disease is the most health-threatening respiratory disease found in 2019 in Wuhan, China. Due to the immunological basis of the disease and the role of copper in the functioning of critical immune cells as well as the preservative effects of zinc in respiratory epithelium, it is hypothesized that elevation in serum levels of essential trace elements such as copper and zinc might be involved in the progression or clinical course of the disease. Thus, the aim of this study was to investigate possible elevated levels of serum copper and zinc in patients with covid-19 and healthy subjects.

Methods: In this case-control study, blood samples from 30 patients (12M and 18F) with covid-19 and 30 healthy age- and sex-matched individuals (12M and 18F) were collected. Sera were separated and copper and zinc levels were determined using colorimetric Biorexfars kits.

Results: There was no difference in mean age between covid-19 patients and control subjects (42.9 ± 3.23 vs 43.8 ± 2.97 years, respectively). The mean age of men (42.2 ± 0.28 years) did not significantly differ from that of women (44.3 ± 0.41 years). Serum copper level was markedly lower in patients (63.87 ± 3.84 ug/dl) compared with the control group (79.21 ± 4.21 ug/dl). Similarly, a significant reduction was observed in serum zinc level in patients (83.54 ± 7.89 ug/dl) compared with control subjects (87.26 ± 6.21 ug/dl).

Conclusion: Findings of this study showed reduced levels of serum copper and zinc in covid-19 patients and therefore, supplementary .copper and zinc might be considered as beneficial therapeutic strategies

Keywords: Copper; COVID-19; Zinc

Role and Mechanism of ACE2 in COVID-19

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Background and Aim: angiotensin-converting enzyme 2 (ACE2) has been recently identified as a receptor for the entry of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) into the cell. also ACE2 by converting angiotensin II to angiotensin1-7 exerts alleviating effect on Coronavirus-induced acute lung injury. actually ACE2 negatively regulated ACE/Ang2/AT1R to alleviate ALI

Results: All studies showed the level of ACE2 on the cell surface decreased after entry SARS-COV2 into host cell and ACE2 negatively regulated ACE/Ang II/AT1R to alleviate ALI and ACE2 with catalytic activity has a protective role in ALI and rhACE2 was found to be capable of binding to the coronavirus directly and the soluble ACE2 ectodomain dose-dependently blocked SARS-CoV2 spike-induced pseudo virus infection of 293T cells

Conclusion: Downregulation of ACE2 after cell infection by SARS-cov-2 exacerbate the symptoms of Acute lung injury therefore interventions targeting ACE2 may improve the treatment of patient with COVID-19 also the level of ACE2 expression can be upregulated by treatment with an ACE inhibitor (ACEI) or AT1R antagonists

Keywords: COVID-19; ACE2

Role of Immunotherapy in The Treatment of COVID-19 Disease

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Background and Aim: Immune-based therapies have found their way in the treatment of a wide range of diseases and recently, they turned into an option for the management of COVID-19 disease. The failure of the anti-viral agents, glucocorticoids, and immunoglobulins in the treatment of the severe cases of COVID-19 disease as well as the partial success of convalescent plasma from patients that have recovered from COVID-19 have suggested that immunotherapies might do well in the treatment of this disease. Recently, the efficacy of natural killer (NK) cells-based therapies, inhibitors of IL-6, JAK inhibitors, mesenchymal stem cell (MSC)-based therapy, and regulatory T (Treg) cell-based therapy against the SARS-CoV-2 virus have been tested in pre-clinical and clinical investigations. MSCs, for example, have shown to not only could control the SARS-CoV-2-mediated cytokine storm but also repair pulmonary tissue damage through secreting several growth factors.

Results: Thus far, several clinical trials are evaluating the effect of MSCs against severe cases of COVID-19. Adoptive therapy with Tregs was also shown to be a valuable approach for restoring the balance of immune responses in COVID-19 patients. By using engineering methods, Tregs could carry markers that facilitate their homing to the lungs, where these cells could shut down the SARS-CoV-2 virus cytokine storm. The early results of the pre-clinical studies suggested that Treg transplantation could prevent lung injury, enhance lung epithelium regeneration, and reduce inflammatory responses.

Conclusion: Taken together, although it is early to suggest whether immunotherapies are beneficial for COVID-19 patients, it seems that manipulating the immune profile of patients could be one of the best approaches to improve SARS-CoV-2 virus-induced lymphopenia and restore inflammatory responses.

Keywords: COVID-19 disease; Immunotherapies; NK cell-based therapies; MSC-based therapies; regulatory T cells.

Screening of A Full Synthetic VHH Antibody Library Against Recombinant Enterokinase Through Phage Display Technology.

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Background and Aim: Mammalian Enterokinase (EC 3.4.21.9) is a duodenal digestive enzyme which has a two-subunit glycoprotein structure and contains a single disulfide bond. Moreover, enterokinase exhibits a remarkable specificity towards the (Asp)₄-Lys sequence inside the protein structures. Hence, it is widely considered as a biotechnological tool in the digestion of fusion proteins and elimination of purification and reporter tags such as poly His, GST, and GFP. In the present study, we aimed to develop novel recombinant monoclonal antibodies against enterokinase enzyme by the screening of a Camelidae VHH library through phage display technology. The isolated antibodies are to be chemically coupled on a chromatographic media as the affinity ligands for purification purposes.

Methods: In this study, we performed the phage display technique through several consecutive biopanning sessions to screen the recombinant nanobodies specific to recombinant enterokinase by the ELISA method. Furthermore, the specificity and affinity of the antibodies against recombinant enterokinase were evaluated using polyclonal phage ELISA. Eventually, the monoclonal phage ELISA has been performed as well.

Results: The specificity and affinity of the selected antibodies were evaluated following the biopanning process. According to data obtained from polyclonal phage ELISA, a distinctively significant difference was observed between the absorbance density of the coated wells and the control ones by about 2 units. In the next step, the eluted phages obtained from round 5 of the biopanning process were used to perform monoclonal phage ELISA and thereby a single VHH-phage clone was isolated which was tightly bound to the surface epitopes of the enterokinase enzyme.

Conclusion: According to the results obtained in this study, the Camelidae VHH library demonstrates efficient specificity and also high binding capacity towards the enterokinase enzyme therefore could act as an effective affinity ligand to be chemically coupled on a chromatographic media for purification purposes.

Keywords: Enterokinase; Phage display; VHH library; Biopanning; Specificity; Phage ELISA

Screening of A Full Synthetic Vhh Antibody Library Against Recombinant Hepatitis B Surface Antigen Through Phage Display Technology

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Background and Aim: Hepatitis B is a potentially life-threatening liver infection caused by the hepatitis B virus (HBV). It is a major global health problem. WHO estimates that 296 million people were living with chronic hepatitis B infection in 2019. The best way to protect against of HBV is vaccination, which various types of vaccines have been produced in different ways. In addition to the use of monoclonal antibodies, the production of recombinant antibodies has developed and because of their advantages, they are a good alternative to conventional antibodies. HBsAg is a component of virus and have been used in vaccine production. Purification of HBsAg is a very important and sensitive step in vaccine production and affects the effectiveness of the vaccine. In the present study, we aimed to develop novel recombinant monoclonal antibodies against HBsAg by the screening of a Camelidae VHH library through phage display technology. The isolated antibodies are to be chemically coupled on a chromatographic media as the affinity ligands for purification purposes.

Methods: In this study, we performed the phage display technique through several consecutive biopanning sessions to screen the recombinant nanobodies specific to recombinant HBsAg by the ELISA method. Furthermore, the specificity and affinity of the antibodies against recombinant HBsAg were evaluated using polyclonal phage ELISA. Eventually, the monoclonal phage ELISA has been performed as well.

Results: The specificity and affinity of the selected antibodies were evaluated following the biopanning process. According to data obtained from polyclonal phage ELISA, a distinctively significant difference was observed between the absorbance density of the coated wells and the control ones by about 2 units. In the next step, the eluted phages obtained from round 5 of the biopanning process were used to perform monoclonal phage ELISA and thereby a single VHH-phage clone was isolated which was tightly bound to the surface epitopes of the HBsAg.

Conclusion: According to the results obtained in this study, the Camelidae VHH library demonstrates efficient specificity and also high binding capacity towards HBsAg; therefore could act as an effective affinity ligand to be chemically coupled on a chromatographic media for purification purposes.

Keywords: phage display; HBsAg ; vhh library

Similarities Between COVID-19 Infection and Rheumatoid Arthritis; Finding a Roadmap to Treat the Disease

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Background and Aim: The role of inflammatory responses in the progression of COVID-19 disease has evolved a hypothesis that perhaps the SARS-CoV-2 virus might exploit the immune system to induce its pathogenesis. Analyzing the profile of secreted cytokines in COVID-19 patients also revealed a similarity between the immune response of the SARS-CoV-2 virus and arthritis rheumatoid. Moreover, the observation of musculoskeletal symptoms in the majority of COVID-19 patients also shed more light on the possible resemblance between the pathogenesis of these diseases. These findings are promising, as it is possible that some of the anti-arthritis treatments could also be used for the management of COVID-19 disease. Although vaccines are the only weapon against the SARS-CoV-2 virus, the emergence of the new variants of the virus necessitates the identification of several therapeutic strategies to prevent the spread of the virus. Among the drugs used in the treatment of arthritis rheumatoid, chloroquine/hydroxychloroquine seems to be a promising option for the treatment of COVID-19 disease.

Results: This agent also showed efficacy against other SARS coronaviruses. Through increasing the endosomal PH and suppression of toll-like receptors (TLRs), chloroquine not only might prevent the fusion of the virus into the target cells but also block the immune responses. The results of the clinical investigations also showed that the administration of chloroquine to COVID-19 patients prevents the exacerbation of pneumonia and reduces the duration of the disease. In addition to chloroquine, IL-1 and IL-6 inhibitors, TNF inhibitors, and Janus kinase (JAK) inhibitors also seem to be effective in controlling COVID-19 disease.

Conclusion: It is believed that JAK inhibitors could prevent receptor-mediated endocytosis of the virus and thereby prevent the involvement of the lungs. Taken together, although there are still mysteries about the SARS-CoV-2 virus, finding similarities between this viral infection and other diseases could help find a roadmap for its management.

Keywords: SARS-CoV-2 virus; Inflammatory responses; Chloroquine; JAK inhibitors; Treatment.

Survey of Royal Jelly Effectiveness on the MDR1 Drug Resistance Gene Expression in PC3 Cell

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Background and Aim: Prostate cancer treatments are unsuccessful for two main reasons, at first several side effects and secondly drug resistance those are induce after treatment, especially after chemotherapy by some agent like doxorubicin. Multi-drug resistant1 (MDR1) is the most operant gene in drug resistance. Royal jelly is one of the natural substance made by honeybees, this product now is a beneficial used material in prevention, treatment and improve patient condition after therapies. The aim of this study was determination of MDR1 expression before and after treatment by doxorubicin. On the other side, characterize royal jelly pretreatment effect in changes of this gene expression after treat by doxorubicin.

Methods: This in-vitro study was performed in 2018, Babol and Shahid Beheshti university of medical sciences. At first PC3 were cultured. Half-maximal inhibitory concentration₅₀ (IC₅₀) of doxorubicin and Royal jelly were determine by 2, 5-diphenyl-2H-tetrazolium bromide (MTT) assay. High performance liquid chromatography (HPLC) was done to check doxorubicin absorption by cells. MDR1 gene expression was evaluated by Real-Time PCR. For analysis, SPSS16 software and analysis of variance (ANOVA) test were used (P-value less than 0.05).

Results: MDR1 did not recognized in PC3 cells before treatment by doxorubicin. After treat by doxorubicin MDR1, significantly increase (P-value less than 0.05). Pretreat by Royal jelly reduces MDR1 gene expression level in PC3 cells (P-value less than 0.05).

Conclusion: This study showed that MDR1 is an important factor in appearance of drug resistance. High expression level of drug resistance gene leads to increase side effects of chemotrophic agents. Solving this issue may improve treatment quality in-patient with cancer, so Royal jelly as a natural substance has a prominent role to make better medical treatment.

Keywords: PC3; Doxorubicin; Royal jelly; MDR1

The Administration of Immunosuppressive Agents Across the COVID-19 Pandemic; the Challenges and The Opportunities

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Background and Aim: As COVID-19 disease spreads around the world, patients with the suppressed immune system were a huge concern. In the early days of the pandemic, many patients discontinued the consumption of immune suppressants; however, this could not last for long, as these agents are necessary for alleviating the disease symptoms. In chronic autoimmune diseases, the sudden shifts in the dose of the drugs could flare the underlying diseases. So, many studies were conducted to evaluate the impact of immunosuppressive agents on the outcome of COVID-19 disease and the response rate to the current treatments. The results of such studies were varied in this case, as some patients who were treated with immunosuppressive agents were successfully recovered from the SARS-CoV-2 virus infection, some developed the more severe forms of the disease, and a group of patients, especially cancer cases, also lost their lives due to viral infection. Even in the same disease, such as rheumatoid arthritis, it seems that the type of immunosuppressive agent might also have an impact on the outcome of COVID-19 disease.

Results: While administration of prednisolone could increase the susceptibility of the individuals to the SARS-CoV-2 virus, another immunosuppressive agent named hydroxychloroquine showed to have protective effects. In another study, it has also been reported that the long-term use of the low dose of methylprednisolone in patients with a history of kidney transplants was associated with the successful recovery from COVID-19 pneumonia. Corticosteroids are another well-known type of immunosuppressive agent in patients with autoimmune diseases that showed to could accelerate the recovery from COVID-19.

Conclusion: Based on these results, it seems that many factors might influence the effect of immune suppressants on COVID-19 outcome and although the prescription of these agents should be taken into consideration if COVID-19 disease was developed, perhaps there is no need for drug discontinuation for the patients.

Keywords: Immunosuppressive agents; COVID-19 disease; SARS-CoV-2 virus; Patient's outcome; Corticosteroids.

The Antioxidant Effect of Allopurinol Bound to GO/CuFe₂O₄/IRMOF-3 Nano-composite and Its Functions

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Background and Aim: Uric acid is the end product of purine metabolism, which is mainly produced by the liver. The liver xanthine oxidase (xanthine dehydrogenase) (XOD) catalyzes the oxidation of hypoxanthine to xanthine and then xanthine to uric acid. Hyperuricemia is one of the risk factors for gout and oxidative stress conditions such as cancer and cardiovascular disease. Allopurinol (Allo) is the most widely used inhibitor of XOD, which is used to treat hyperuricemia. Allo, not only has a rapid clearance but it also causes some side effects. We prepared a new metal-organic framework (MOF) and compared the release rate and side effects of free Allo and encapsulated in nano composite in male Wistar rats.

Methods: Nano scale GO/CuFe₂O₄/IRMOF-3 was synthesized by hydrothermal method and Allo was encapsulated in its pores. The structural properties of nano particles and efficiency of the free and encapsulated drug in addition to the drug release rate were investigated. 48 male Wistar rats weighing approximately 250 g were adopted for one week in a room with a temperature of 25-28 °C without restriction in water and food and then treated in two phases. In the first phase, a 10 % fructose solution was used to induce hyperuricemia. In the second phase, pure drug per kg of animal body weight (or the equivalent weight of drug encapsulated in nano composite) was used to treat the disease.

Results: TGA, FTIR, BET, XRD analyzes confirmed the formation of the MOF and the drug encapsulation. SEM images showed dimensions of nano particles before/after drug loading was 30, 50 nm, respectively. Encapsulation caused the slow release of the drug (88 % of the loaded drug released in four days). Serum uric acid level in the hyperuricemic group receiving encapsulated Allo was significantly reduced.

Conclusion: Encapsulation of Allo in GO/CuFe₂O₄@IRMOF-3 can be employed to increase the efficacy, reduce the side effects, and lower the release rate of the drugs including treating hyperuricemia and oxidative stress conditions.

Keywords: Hyperuricemia; Allopurinol; MOF; Liver/kidney function; Oxidative stress

The Assessment of Hesperidin Effect on Gene Expression of SIRT1 in Cholestatic Rats

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Background and Aim: Bile duct ligation (BDL) is a typical model of biliary cholestasis in rodents. Hesperidin is known as a bioflavonoid that has hepatoprotective effect; however, its underlying mechanisms are not absolutely recognized. Sirtuin 1 (SIRT1), a class III NADH-dependent histone deacetylase, is involved in glucose, lipid, and bile acid metabolism. The expression of this gene may change in cholestasis. Therefore, in present study we assessed the effect of Hesperidin on the expression of this gene in BDL-induced cholestasis.

Methods: In this study, 24 male Wistar rats weighing 200-250 g, were divided into 4 groups (n=6): sham, BDL, BDL + Hesperidin (100 and 200 mg/kg). After 14 days of treatment, the value of mRNA expression of SIRT1 gene was carried out by using quantitative Real time polymerase chain reaction (qRT-PCR).

Results: Bile duct ligation attenuated hepatic mRNA expression of SIRT1 compared to sham. Administration of hesperidin (100 and 200 mg/kg) after BDL significantly increased gene expression of SIRT1 (p. Value less than 0.0001).

Conclusion: Hesperidin increase SIRT1 expression in liver of rats subjected to BDL and could be considered as a hopeful treatment in the therapy of cholestasis.

Keywords: SIRT1; Hesperidin; BDL; Rat

The Association of Insulin-like Growth Factor-1 Receptor (IGF-1R) Protein Expression with Tumor Grade, Metastatic Progression and Recurrence in Patients with Primary Bone Cancers.

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Background and Aim: Primary bone neoplasms are considered as rare and mesenchymal solid tumors which occur in people at all age groups. Bone tissue can undergo changes in cellular behavior and its microenvironment, which in turn can initiate the process of proliferation, tumorigenesis, angiogenesis and metastasis. Despite recent technologies and advances in imaging and laboratory techniques, more investigation is required to understand underlying molecular mechanisms due to the necessity of finding specific diagnostic markers and early detection. This study is aimed to evaluate the expression of insulin-like growth factor-1 receptor (IGF-1R) protein level and its association with tumor pathophysiological features in primary bone cancers.

Methods: This study was conducted by enrolling bone tumor tissue and peripheral blood samples of 90 patients and 30 healthy controls with informed consent. Immunohistochemistry was applied to assess the level of IGF-1R protein expression in tissue samples. Additionally, ELISA and calorimetric methods were used to evaluate the biochemical profile of blood samples. while using unpaired t-test for normal distributed variables and nonparametric Mann–Whitney U test for not normally distributed variables, Side-to-side comparisons were undertaken. The correlation between IGF-1R and the variables (age, tumor size, malignancy, tumor grade, chemotherapy status, metastasis, and recurrence) was evaluated by Spearman's Rho coefficient.

Results: Patients with bone tumors showed increased level of the IGF-1R protein level compared to the normal adjacent bone tissue. The concurrent overexpression of IGF-1R was detected in malignant bone tumors and those patients who received chemotherapy and had high grade, metastasis and recurrence bone tumor. Also, local protein expression of IGF-1R was correlated with malignancy, tumor size, chemotherapy-received history, grade, tumor recurrence and metastasis status.

Conclusion: Our results demonstrate the possible role of IGF-1R as a major component of IGF-axis signaling pathway and a diagnostic local biomarker in patients with primary bone tumors.

Keywords: Receptor, IGF Type 1; Bone Neoplasms; Neoplasms; Neoplasm Metastasis

The Association of Transcript Levels of Proinflammatory Cytokines in Adipose Tissues with Various Adiposity Indices in Women with Obesity

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Background and Aim: To investigate the association of adipose tissue transcript levels of IL-1 β , IL-6, TNF- α , and MCP-1 with various adiposity indices in obese women.

Methods: Real-time PCR was carried out to investigate the mRNA expression level of the mentioned genes in VAT and SAT from all participants.

Results: The results presented higher mRNA levels of IL-6 and MCP-1 in SAT and VAT of obese women, compared to normal-weight women. As well, results showed a positive correlation of IL-6 and MCP-1 with HOMA-IR. Obesity indices including BMI, hip, and WHtR were considerably higher in the obese group in comparison with the control group. More importantly, we observed a positive correlation of mRNA expression of these pro-inflammatory factors in adipose tissues with some obesity indices.

Conclusion: We have shown here that adipose tissue transcript levels of pro-inflammatory cytokines were significantly higher in obese participants than non-obese participants. In obese individuals, this pro inflammatory molecules was significantly correlated with various obesity indices. These results suggest that targeting obesity and adipose tissue could prevent the high expression of cytokine.

Keywords: Adipose Tissue; Pro-inflammatory Cytokines; Adiposity Indices; Anthropometric Index; Obesity

The Critical Role of Competing Endogenous RNAs (CeRNAs) in Neurological Disorders: A systematic Review

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Background and Aim: Neurological disorders (NDs) are regarded as a broad range of diseases affecting the nervous system. These most prevalent diseases have a high rate of mortality all over the world, especially in elderly persons. In the more recent years, new data drove our attention to the crucial function of non-coding RNAs in the pathogenesis of NDs. Interestingly; a recent investigation demonstrated that there are close crosstalk networks among RNA transcripts such as mRNAs, long non-coding RNAs (lncRNAs), circular RNAs (circRNAs), and pseudogenes for regulating each other's expression via competing for shared sequences in microRNAs (miRs). This term is a novel area of ongoing research explored as competing for endogenous RNA (ceRNA) hypothesis. These are novel regulatory molecules involved in a wide range of biological stages and pathological contexts. In fact, the dysregulation of ceRNA networks (ceRNETs) may affect neural development genes and increase neuropathological alterations leading to the progression of NDs. Hence, exactly understanding the correlation of ceRNETs with NDs will open a new window for expanding our knowledge about this area of science, as well as providing a novel avenue for developing diagnostic biomarkers for NDs management. Thus, we will summarize the evidence supporting ceRNETs in the regulation of NDs-related

Methods: We systematically searched the PubMed, google scholar, and Scopus databases to explore the role of CeRNAs in NDs between March 2010 and July 2021. After performing the literature search and review, 200 eligible studies were identified.

Results: Our findings showed that modeling the post-transcriptional regulation of gene expression by miRNAs has been a broadly studied field of research in the last decade. Moreover, several studies have investigated the effect of ceRNET on NDs and demonstrated very exciting results. It is indicated that CeRNAs play an important role in several types of NDs, including Alzheimer's disease, Parkinson's disease, glaucoma, multiple sclerosis, stroke, epilepsy, and glioblastoma.

Conclusion: It seems that researchers should have a comprehensive view of factors affecting ceRNAs activities and the general steps of conducting ceRNA researches in the field of NDs. Of note, although this systematic review revealed the importance of the crosstalk between ceRNET and many types of NDs, the regulation of ceRNA remains unexplored in Huntington's disease (HD). Nevertheless, these findings are still limited and future investigations in this field can result in a better understanding of the previously discovered processes.



Keywords: Neurological disorders; Competing endogenous RNA (CeRNA); Gene expression



The Diagnostic Value of β -Catenin to Detect Invasive and Non-Invasive Functional and Non-Functional Pituitary Adenomas

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Background and Aim: Background and aim: Pituitary adenomas as multifactorial intracranial neoplasms impose a massive burden of morbidity on patients and characterizing the molecular mechanism underlying their pathogenesis has received considerable attention. Despite the appealing role of canonical Wnt/ β -catenin pathway in cancer pathogenesis, their relevance to pituitary adenoma pathogenesis and possible effect in pituitary tumor invasion is debated that is perused in the current study.

Methods: Materials and Methods: The 109 tumor tissues of non-functional, growth-hormone (GH) secreting and prolactin-secreting pituitary adenomas were obtained from patients who were subjected to the endoscopic transnasal transphenoidal surgery (ETSS) at the neurosurgery department of our institute as well as 30 cadaveric healthy pituitary tissues. The anterior epithelial lobe of pituitary with no pathological evidence was collected and the invasive pituitary tumor was determined based on the Knosp classification system. The β -catenin gene and protein level in tissues were determined using Real-Time PCR and immunohistochemistry, respectively. The receiver operating characteristic (ROC) curves and calculation of area under the curve (AUC) were used to determine the diagnostic value of β -catenin.

Results: Results: The over-expression of β -catenin was detected in invasive compared to non-invasive functional and non-functional pituitary tumors and the β -catenin expression was associated with tumor size and invasion. Therefore, the β -catenin gene and protein expression revealed consistency in different types of pituitary tumors also the simultaneous over-expression of β -catenin in local sites of tumors and patients' PBMCs indicates the possible role of β -catenin as a diagnostic marker in pituitary tumors. The correlation of β -catenin expression with patients' age was only detected in patients with non-functional pituitary adenomas. The diagnostic value of β -catenin to distinguish non-functional pituitary adenomas from GH-secreting and prolactinoma was considerable.

Conclusion: Conclusion: Data from the current study provides expression pattern of β -catenin in prevalent pituitary tumors and their association with patients' clinical features which may open up new molecular targets for early diagnosis/follow up of pituitary tumor growth.

Keywords: β -catenin; functional pituitary adenoma; tumor size; tumor invasion; cancer

The Effect of Androgen Sensitivity in Severity of COVID-19 Disease

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Background and Aim: The coronavirus disease 2019 (COVID-19) has turned into a worldwide complication. Since the beginning of the COVID-19 pandemic there has been several reports indicating higher rates of morbidity and mortality in men, suggesting the potential role of androgens in the disease. Steroid hormones like testosterone, dihydrotestosterone, and estrogen which increase in puberty have a significant role on regulation and immune response because of their effect in adaptive and innate immunity using neutrophils and macrophages. Androgens exist in both sexes but higher in males. Estrogen increases immune system while testosterone suppresses it. Thus, there may be a relationship between androgens and COVID-19 infection. In this study, out of 150 primary articles searched in PubMed and Google Scholar databases from 2017 to 2021, 45 articles with Androgen, Sex hormones, COVID-19, TMPRSS2, testosterone keywords were selected and studied.

Results: SARS-CoV2 virus spike proteins use Transmembrane protease serine 2 (TMPRSS2) to enter the host and androgen receptors are promoters for TMPRSS2 transcription. Thus, facilitating entry of virus that results in reducing the viral recognition of neutralizing antibodies and triggering SARS-CoV-2 virus-cell fusion. Androgen sensitivity which is determined by genetic variants of the androgen receptor may have an important role in severity of COVID-19 disease. Genetic variants are related to prostate cancer, benign prostatic hyperplasia, and androgenetic alopecia may determine host susceptibility. increased susceptibility of males to COVID-19 may be explained by TMPRSS2 expressed in lung tissue, controlled by androgen-regulated gene which is mostly expressed in prostate. Furthermore, angiotensin-converting enzyme 2 (ACE-2) pathway is affected by androgens and behaves like a receptor for SARS-CoV-2 and facilitates virus entry into host.

Conclusion: The difference in mortality rate between genders and pre pubertal Children may be explained by TMPRSS2-mediated actions of androgens in SARS-CoV2 infection. The seriousness of COVID-19 disease could be directly under the influence of androgen sensitivity.

Keywords: Androgen; Sex hormones; COVID-19; TMPRSS2; Testosterone

The Effect of Capsaicin on The Expression of Genes Affecting Apoptosis in LS-180 Colorectal Cancer Cell Line

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Background and Aim: Capsaicin is a phenolic compound that causes the pungent taste of red pepper and has antioxidant, antimicrobial and anti-cancer properties. Several studies have shown its anti-tumor potential of Capsacin. Therefore, the aim of this study was to investigate the effect of this substance on the expression of genes effective in apoptosis in LS-180 cell line.

Methods: In this study, after 24 hours of treatment of human colorectal cancer cells (LS-180) with different concentrations of capsaicin, the expression of genes affecting apoptosis (Bax, Bcl2, Caspase3) were determined by real time-PCR.

Results: The results of study showed that capsaicin can increase the expression of Bax, Bcl2, Caspase 3 in LS-180 cells.

Conclusion: According to the results of this study, it seems that the anti-cancer effect of capsaicin is through induction of apoptosis and increased expression of genes effective in this process.

Keywords: Capsaicin; Apoptosis; LS-180

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The Effect of Crocin on Serum Insulin Levels, Insulin Resistance and Histopathological Changes in The Pancreas of Diabetic Rats

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Background and Aim: Diabetes mellitus (DM) is one of the most common endocrine diseases. Diabetes causes damage to other organs in the body, including the pancreas, by creating free radicals. The aim of this study was to evaluate the effect of crocin separately and in combination with losartan on serum insulin levels, insulin resistance, and histopathological changes in the pancreas in an experimental model of diabetes.

Methods: Male Wistar rats were treated with a dose of streptozotocin (STZ) (50 mg/kg/IV). After 4 weeks of STZ injection, the animals were randomly divided into 5 groups (N=8): healthy, diabetic, DM+Crocin, DM+Losartan, DM+ Losartan-Crocin. The treated animals received crocin (50 mg/kg) and losartan (25 mg/kg) orally for four weeks, and at the end of the eighth week the animals were sacrificed and Serum samples (for measuring FBS and insulin) and pancreatic tissue (for histopathology) were collected.

Results: In the diabetic group, compared to the healthy group, FBS and HOMA IR increased significantly ($P = 0.001$ and $P = 0.002$, respectively) and serum insulin levels decreased significantly ($P = 0.001$). The results showed that four weeks after crocin and losartan administration, the levels of FBS and HOMA-IR in the diabetic group treated with crocin were significantly reduced compared to the diabetic group ($P = 0.001$ and $P = 0.02$, respectively). While serum insulin levels in the group treated with losartan-crocin increased significantly compared to the diabetic group ($P = 0.001$). The results of pancreatic tissue histopathology showed that in the diabetic group compared to the healthy group, severe atrophy occurred in the islets of Langerhans, while in the treated groups compared to the diabetic group, the atrophy of the islets of Langerhans decreased. In addition, pancreatic tissue histopathology showed that losartan-crocin performed better than crocin and losartan in reducing the atrophy of the islets of Langerhans.

Conclusion: The findings of the present study show that the protective effects of crocin on the complications of diabetes are due to its effect on improving β -cells of the islets of Langerhans and reducing insulin resistance, which leads to its hypoglycemic effects. We have also shown that crocin in combination with losartan improves the effectiveness of losartan against diabetes and its complications. In conclusion, we suggest that the combination of crocin with other chemical drugs could be a promising therapeutic agent in the treatment of diabetes complications. However, more human studies are needed to confirm this.

Keywords: Diabetes; Crocin; Losartan; Insulin; HOMA-IR; Pancreas

The Effect of Flaxseed Oil on Improving Biochemical Parameters and Liver Tissue Indices in An Animal Model of Non-Alcoholic Fatty Liver

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Background and Aim: Non-alcoholic fatty liver disease (NAFLD) is one of the most common forms of chronic liver disease worldwide due to changes in the lifestyle of people. NAFLD is one of the current public health problems. Flaxseed oil is one of the richest vegetarian sources of α -Linolenic acid. Omega-3 fatty acids have demonstrated multiple impacts on human diseases as well as in animal models.

Methods: Twenty-four male Wistar rats were divided into three groups (N=8): 1) control, 2) HFD and 3) HFD+ flaxseed oil. HFD groups received HFD for 15 weeks in order to induce NAFLD. After confirmation of induction of NAFLD model by pathological evaluation, flaxseed oil (40mg/kg/day; gavage) was administered for 8 weeks. After treatment, the animals were anesthetized with ketamine and Xylazine and the serum level of liver enzymes and lipid profiles were determined.

Results: TG and CHO in HFD had a significant increase and LDL level decreased compared to the control group. The levels of AST and ALP were significantly increased in the HFD. Also, TG, CHO, Liver enzymes, and FBS levels were reduced significantly in HFD + flaxseed oil compared to HFD ($P \leq 0.05$). The results of liver tissue pathology indicate that in HFD + flaxseed oil group, vacuolar degeneration, congestion, and dilation of sinusoids have improved. Focal necrosis, lobular inflammation, and NAS indices were reduced.

Conclusion: Flaxseed oil controls lipid profile and liver enzymes. Therefore, it can be stated with caution that flaxseed oil can be used to improve lipid profile and reduce the complications of NAFLD.

Keywords: Flaxseed Oil; High-Fat Diet; Nonalcoholic Fatty Liver Disease

The Effect of Heat-Killed *Lactobacillus Plantarum* on The Testis and Sperm Parameters of Bile Duct-Ligated rats

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Background and Aim: Liver plays important role in maintaining endocrine homeostasis, therefore one of the liver fibrosis complications is male reproductive dysfunction. *Lactobacillus plantarum* (L. plantarum) was demonstrated to have probiotic behavior and functions, including antioxidant and anti-inflammatory activity. The present study was carried out to investigate the protection effect of heat-killed L. plantarum on the cholestasis induced male reproductive dysfunction.

Methods: Cholestatic liver injury was produced by ligation common bile duct in male wistar rats. The study included 4 groups (n=8). Two groups were bile duct ligated (BDL) and received heat-killed L. plantarum (3×10^9 cfu/ml) or vehicle. One group was sham-operated and one group included normal control. L. plantarum was orally administrated using gastric gavage for a week prior to BDL and 21 days after BDL. At the end of treatment, the rats were sacrificed, blood was collected and subjected to evaluation of sexual hormones by Elisa kit. Also, epididymis was isolated from the testis and sperm concentration, the percentage of live and abnormal sperm were evaluated. Also, testicular tissue was assayed for antioxidant parameters Such as total antioxidant capacity (TAC), Nitric oxide (NO), Glutathione (GSH), Malondialdehyde (MDA), Catalase (CAT) and Superoxide dismutase (SOD) Activity.

Results: Liver fibrosis caused significantly increase sperm abnormality and decrease live sperm, sperm concentration and LH and testosterone level in serum ($p \leq 0.05$). Heat Killed L. plantarum administration improved Sperm abnormality and sexual hormone level such as testosterone and LH in comparison to BDL group ($P \leq 0.05$). Also, total antioxidant capacity was higher and Glutathione was lower in BDL+ Heat-killed L. plantarum group when compared to BDL group ($P \leq 0.05$). Other antioxidant parameters, sperm parameters and FSH level didn't significantly Changed Compared to BDL group ($P \geq 0.05$).

Conclusion: Our results indicate that heat-killed L. plantarum has potential for use in the context of infertility on liver fibrosis.

Keywords: infertility; *Lactobacillus plantarum*; liver fibrosis; testis

The Effect of Hepatobel Capsule on Fatty Liver Indexes in Patients with Non-Alcoholic Fatty Liver Disease

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Background and Aim: Non-alcoholic fatty liver disease (NAFLD) is rapidly becoming the most common liver disease worldwide. The prevalence of NAFLD in the general population of western countries is 34-45% and in Iran is 21.5-31.5%. Today, the use of medicinal plants instead of synthetic drugs is increasing, due to their safety. Therefore, we designed a randomized controlled clinical trial to evaluate the effects of hepatobel Capsule (Made from a combination of Cichorium intybus and Portulaca oleracea) supplementation on fatty liver indexes (FLI: fatty liver index, HSI: hepatic steatosis index) in patients with NAFLD.

Methods: This randomized, double-blind, placebo- controlled trial was performed on 60 subjects with, aged 20–40 years old. NAFLD patients were randomly divided into two groups. Intervention group received three hepatobel capsule (H. Capsule) a day for 6 weeks ($n = 30$), and the placebo group was given placebo capsule for the same time duration and dosage ($n = 30$). FLI and HIS were used to diagnose fatty liver, grouping of participants, and evaluate the effect of the H. Capsule. FLI and HIS were. TG, GGT, ALT and AST were measured using spectrophotometric methods. T-test analysis was used to analyze the obtained data.

Results: As showed in table-1, the results showed that the H. Capsule supplementation significantly improved the TG, GGT, ALT and AST, compared with placebo group (P less than 0.001). As showed in table-2, the results showed that the H. Capsule supplementation significantly improved the FLI and HIS, compared with placebo group (P less than 0.001). Also, the results showed that H. Capsule had no significant effect on BMI in H. Capsule group (25.4 ± 2.2) compared to the placebo (26.5 ± 1.3) group for 6 weeks (P greater than 0.05).

Conclusion: According to the results of this study, H. Capsule supplementation can be effective as an adjunct therapy to improve NAFLD.

Keywords: Non-alcoholic fatty liver disease; hepatobel Capsule; supplementation; fatty liver index; hepatic steatosis index intensity

The Effect of Homocysteine on CD28 Costimulatory Receptor Exposure and IL-2, TNF- α and INF- γ Release from CD4+ T Cells

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Background and Aim: There is evidence suggesting that Hyperhomocysteineemia accelerates cardiovascular disease development by modulating CD4+ T cell functions. However, the effects of mild, moderate and severe hyperhomocysteinemia on induction of functional characteristics relevant to the development of cardiovascular disease in CD4+ T cells is not fully understood. Therefore, at the present study, our aim was to examine the direct effects of Hcys on CD4+ T cells.

Methods: Untouched CD4+ T cells were purified from peripheral blood, using a negative selection strategy. The purified CD4+ T cells were treated with different concentrations of Hcys (6.25, 12.5, 25, 50, 75 and 100 μ M) for 24 hours after activation.

Results: Lactate dehydrogenase assay and caspase-3 activity measurement showed that Hcys at concentrations $\geq 12.5 \mu$ M had cytotoxic effects toward CD4+ T cells in a concentration-dependent manner. However, at concentration $\leq 6.25 \mu$ M, Hcys promotes CD4+ T cells proliferation via IL-2 independent pathway. Also, at concentrations $\geq 12.5 \mu$ M, Hcys significantly enhanced the release of IL-2, INF- γ and TNF- α from activated CD4+ T cells. Hcys does not have an effect on CD28 exposure in resting CD4+ T cells, however, the down-regulation of CD28 after activation was significantly decreased in Hcys treated CD4+ T cells compared to controls.

Conclusion: Our results indicate that severe hyperhomocysteinemia induced apoptotic cell death in CD4+ T cells, however, mild and moderate hyperhomocysteinemia significantly increase atherogenic cytokines release from activated CD4+ CD28+ T cells possibly by prolonging CD28+ exposure time. This novel finding may improve our understanding of the mechanism whereby Hcys impaired pro-inflammatory cytokine release.

Keywords: Homocysteine; CD4+; atherosclerosis; cytokine

The Effect of L-Lysine Amino Acid on Biochemical Factors and Heat Shock Protein 70 (HSP-70) Concentration in Rats with Diabetic Nephropathy

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Background and Aim: Millions of people around the world suffer from diabetes mellitus. Diabetic nephropathy (DN) is a micro vascular complication in diabetic patients and a common cause of end-stage renal disease (ESRD). The aim of this study was to investigate effect of L-Lysine Amino acid on metabolic parameters and diabetic nephropathy in rat.

Methods: In this experimental study, 24 male rats were randomly divided into three groups of eight (first group: healthy control, second group: diabetes control, third group: diabetic rats + L-lysine 0.15%). Diabetes was induced by i.p injection of streptozotocin at a dose of 55 mg/kg. After intervention, serum levels of glycemic indexes, renal parameters in renal tissue homogenates and histopathology examination were performed.

Results: The results showed that the treatment of amino acid L-lysine for 8 weeks caused a significant reduction in fasting blood glucose (FBS) indices, rat weight, AGEs, TG, LDL-c, total cholesterol, serum creatinine, GFR, BUN and urinary micro albumin levels ($P \leq 0.05$). Furthermore, a significant increase in HDL-c level, serum insulin level was observed between the study group in compared to the diabetes control group ($P \leq 0.05$). HbA1C didn't show any significant relationship between the treated and control group of diabetes ($P \geq 0.05$). In terms of the effect on antioxidant indices in serum and kidney tissue homogenate, it was observed that treated group caused a significant decrease in nitric oxide, MDA, carbonyl protein and caused a significant increase in HSP-70, GSH, SOD, Catalase and TAC compared to the diabetes control group ($P \leq 0.05$). Pathological findings also showed that induction of diabetes in rats increased mesenchymal matrix and swelling of glomeruli and urinary tubes.

Conclusion: The results showed that L-lysine amino acid treatment for 8 weeks may play an effective role in preventing and slowing the progression of diabetic nephropathy.

Keywords: Diabetic nephropathy; L-lysine; Oxidative stress; Heat shock protein.

The Effect of Metformin on The Metabolism of High Glucose-Induced Human Vascular Smooth Muscle Cells

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Background and Aim: Metformin is commonly used to lower blood glucose levels in type 2 diabetic patients. The purpose of this study was to investigate the effects of metformin on the metabolism of vascular smooth muscle cells under high glucose conditions.

Methods: The vascular smooth muscle cells were cultured in DMEM F12 and high glucose (25 mM) for 24 hours. MTT method was used to determine cellular viability. The cells were treated with metformin in values of 1mM, 5mM, and 7 mM. Biochemical parameters including lactate, glucose, total protein, creatinine, and triglyceride were measured in the cell culture after 24 hours. The oil red o staining method was used to stain the lipids in the cells.

Results: Metformin reduced dose-dependently the cellular growth of VSMCs (P-value less than 0.001). The cellular lipid deposition did not change with the increase of glucose uptake. Other biochemical parameters such as lactate, triglyceride, total protein, and creatinine, changed significantly in cellular culture (P-value less than 0.05). Other biochemical parameters such as cholesterol, lactate dehydrogenase, phosphorus, and uric acid did not show significant differences between the control group and different groups treated with metformin (P-value greater than 0.05).

Conclusion: Metformin can increase glucose uptake impacting metabolic pathways. It increased the lactate secretion, protein metabolism without the change in cellular lipid deposition in high glucose conditions.

Keywords: Metformin; Metabolism; Vascular smooth muscle cell; High-glucose

The Effect of Nigella Sativa (NS), Date Palm Pollen (DPP), Vitamin D and Omega 3 as Dietary Supplements on Blood Biochemical Markers in Mice

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Background and Aim: Infertility is one of the major problems among young couples can occur for a variety of reasons. Numerous findings showed that reactive oxygen species (ROS) can strongly affect various sperm parameters. Research showed that the use of antioxidants such as Nigella sativa (NS), Date palm pollen (DPP), vitamin D and omega 3 can be effective in reducing the damage caused by free radicals. The aim of this study was to investigate the effects of mentioned supplements on blood biochemical, inflammatory factor and sex hormone levels in male mice.

Methods: Animals were divided to 2 groups (6 rats in each group) as follows: control group that received saline solution /kg and group 2 that administrated combination of NS (200mg/Kg), DPP (200mg/Kg), and vitamin D (1.5 IU /g) and omega-3 (300mg/Kg). All solutions were administrated during 7 weeks via a gavage tube. Blood samples are taken from the hearts of rats to measure laboratory markers, mineral element, sex hormone and inflammatory factor of C-reactive protein (CRP) using an ELISA kit.

Results: The comparative plasma biochemical parameters (Triglyceride, Cholesterol, HDL, LDL, Urea, Creatinine, Total Protein, AST, ALT, LDH and CRP), minerals (Calcium, Phosphorus, Sodium and Potassium) and sexual hormone (Testosterone) were shown no significant different between control and experimental group except in FBS level that significantly reduced in treatment group and p value amount was less than 0.05.

Conclusion: The results showed that the combination therapy oh herbal medicine and vitamin has no side effects on body organs and blood markers

Keywords: Supplements; Laboratory markers; Oxidative stress.

The Effect of Platelet-Rich Plasma on The Osteoblastic Differentiation of Human Adipose Tissue-Derived Mesenchymal Stromal Cells

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Background and Aim: Mesenchymal stem cells (MSCs) are cell populations that have the potential to proliferate and differentiate. The process of differentiating stem cells from pluripotent to bone cells requires general changes in the pattern of gene expression in them, the most well-known of which are changes in miRNA-dependent settings. Platelet-enriched plasma (PRP) releases growth factors that are mitogenic to mesenchymal cells and can accelerate the process of osteogenic differentiation. The aim of this study was to investigate the effect of PRP on the expression changes of Let-7a, mir-27a and mir-106a during osteogenic differentiation.

Methods: MSC cells were extracted from adipose tissue resulting from abdominoplasty and evaluated by flow cytometry. PRP was prepared from healthy volunteers by 2-step centrifugation. The effect of PRP 10% on the process of osteogenic differentiation was investigated by measuring alkaline phosphatase activity and calcium deposition. As well as, the expression of Let-7a, mir-27a and mir-106a Was evaluated using Real Time PCR technique.

Results: Statistically, the use of PRP resulted in a significant increase in alkaline phosphatase activity and calcium deposition. The increase in let-7a expression was significant on the 14th day compared to the 3rd day. Mir-27a expression in the PRP10% group increased significantly on the 3rd days. Mir-106a expression showed a significant decreasing tendency between days 3 and 14 in a time-dependent pattern.

Conclusion: These findings indicate that PRP accelerates the process of differentiation into bone. Platelet-rich plasma as a biological catalyst showed a clear and different effect on the miRNA regulating bone differentiation of human mesenchymal cells. However, more research is needed to apply PRP, especially in the clinical field.

Keywords: Mesenchymal stem cells; Platelet-enriched plasma; miRNA

The Effect of Silymarin on Nrf2 Gene Expression and Antioxidant Enzyme Activity in HCT-116 Colorectal Cancer Cell Line

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Background and Aim: Silymarin is a natural polyphenolic compound in the thistle plant that has different properties. The aim of this study was to evaluate the effect of this substance on Nrf2 gene expression and the activity of antioxidant enzymes superoxide dismutase, catalase and glutathione peroxidase in HCT-116 cell line of colorectal cancer

Methods: In this study, at first HCT-116 cells were treated with different concentrations of silymarin for 24 hours, after that the level of Nrf2 gene expression was determined by Real time-PCR and the activity of antioxidant enzymes were calculated by calorimetric methods.

Results: Analysis of gene expression results showed that silymarin treatment significantly increased in Nrf2 gene expression and the activity of superoxide dismutase, catalase and glutathione peroxidase in the study cell compared to the control group.

Conclusion: It seems that silymarin increases the activity of antioxidant enzymes in colorectal cancer cells by increasing the expression of Nrf2 gene.

Keywords: Silymarin; Antioxidant enzymes; Nrf2; HCT-116

The Effects of Carvacrol on VCAM-1 Gene Expression in Experimental Autoimmune Encephalomyelitis (EAE) Rat Models

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Background and Aim: Multiple sclerosis (MS) is an autoimmune disease of the central nervous system (CNS). MS causes events including axonal destruction, demyelination of nerve cells, disruption of the blood-brain barrier, infiltration of macrophages, lymphocytes, and also nerve inflammation. Various studies have also shown that angiogenesis mechanisms are of particular importance in MS (1-4). Moreover, various studies have shown that angiogenesis mechanisms are of particular importance in multiple sclerosis (5-7). Experimental autoimmune encephalomyelitis (EAE) is an experimental model of MS. Nowadays, there are many studies in which compounds with anti-inflammatory and antioxidant properties have been identified as effective drugs in preventing and improving neurological disorders (8). Based on these results, in this study, we investigated the effects of carvacrol on the expression of the VCAM-1 gene affecting angiogenesis in rats of the EAE rat models.

Methods: In order to study the effect of carvacrol on the expression of effective genes in angiogenesis including VCAM-1, we used, 24 rat's spinal cord tissue belonging to three groups (n = 8): Healthy control, EAE model and EAE treated with a concentration of 25 mg/kg of carvacrol for 17 days obtained in Lorestan University of Medical Sciences. In this study, we evaluate the VCAM-1 gene expression using the Real-Time PCR technique.

Results: Our study showed that the gene expression level of VCAM-1 in the carvacrol treated-EAE group significantly increased as compared to the control group (P-value less than 0.05). Moreover, our study showed that the VCAM-1 gene expression in EAE rat models was significantly increased as compared to the control group (P-value less than 0.05). Also, our results indicated that, there is no significant difference in ICAM-1 gene expression between EAE and carvacrol-treated EAE rats (P-value greater than 0.05).

Conclusion: In this study, we observed different effects of carvacrol on the gene expression involved in the angiogenesis in EAE rats. Based on these results, our hypothesis is that carvacrol can play an important role in the VCAM-1 gene expression in EAE-rat models. In summary, the results of our study indicated that carvacrol possibly increased the expression of the ICAM-1 gene. Further studies in animal and human studies and also in-vitro and in vivo experiments are needed to examine the molecular mechanisms of carvacrol in modulating angiogenic processes and preventing MS.



Keywords: Experimental autoimmune encephalomyelitis (EAE); carvacrol; gene expression; angiogenesis; VCAM-1.



The Effects of Chromium and Vitamin D3 Co-Supplementation on Insulin Resistance and Tumor Necrosis Factor-Alpha in Type 2 Diabetes

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Background and Aim: Type 2 diabetes mellitus (T2DM) as one of the most common metabolic disorders cause devastating chronic complications, including micro- and macrovascular diseases. Improvement of the insulin resistance (HOMA-IR) as a predictor of T2DM and its complications might have beneficial effects on glucose control and also on complications such as cardiovascular diseases in patients with T2DM. Inflammatory cytokines such as tumor necrosis factor- α (TNF- α) and interleukin 6 (IL-6) induce HOMA-IR in T2DM by altering the function of β -cells. In recent years, due to serious adverse effects of HOMA-IR modifying drugs, many studies have investigated the effects of diet and exercise on alleviation of HOMA-IR. Types of micronutrients, including vitamin D3 (25 (OH) D) and chromium (Cr+3) play a key role in regulation of carbohydrate metabolism and insulin action. To assess the efficacy of these popular nutritional supplements, we performed a randomized controlled trial to investigate effects of CrPic and vitamin D3 co-supplementations for four months on the serum measures of FBS, HOMA-IR, TNF- α and lipid profile as well as HbA1c in whole blood on subjects with T2DM

Methods: 92 patients with T2DM were randomly allocated to four groups to intake oral: (I) placebo of vitamin D3 (n=23); (II) vitamin D3 supplement at a dose of 50000 IU/ week (n=23); (III) CrPic supplement at a dose of 500 μ g/day (n=23) and (IV) both vitamin D3 at a dose of 50000 IU/ week and CrPic at a dose of 500 μ g/day (n=23) for four months, respectively. Serum FBS, high density lipoprotein cholesterol (HDL-C), total cholesterol (TC), triglyceride (TG), calcium (Ca²⁺) , and phosphorus (P) levels were measured by an enzymatic method using a Liasys autoanalyzer. Low-density lipoprotein cholesterol (LDL-C) was calculated using the Friedewald's formula (Chen et al. 2010). Serum levels of 25(OH) D (D3) vitamin were assayed using the VIDAS automated enzyme linked fluorescent immunoassays technique with a Vidas immunoassay analyzer. Serum insulin and parathyroid hormone (PTH) were determined by a Tosoh immunoassay analyzer (Elecsys 1010 System, Tosoh, Tokyo, Japan). HbA1c on whole blood was determined by HbA1c point-of-care testing. Serum TNF- α was measured through the ELISA method according to the manufacturer's instructions. Insulin resistance. HOMA-IR was calculated by the formula

Results: HOMA-IR levels increased significantly in groups I and II after the intervention. However, this increase in group I was significantly higher than that in group II after the treatment. HOMA-IR levels were controlled in groups III and IV during the intervention. TNF- α decreased significantly in



groups II, III, and IV after the intervention. FBS, HbA1c, and lipid profile did not change significantly in total groups after the intervention.

Conclusion: It seems that chromium and vitamin D3 co-supplementation are probably effective in controlling HOMA-IR by decreasing TNF- α in T2DM.

Keywords: Type 2 diabetes mellitus; Vitamin D3; Chromium picolinate; Insulin resistance; Inflammatory response; Hemoglobin A1c



The Effects of *Elaeagnus angustifolia* L. On Lipid Profile and Cardiovascular Function in Menopausal Women: A Double-Blind, Randomized, Placebo-Controlled Study

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Background and Aim: The reduced production of ovarian hormones is known as the cause of an increase in the incidence of heart disease in post-menopausal women. Phytosterols are found in various herbal sources and have structural similarity with estrogen. Accordingly, they are considered as an alternative to hormone therapy. *Elaeagnus angustifolia* L., known as Senjed in Persian, contains valuable compounds, including two types of phytoestrogens and is used in Iranian traditional medicine with various medicinal properties. In the present study, the efficacy of *E. angustifolia* fruit on the cardiovascular function and lipid profile in postmenopausal women was investigated.

Methods: In this double-blind placebo-controlled clinical trial, 58 postmenopausal women were randomly assigned into two experimental groups of medicinal herb (15 g/day of the whole *E. angustifolia* fruit powder) and placebo (15 g/day of isomalt + corn starch). At the beginning and end of the trial, cardiovascular function (heart rate, blood pressure), and lipid profile (total cholesterol, TC; triglyceride, TG; LDL-C and HDL-C) were measured.

Results: Heart rate and serum levels of LDL-C and HDL-C significantly decreased after treatment with *E. angustifolia*

Conclusion: After 10- week treatment with *E. angustifolia*, the overall health of the participants, as well as cardiovascular function and lipid profile were improved.

Keywords: Menopause (MeSH Unique ID: D008593); *Elaeagnus angustifolia* L; Lipid Profile; Cardiovascular Function (MeSH Unique ID: D002319); Iran

The Effects of Green Synthesized Copper Nanoparticles on Liver Function and Hematological Parameters in Mice

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Background and Aim: The present investigation is based on the green synthesis of copper nanoparticles (CuNPs) from aqueous extract of *Capparis spinosa* L. fruit. Their effects on liver function and hematological parameters in mice were evaluated.

Methods: The green synthesis of CuNPs by means of *C. spinosa* extract was achieved. Ultraviolet-visible spectroscopy, fourier transform infrared (FTIR) spectroscopy, scanning electron microscopy (SEM), and energy dispersive X-ray spectroscopy were used to identify the synthesized nanoparticles. BALB/c mice were orally administrated CuNPs at doses of 1000, 2000, and 5000 µg/kg for 2 weeks. Later, the effects of CuNPs on liver function in the treated mice were evaluated by measuring the serum levels of enzymes such as alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, and bilirubin as well as hematological parameters including hemoglobin, hematocrit, white blood cell, red blood cell, and platelet counts.

Results: A maximum peak at wavelength 414 nm confirmed the biosynthesis of CuNPs. FTIR spectrum analysis revealed that the factor groups shaped a coating extract on the surface of the nanoparticles. SEM images demonstrated a particle size between 17 and 41 nm. Although some liver enzymes and hematological parameters increased with increasing dose of extract, there was no significant difference ($p>0.05$) between oral administrations of CuNPs at doses of 1000, 2000, and 5000 µg/kg and the control group.

Conclusion: The findings revealed that CuNPs biosynthesized from aqueous extract of *C. spinosa* fruit have no toxic effects on the liver functions and hematological parameters of mice. However, more studies are needed for evaluation of the hepatoprotective effects of CuNPs.

Keywords: Nanoparticles; copper; liver; hematology; *Capparis spinosa*; BALB/c mice

The Effects of Methizumol and Levothyroxine on DNMT3A Gene Expression in Rat Thyroid Tissue

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Background and Aim: Thyroid hormones (T3 and T4) are essential for proper cellular development (1-4). These hormones, bound to their receptors, regulate gene transcription in part by modulating posttranslational modifications of histones, microRNAs, and DNA methylation (5-8). DNA methylation, is established by a family of DNA methyltransferase (DNMT) including DNMT1, DNMT3A, and DNMT3B influencing gene transcription (9-10). In this study, we have investigated the influence of thyroid hormones (T3 and T4) on DNMT3A expression in rat thyroid tissue.

Methods: Thirty Sprague- Dawley female rats allotted into 3 dietary groups of 20 animals in hypothyroid, 20 animals in hypothyroid, and 10 animals in euthyroid (control). Each group received experimental diets containing methizumol and levothyroxine every day as follows: control group fed a normal diet and water; hypothyroid group received methizumol 0.1% in drinking water/day for two weeks and the hyperthyroid group received levothyroxine 96 mg /30 ml of drinking water for one month. The expressions of the DNMT3A gene were quantitated using real-time RT-PCR.

Results: The average expression level of the DNMT3A mRNA was found to be 7.95-fold higher in the thyroid of hyperthyroid rats as compared to euthyroid control rats (p-value less than 0.05). Moreover, our results indicated that the gene expression level of DNMT3A in the hyperthyroid group showed a significant increase as compared to the hypothyroid group (p-value less than 0.05).

Conclusion: Thyroid hormones could modulate the expressions of DNMT3A gene expression in the thyroid of hyperthyroid rats. To the best of our knowledge, there is no previous report of the effect of thyroid hormones on DNMT expression in animals.

Keywords: DNA methyltransferase (DNMT); gene expression; thyroid hormones

The Effects of *Saccharomyces Boulardii* on Liver Function Tests and Oxidant/Antioxidant Parameters in Rats with Obstructive Jaundice

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Background and Aim: The yeast *Saccharomyces boulardii* (*S. boulardii*) is a probiotic that it has been received more attention due to immune responses regulation, reducing oxidative stress and maintaining intestinal eubiosis. Several studies showed that some live bacteria and yeasts attenuate liver fibrosis, which is an inflammatory response results from chronic hepatic injury. Nevertheless, the safety of live probiotics is an important issue. Thus, the aim of this study was to investigate the effects of heat-killed *S. boulardii* on an experimental model of liver fibrosis in rat.

Methods: Bile duct ligation (BDL), was performed in male Wistar rats to induced liver fibrosis. The study included four groups (n=8), two groups were Bile duct ligated, one group received heat-killed *S. boulardii* and another one vehicle. Sham-operated and normal controls either received vehicle. Heat-killed *S. boulardii* were administered by gastric gavage once a day for 28 days. At the end of treatment, the body weight gain was analyzed. Serum activities of enzymes ALT, AST, ALP, LDH, level of bilirubin and FBS were measured. Antioxidant parameters including malondialdehyde (MDA), nitric oxide (NO), glutathione (GSH), total antioxidant capacity (TAC), and activity of Catalase (CAT) and Superoxide dismutase (SOD) enzymes were measured in liver tissue.

Results: Liver function test levels such as; ALT, AST, ALP, LDH and bilirubin were lower in the BDL+ *S. boulardii* group when compared to the BDL control group ($P \leq 0.05$). However, FBS and the body weight gain were not significantly changed ($P \geq 0.05$). There were low liver contents of MDA and NO and a high level of TAC, GSH, CAT and SOD in BDL+ *S. boulardii* group compared to the BDL control group ($P \leq 0.05$).

Conclusion: Results in this study showed that heat-killed *S. boulardii* had a beneficial effect on liver function and reducing oxidative stress in liver tissues. The findings suggest that heat-killed *S. boulardii* have beneficial effects against liver fibrosis progression.

Keywords: *Saccharomyces boulardii*; Bile duct ligation; Liver fibrosis; Antioxidant

The Evaluation of Hesperidin Effect on Serum Lipid Profile in The Bile Duct Ligation rats

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Background and Aim: Cholestasis is a condition in which the bile flow from liver is reduced or blocked. It can be caused by a disorder in the liver cells, intrahepatic bile ducts or extrahepatic bile ducts. Inadequate bile flow leads to the accumulation of bile contents, including bile acids, in the liver and finally in the blood. It also disrupts fat metabolism and result in hyperlipidemia. Bile duct ligation (BDL) is a recognized experimental model used to cause obstructive cholestatic injury in rodents. The aim of the present study is to investigate the effect of Hesperidin (classified as a bioflavonoid) on serum lipid profile in cholestasis.

Methods: Twenty and four male Wistar rats were categorized into four groups (n = 6, 250-300 g). sham group, the bile duct ligation (BDL), and BDL + hesperidin groups. Bile duct ligated rats have been treated with hesperidin at doses of 100, 200 mg/kg orally for 14 days. In serum samples, lipid profile parameter such as triglyceride (TG), total cholesterol (TC), low density lipoprotein (LDL) and high-density lipoprotein (HDL) were measured via automated chemistry analyzer.

Results: Serum levels of TG, TC, LDL were significantly high in the BDL group compared with sham group, while HDL level significantly decreased in the BDL group.

Conclusion: The present study suggests that Hesperidin may improve serum lipid profile in rats following BDL.

Keywords: liver; bile duct ligation; hesperidin; lipid profile; rat

The Evaluation of Relationship Between Lead and Cadmium Concentration in Semen and Serum of Infertile Men and The Control Group, in Bushehr City During the Year 2017

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Background and Aim: Infertility is a common disease with consequences of psycho-economic and medical problem. It is a disease of reproductive system, defined by failure to achieve a clinical pregnancy after 12 months of having regular unprotected sexual inter course. Some studies have shown that lead and cadmium could significantly affect the infertility in men. The aim of this study was to evaluate the relationship between lead and cadmium concentration in semen and serum with infertility in men in Bushehr city.

Methods: In a case –control study 43 men were participated and divided into two groups (24 in case group and 19 in the control group). A questionnaire was filled showing Demographic datas and infertility period. Blood (5 cc) and semen samples were collected from each patient. Lead and cadmium concentration were measured in all the sampels by using atomic absorption method. Statistical analysis was perforemed by SPSS number 18 - version.

Results: There was no significant difference between the two groups regarding the mean levels of lead and cadmium in serum and semen. However, the concentration of lead and cadmium in infertile men were higher than in normal men. The chance of infertility was not significantly affected by the levels of lead and cadmium in semen and serum as compared with normal. Moreover, there was a significant positive relationship between cadmium levels in semen with WBC. Also, a negative correlation was found between serum concentration of lead and WBC in semen infertile men.

Conclusion: There was no significant relationship between the two groups regarding the mean levels of lead and cadmium in serum and semen.

Keywords: Lead; cadmium; serum; semen; infertility; concentration

The Footprint of Nuclear Factor (NF)- κ B in The Pathogenesis of the SARS-CoV-2 Virus; a Valuable Target for Therapeutic Intervention

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Background and Aim: In inflammatory diseases and conditions, the nuclear factor (NF)- κ B signaling axis plays a critical role. Through enhancing the expression of genes encoding pro-inflammatory cytokines and polarization of macrophages into pro-inflammatory phenotype, NF- κ B has been identified to be the main executor of cytokine storm and its related tissue damage. In COVID-19 disease, NF- κ B have also a solid foot in the induction of severe acute respiratory syndrome-coronavirus-2 (SARS-CoV-2), which is the main reason why patients were subjected to intense care unit (ICU). As the SARS-CoV-2 infected the host's cells via the interaction between the Spike protein and ACE2R, the activated CE/Ang II/AT1R axis recruits STAT signaling pathway to increase the transcription activity of NF- κ B.

Results: The produced pro-inflammatory cytokines such as IL-6 could also make a vicious cycle and, in turn, more vigorously enhance the activity of NF- κ B. Since this nuclear transcription factor bifurcates with numerous signaling pathways, its activity due to COVID-19 disease could bring patients other complications. In this vein, the interaction of NF- κ B with Nrf2 has been proposed to could induce long-term neuroinflammation in the patients. Or, the connection between NF- κ B and the PI3K/Akt signaling pathway could lead to multiple organ failure. Additionally, it should be noted that the upregulation in the expression of NF- κ B with the advance of age or due to the several underlying diseases increases the susceptibility of individuals to COVID-19 infection. So far, several microRNAs, including miR-9, miR-98, miR-223, and miR-214 were identified that could prevent the activation of NF- κ B in inflammatory disease.

Conclusion: Given these, it has been proposed that targeting the miRNA network not only may change the therapeutic pattern of COVID-19 disease but also could prevent the activation of NF- κ B in high-risk patients and thereby reduce the mortality rate of the disease.

Keywords: Inflammation; NF- κ B; MicroRNAs; SARS-CoV-2 virus; Cytokine storm

The Gene Expression of Long Non-Coding RNAs (lncRNAs): MEG3 And H19 in Adipose Tissues from Obese Women and Its Association with Insulin Resistance and Obesity Indices

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Background and Aim: Data from animal models and in vitro studies point to the role of two lncRNAs; MEG3 and H19 the patho mechanism of obesity and related disorders. Here, we aimed to evaluate the expression of MEG3 and H19 in visceral adipose tissues (VAT) and subcutaneous adipose tissues (SAT) of obese women (n=18), as compared to normal-weight women (n=17). Moreover, we sought to identify the association of expression of MEG3 and H19 in SAT and VAT with obesity parameters, insulin resistance, and the mRNA expression of possible target genes involved in adipogenesis and lipogenesis including peroxisome proliferator-activated receptor gamma (PPAR γ), fatty acid synthase (FAS), and acetyl-CoA carboxylase (ACC)

Methods: Real-time PCR was performed to investigate the mRNA expression of the above-mentioned genes in VAT and SAT from all participants

Results: The results showed lower mRNA levels of H19 in SAT of obese women, compared to normal-weight women. While MEG3 expression was significantly higher in the SAT of the obese group rather than controls. Correlation analysis indicated that the transcript level of H19 had an inverse correlation with obesity indices and HOMA-IR values. However, MEG3 expression displayed a positive correlation with all the indicated parameters in all participants. Interestingly, a positive correlation was found between transcript level of MEG3 in SAT with FAS and PPAR γ . However, there was an inverse correlation between SAT expression of H19 and FAS.

Conclusion: It appears that lncRNAs; MEG3 and H19 are involved in obesity-related conditions. However, more clinical studies are still required to clarify the relationships between lncRNAs with obesity and related abnormalities.

Keywords: Long non-coding RNAs (lncRNAs); Obesity; Lipogenesis; Adipogenesis

The Human IGFBP-3 Serum Level in Adult Respiratory Distress Syndrome Status in COVID-19 Patients

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Background and Aim: Coronavirus (COVID-19) is caused by SARS-CoV2 and is considered to be the cause of a dangerous disease that has raised widespread welfare concerns around the world. Coronavirus is one of the major pathogens that target the human respiratory tract. Previous episodes of Coronavirus have included Severe Respiratory Disorder (SARS) -CoV and East-Central Respiratory Disorder (MERS)-CoV, which have long been known to be exceptionally disabling for open well-being.

Results: The first sights of SARS-CoV-2 were published and revealed that the existence of SARS-CoV-2 in CSF of COVID-19 cases display that SARS-CoV-2 is expensing to the CNS. Novel effective researches of COVID-19 cases with non-acute indication identical to wholesome control illustrate a notable decline in GH and IGFBP-3 pending hospitalization. Further, the TSH, FT3, P1NP, N-MID OC, 25 (OH) D3, GH, and IGFBP-3 level was diminishing in COVID-19 cases set against the control participants. GH and IGFBP-3 were considerably diminished in the serum of COVID-19 patients. On the other hand, Previous research reported that the diagnosis of SARS-CoV-2 in the CSF of COVID-19 patients demonstrates that the Central Nervous System might be forayed by SARS-COV-2. Recent studies have presented that serum levels of IGF-1 and IGFBP-3 are arising in high-risk patients who have not experienced early adult respiratory distress syndrome. Survival relative to survivors indicates epithelial damage. However, they declined in the late ARDS.

Conclusion: ARDS is strongly correlated with IGF-1 and IGFBP-3 levels in acutely ill patients, making them a potential biomarker.

Keywords: IGFBP-3; ARDS; SARS-COV-2

The Impact of A β on ABCA1 in Microglia, Astrocytes, and Neurons Isolated from C57BL/6 Mice

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Background and Aim: Impaired of cholesterol metabolism has been reported in Alzheimer's disease. Since, ABCA1 is one of the main players in the brain cholesterol homeostasis, here we investigated and combined the in vivo data associated with the effect of amyloid beta (A β) on the protein expression of ABCA1 in microglia, astrocytes, and neurons with the in-silico experiments of A β /ABCA1 interaction.

Methods: microglia, astrocytes, and neurons were isolated, cultured, and exposed to beta amyloid. ATP-binding cassette transporter A1 (ABCA1) in cell lysates were determined by Western blotting technique and cholesterol efflux was measured in the conditioned media.

Results: The results showed that the protein expression of ABCA1 is significantly increased by A β treatment in microglia, astrocytes, and neurons, however it is not reflected in its function to increase cholesterol efflux.

Conclusion: our findings showed that the effect of A β on increasing of ABCA1 protein levels in different cells of brain is a general and cell-type independent effect(s). We believe that the spatial conformational changes of ABCA1 will lead to the subsequent events that include its stability and decreased its ability to release the cellular cholesterol. A possible explanation for ABCA1 stability is that altered spatial conformation causes ABCA1 to become less accessible (ABCA1 becomes resistant to calpain) to the proteases like calpain family.

Keywords: Amyloid beta-Peptides; cholesterol; ATP Binding Cassette Transporter 1

The Impact of Continuous Positive Airway Pressure Therapy on Total Antioxidant Capacity in Obstructive Sleep Apnea: A Systematic Review and Meta-Analysis

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Background and Aim: Obstructive sleep apnea (OSA) is a highly common respiratory disorder characterized by chronic intermittent hypoxia and fragmented sleep due to obstruction of the upper airways. Hypoxia / re-oxygenation is associated with an impaired mitochondrial respiratory chain leading to the production of Reactive Oxygen Species (ROS). Total Antioxidant Capacity (TAC) is an integrated parameter in the study of oxidative stress in various pathological conditions. Continuous positive airway pressure (CPAP) has been confirmed as the first line of treatment and the most effective therapy for OSA patients. CPAP, as a positive airway pressure, prevents intermittent hypoxia by injecting airflow. Thus, this systematic review and meta-analysis were performed to evaluate the effect of CPAP treatment on TAC levels in OSA patients.

Methods: We searched systematically online databases including PubMed, Web of Science, Scopus, EMBASE, and Cochran library published to January 2021, to identify the relevant investigating that evaluated TAC in patients with OSA before and after CPAP therapy. The following inclusion criteria were used to select the related articles: using polysomnography to the diagnosis of OSA, measuring TAC levels before and after CPAP therapy, using CPAP at least for 24 hours, and the study provides adequate data for meta-analysis. Quality assessment of studies was performed using Cochrane criteria and statistical analyses were performed using Stata version 14.2 (Stata Corp, College Station, TX, USA).

Results: Twelve articles were included in this study, In this meta-analysis demonstrated that CPAP therapy significantly increased TAC levels in OSA patients [SMD (+1.51) (95% CI, +2.06 to +0.97) $p < 0.05$].

Conclusion: As a result, the current systematic review and meta-analysis have shown the beneficial effects of CPAP therapy on TAC levels in OSA patients.

Keywords: Obstructive sleep apnea; Continuous positive airway pressure; ROS; TAC; meta-analysis

The Increased Incidence Rate of Guillain-Barre Syndrome in COVID-19 Pandemic

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Background and Aim: SARS-CoV-2 is a single-stranded RNA enveloped virus that has caused the coronavirus disease 2019 (COVID-19) pandemic. Guillain-Barre Syndrome (GBS) is an inflammatory polyradiculoneuropathy associated with several viral infections. *Campylobacter jejuni* is the most common pathogen associated with GBS along with viral pathogens including cytomegalovirus (CMV), Epstein-Barr virus (EBV), and Zika-virus (ZIKV). GBS has also been associated with influenza infections and vaccinations.

Results: GBS is not a usual form of COVID presentation, but the relative frequency and standardized incidence in COVID patients were higher than in non-COVID patients. It is suggested that COVID-19 may cause peripheral nervous system complications which meets the diagnostic criteria of an acute sensory and motor poly radiculoneuritis. On average it took 11 days from COVID-19 symptoms to GBS symptoms. COVID-19 may dysregulate immune system due to systemic hyper inflammation in COVID-19 patients with a macrophage activation syndrome (secondary hemophagocytic lymphohistiocytosis) or alterations in lymphocytes. Moreover, SARS-CoV-2 is able to cause an immune reaction with an increased level of interleukin-6 (IL-6) which stimulates the inflammatory cascade and damages tissues. COVID-19 may indirectly cause GBS through the inflammatory response of the immune system or directly through neuroinvasive capacity (ACE2 receptors of neuronal tissues). Therefore, inflammatory factors may have an important role in the organ dysfunctions of COVID-19 patients responsible for most of the neurological manifestations.

Conclusion: Taken together, GBS associated COVID-19 appears to be a rare condition with similar patterns to GBS before the pandemic. Finally, it is wise not to limit the research between COVID-19 and nervous system to the current pandemic so we can be ready for the potential new variants and viruses.

Keywords: COVID-19; Guillen-barre syndrome; SARS-CoV-2

The *Lactobacillus Reuteri* Attenuates Oxidative Stress and Liver Damage in a Rat Model of Obstructive Cholestasis

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Background and Aim: Liver fibrosis is a complex fibrogenic and inflammatory process that results from chronic liver injury and represents an early step in the progression of liver cirrhosis. One species of *Lactobacillus*, *Lactobacillus reuteri* (*L. reuteri*), due to its strong modulatory effects on host microbiota and immune responses is a good strategy against inflammatory diseases. This study was conducted to determine the hepatoprotective effect of heat-killed *L. reuteri* in a liver fibrosis model.

Methods: Liver fibrosis was induced by common bile duct ligation (BDL). The study included four groups (n=8), BDL group received heat-killed *L. reuteri*, BDL control, sham-operated controls and normal controls that these groups received vehicle. *L. reuteri* or vehicle were administered daily by gastric gavage. Heat-killed *L. reuteri* prescribed 7 days before BDL and then for 21 consecutive days after BDL. At the end of the experiment, rats were sacrificed. We measured serum levels of FBS, ALT, AST, ALP, LDH and bilirubin. Also, Liver tissues were assessed for oxidant and antioxidant parameters such as MDA, nitric oxide, total antioxidant, GSH, Catalase and SOD activity.

Results: Serum levels of ALT, AST, ALP, LDH and bilirubin in BDL group received heat-killed *L. reuteri* were lower than BDL control ($P \leq 0.05$). Oxidative stress markers include; MDA and nitric oxide were lower and total antioxidant capacity, glutathione, catalase and superoxide dismutase activity in liver tissue were higher in BDL group received heat-killed *L. reuteri* in compare to BDL control. ($P \leq 0.05$). Nevertheless, the difference in serum level of FBS and body weight gain were no significant ($P \geq 0.05$).

Conclusion: Results showed that heat-killed *L. reuteri* significant reduced oxidative stress and it improved liver function in BDL rats. The findings suggest that heat-killed *L. reuteri* beneficial effect against liver injury via oxidant/antioxidant pathways.

Keywords: *Lactobacillus reuteri*; Bile duct ligation; Liver Function Test; Oxidative stress

The Protection Role of Gallic Acid Against Cisplatin-Induced Nephrotoxicity

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Background and Aim: Cisplatin is a chemotherapeutic drug used in the treatment of solid tumors. Aggregation of the drug in renal tissue causes nephrotoxicity. Researches showed nephrotoxicity as limiting side effect. One of the Cisplatin side effects is generation of reactive oxygen species, which leads to oxidative stress and finally renal damage. The purpose of this study was evaluation of Gallic acid effect on Cisplatin-induced toxicity including biochemical and histopathological variations in adult male Wistar rats.

Methods: Rats were divided into 4 groups including GpI (control group) (received distilled water for 10 days); GpII:(Gallic acid 50 mg/kg, for 10 days); GpIII (Cisplatin 6 mg/kg bw, I.P. on 5th day); GpIV:(Gallic acid 50 mg/kg, for 10 days and Cisplatin 6 mg/kg, I.P., on 5th day). Then all rats were anaesthetized and plasma collected to estimate MDA, SOD, Catalase, GPX, and GSH and kidneys removed for histopathological study.

Results: The Cisplatin - induced nephrotoxicity was evident from the elevated levels MDA P less than 0.0001 , decreased levels of SOD , CAT , GPX , GSH , P less than 0.0001 . GA modulate nephrotoxicity ("Cis-gal vs. Cis" MDA: P less than 0.0001, SOD: P less than 0.0001, CAT: P less than 0.0001, GSH: P less than 0.0001, GPx: P less than 0.001). GA Comparing the cis. group with cnt.group: cisplatin in cases of vacuolization , P equal to 0.006, thickness of proximal, P equal to 0.002, cell necrosis, P equal to 0.006, eosinophilic casts , P equal to 0.006 , caused damage and GA showed protective effects .

Conclusion: The Cisplatin-induced nephrotoxicity was evident from the elevated levels MDA and decreased of SOD, CAT, GPX, GSH and tissue damage of kidney. Outcomes of the present investigation suggest that Gallic acid provides protection against cisplatin induced nephrotoxicity, but for application in medicine, further studies are needed.

Keywords: cisplatin; nephrotoxicity; Gallic acid; antioxidant parameters; histopathological

The Relationship Between Serum Levels of Interleukin-2 And IL-8 with Circulating MicroRNA-10b in Patients with COVID-19

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Background and Aim: The role of cytokine storm in the immunopathogenesis of coronavirus disease 2019 (COVID-19) has been implicated. To determine the association of microRNA (miRNA)-10b and serum levels of IL-2 and IL-8 in patients with COVID-19.

Methods: Blood samples were obtained from 33 COVID-19 patients and 29 healthy subjects. After RNA extraction and cDNA synthesis, the transcript level of miR-10b was determined by Realtime PCR. In addition, the serum levels of IL-2 and IL-8 were measured in subjects using ELISA.

Results: The patient group comprised of 33 patients with COVID-19 (62.4 ± 3.7 years old), containing 13 (39%) males and 20 (61%) females. In the control group, 29 subjects (56.6 ± 1.6 years old) containing 9 (31%) males and 20 (69%) females were included. The expression of miR-10b was significantly downregulated in the peripheral blood of COVID-19 patients in comparison to the healthy controls (fold change= 0.12, $P < 0.0001$). The levels of IL-2 ($P < 0.001$) and IL-8 ($P < 0.001$) were significantly increased in the serum samples of COVID-19 patients compared to the healthy subjects. The expression level of miR-10b was correlated significantly with the serum levels of IL-2 and IL-8 as well as with the age of patients, ESR and CRP level.

Conclusion: miR-10b is downregulated in the COVID-19 patients and might result in increased levels of IL-2 and IL-8, hence contributing to cytokine storm.

Keywords: Coronavirus disease 2019; MicroRNA; Cytokine storm; Interleukin-2; Interleukin-8

The Relationship Between Serum Vitamin E Levels and Colorectal Cancer Incidence

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Background and Aim: Colorectal cancer (CRC) is the one of must leading cause of cancer death, as it the second and third common cancer in women and men, respectively. Recent examinations Vitamin E is a type of fat-soluble vitamin and has antioxidant properties that can be anti-cancer factor.

Methods: In the present study, 50 CRC patients and 50 healthy individuals were entered according to inclusion and exclusion criteria. Peripheral blood was taken from the subjects and the serum was separated from the samples on the same day and stored in freezer - 20 CO until the final test. Vitamin E levels were measured by spectrophotometry method. The level of serum vitamin E was measured. Data was analyzed by t-test and the Mann Whitney test. Confidence interval (CI) 95% was considered.

Results: The outcomes indicated that vitamin E values were significantly showed lower concentrations in patients with CRC compared with healthy controls (P value: ≤ 0.5)

Conclusion: Insufficient vitamin E of serum can be a reason that led to an increase in CRC incidence and progression. CRC is associated with a lower concentration of serum vitamin E. Therefore, monitoring of vitamin E levels can be a factor in assessing the risk of colorectal cancer that may play a role in CRC screening and maintaining vitamin E levels at the normal level can be a preventive factor in colorectal cancer.

Keywords: Colorectal cancer; Vitamin E; Screening; Serum

The Relationship Between Vitamin D Deficiency and Increased Oxidative Stress in Patients with Colon Cancer

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Background and Aim: Colorectal cancer (CRC) is one of the most prevalent cancers worldwide. Oxidative stress is one of the involved factors in CRC onset and progression. Recent examinations have revealed antioxidant characteristics of vitamin D. Given the vital role of this vitamin in balancing free radicals and antioxidant capacity, in this study we intended to review the association between vitamin D deficiency and oxidative stress in CRC patients.

Methods: In the present case-control study, 30 CRC patients and 32 healthy individuals were entered, based on the defined inclusion and exclusion criteria. Peripheral blood was taken from the subjects. Thiobarbituric acid reactive substance (TBARS) values, total antioxidant capacity, and serum vitamin D were measured. Data were interpreted using SPSS 18 software; t-test and the Mann Whitney test were applied.

Results: The outcomes explained that TBARS values were significantly greater in patients group (P value: less than 0.005), but no meaningful difference was monitored in the total antioxidant capacity. 21 (70%) patients and 14 (44%) control subjects had inadequate vitamin D. There was a significant association between serum vitamin D in both groups (P value less than 0.005). A notable negative relationship was found between vitamin D values and oxidative stress indicator ($p=0.05$, $r=-0.249$).

Conclusion: Insufficient vitamin D can lead to an increase in oxidative stress, which is directly associated with CRC. Serum vitamin D levels were also inadequate in high percentage of cancer patients. Given the predominance of vitamin D insufficiency in the population, more extensive studies are required to prove the impact of deficiency on disease pathogenesis.

Keywords: Colorectal cancer; Vitamin D; Oxidative stress

The Role of Circular RNAs in Diabetic Nephropathy: Mechanism of Action and Therapeutic Targets

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Background and Aim: Diabetes mellitus (DM) is a complex metabolic disease in which glucose homeostasis is impaired. Today, diabetes has become one of the most critical causes of death worldwide and has major negative effects on health systems. Chronic high blood sugar over a long period of time leads to damage to various organs such as the eyes, kidneys and nervous tissues. Damage to these organs can be caused by microvascular defects and pathological events such as retinopathy, nephropathy, and neuropathy. Diabetic nephropathy (DN) is a severe renal complication of DM. Non-coding RNAs (ncRNAs), which play an important role in regulating physiological and pathological processes, are responsible nearly 60% of the human genome's transcripts as non-protein coding RNA transcripts. CircRNAs are a new class of ncRNAs that are protected against RNA exonucleases due to their covalently closed structures. CircRNAs have different effects on cell signal processes by targeting different molecular mechanisms. Based on studies, it has been reported that the pathogenesis of DN is positively affected by circRNA. CircRNAs can target miRNAs as a key regulatory axis, from their molecular targets, and can be considered as miRNA sponges. Thus, circRNAs may prevent DN progression by suppressing specific miRNAs.

Results: CircRNAs have today attracted much attention due to their various functions in the physiological and pathological conditions of the human body. In various pathological conditions and diseases, including diabetes and its complications, circRNAs act through a network of modulatory mechanisms. One of the well-studied regulatory mechanisms associated with circRNAs is sponge miRNAs, in which the circular structures of circRNAs target specific miRNAs, thereby increasing or decreasing genes, proteins, and specific transcription factors. DN, which is one of the most serious complications of diabetes, is considered to be the main cause of ESRD and even death in these patients. For this reason, it is important to find the basic and new mechanisms involved in the pathogenesis of this disease. Due to the influence of circRNAs in regulating the pathways involved in DN, in accordance with the development of desirable strategies to mediate the various circRNAs that play a role in this disease, in the near future, therapeutic measures to target these RNA molecules to improve DN and other complications of diabetes can be effective. Fortunately, a new research insight into DN in the coming years is likely to be the field of circRNA biology.



Conclusion: In this study, it can be concluded that circRNAs, due to their undeniable modulating functions in DN progression, can be considered as a promising target in the treatment of this disease. However, there are several limitations that may decelerate the successful development of circRNAs in the clinic. These include the ambiguity of biogenic mechanisms and their elimination, the complexity of their secondary structures and doubts about whether they can be considered as diagnostic biomarkers. These will be identified through extensive research and consideration of the role of circRNAs in DNA.

Keywords: Diabetic nephropathy; Circular RNAs; MicroRNAs; Cellular mechanism



The Similarity Between the Pathogenesis of COVID-19 Disease And Autoimmune Disease; A Lesson We Could Learn From Previous Findings

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Background and Aim: A recent member of the Coronaviridae family, the SARS-CoV-2 virus has made a global crisis. By targeting ACE2-expressing cells, this virus could infect the human respiratory system, leukocytes, and platelets. Although severe acute respiratory syndrome (SARS) is one of the most devastating side effects of this virus, excessive release of pro-inflammatory cytokines, known as cytokine storm, is the main cause that leads to multiple organ failure and subsequently disease mortality. Interestingly, the incidence of the cytokine storm is a parameter that makes the pathogenesis of COVID-19 disease close to the autoimmune disease. It seems that in both diseases, T helper (Th)-17, JAK/STAT signaling pathway, and nuclear factor (NF)- κ B transcription factor might be involved in reinforcing the production of pro-inflammatory cytokines such as TNF- α , IL-1, IL-6, and IL-17.

Results: The similarity between the pathogenesis of COVID-19 infection and several autoimmune diseases including systemic lupus erythematosus (SLE) as well as rheumatoid arthritis (RA) brings up the idea that perhaps the same therapeutic approach could be engaged for both diseases. Anifrolumab (a monoclonal antibody against INF-I), Hydroxychloroquine (HCQ), and Quinapril (ACE inhibitor) are the best examples of the autoimmune-related drugs that showed to be successful in the treatment and the management of COVID-19 disease. For example, HCQ has received FDA approval for the treatment of COVID-19 as an emergency use authorization (EUA) in 2020. This drug has been shown to suppress the SARS-CoV-2 replication, prevent lung involvement, and ameliorate respiratory symptoms and pulmonary inflammation in patients.

Conclusion: These findings suggest that although there is little time to understand the biology and the behavior of the SARS-COV-2 virus, using previous knowledge of the autoimmune diseases might help design drugs and treatment strategies against COVID-19 disease.

Keywords: Autoimmune disease; SARS-CoV-2 virus; Viral pathogenicity; Hydroxychloroquine; Pro-inflammatory cytokines.

The Study of The Effect of Hydroxytyrosol (HT) on Expression of Matrix Metalloproteinase-2, 9 (MMP-2, 9) in Hepatocellular Carcinoma Cell Line (HepG2)

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Background and Aim: Hepatocellular carcinoma (HCC) is one of the most common types of cancer in the world, accounts for 90% of all incidents of primary liver malignancy. Most cancer-related deaths are affected by the invasion, migration, and metastasis of cancer cells to other organs. In the last two decades, the anticancer and anti-metastatic properties of natural polyphenols have become a hot topic in many laboratories. Hydroxytyrosol is a potent antioxidant that has numerous activities in different malignancies, such as abilities to regulate molecular targets, like matrix metalloproteinase-2, 9 (MMP-2, 9), and also signaling pathways, which were associated with cell survival, invasion, migration, and finally metastasis of cancer cells. In this study, we aimed to find out the effect of hydroxytyrosol on the expression of MMP-2, 9 in hepatocellular carcinoma cells.

Methods: In the current investigation, the human Hepatocellular carcinoma cell line HepG2 was treated with different concentrations of hydroxytyrosol for 24 hours. The expression of MMP-2 and MMP-9, were determined by real-time PCR.

Results: Examination of gene expression showed that hydroxytyrosol significantly reduced the expression of MMP-2 in all groups of concentrations compared to the control. Also, the results indicated that hydroxytyrosol distinctly reduced the expression of MMP-9 in all treatment groups compared to control.

Conclusion: Our findings suggested that hydroxytyrosol probably plays a critical role in hepatocellular carcinoma metastasis by reducing the expression of MMP-2 and MMP-9, so it can be a helpful agent for preventing the progression of cancer cells.

Keywords: Hepatocellular carcinoma; Hydroxytyrosol; Metastasis; Matrix metalloproteinase

The Triangle Between Metabolic Disorders, Redox System, And the Severity of the COVID-19 Disease; The Probable Role of G6PD And GSH in The Pathogenicity of The SARS-COV-2 Virus

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Background and Aim: From the first description of the SARS-COV-2 virus, there was no doubt that this virus might recruit the redox system of the host to induce tissue damage, especially in the lungs. In many cases, it has been proposed that the disturbance in the redox system is the main reason for acute respiratory distress syndrome (ARDS) development in the patients. On the other hand, obesity and metabolic diseases such as diabetes were proposed to increase the risk of the development of the severe type of the disease in the individuals. It seems that there is a correlation between diabetes/obesity, the balance of the redox system, and the tissue levels of glutathione (GSH), a component of the antioxidant system that apart from scavenging free radicals, could regulate the metabolism of 25-hydroxyvitamin D (25(OH)VD) (vitamin D).

Results: But how does obesity/diabetes change the game in favor of COVID-19 development? The answer could be found in the suppression of glucose-6-phosphate dehydrogenase (G6PD), an enzyme that regulates GSH levels. GPD deficiency has been reported to increase the susceptibility of individuals to the SARS-COV-2 virus. In other studies, GSH depletion has also been indicated that could enhance the mortality rate of COVID-19 disease. Given these and putting all pieces together, it is reasonable to assume the presence of a triangle between obesity/diabetes, G6PD-mediated downregulation of GSH, and the development of severe cases of COVID-19.

Conclusion: Taken together, through impairing the balance of the redox system and shutting down the antioxidant arm of the redox system, it seems that metabolic disorders could pave the way for the SARS-COV-2 virus to increase the production of ROS, induce excess oxidative damage, activate inflammatory responses, and propagate the cytokine storm.

Keywords: SARS-COV-2 virus; Obesity; Redox system; glutathione; G6PD; Risk factor.

Toxic Heavy Metal Concentrations in Multiple Sclerosis Patients: A Systematic Review and Meta-Analysis

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Background and Aim: The present meta-analysis was performed to assess the association between multiple sclerosis (MS) patients and control subjects in terms of their circulating levels of arsenic (As), lead (Pb), mercury (Hg), and cadmium (Cd).

Methods: We searched Medline/PubMed, Scopus, Web of Science, and Embase up until June 2020 to identify all studies that examined the concentrations of heavy metals in MS patients. Statistical tests used to assess inter-study heterogeneity were Cochrane's Q test and the I² statistic. Given the observed significant heterogeneity, the random-effects model was employed to pool the weighted mean differences (WMDs) and the corresponding 95 % confidence intervals (CIs).

Results: Out of a total of 1181 articles, 16 studies with 1650 participants (772 patients with MS and 878 controls) were included in this review meta-analysis. Pooled results using random-effects model showed that the levels of Pb (WMD= 0.73 µg/L, 95 % CI= 0.33 to 1.12, P less than 0.001), As (WMD= 2.48 µg/L, 95 % CI= 1.44 to 3.53, P less than 0.001; I²= 98.9 %, P less than 0.001), and Cd (WMD= 0.17 µg/L, 95 % CI= 0.09 to 0.26, P less than 0.001) were significantly higher in MS patients than those of the controls. However, there were no significant differences in the levels of Hg (WMD= -0.14 µg/L, 95 % CI= -0.77 to 0.49, P = 0.658) among both groups. Sensitivity analysis indicated that after excluding one-by-one studies, the overall pooled WMD of Pb was changed.

Conclusion: This meta-analysis showed that patients with MS had significantly higher levels of circulatory As and Cd compared to the controls. Yet, there was no statistically significant difference between circulating levels of Hg and Pb among MS patients and controls.

Keywords: Heavy metal; multiple sclerosis; meta-analysis; arsenic; cadmium.

Vaccine Therapy as A Better Therapeutic Alternative in Glioblastoma

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Background and Aim: One of the most common primary malignancies in the brain is Glioblastoma (GBM), that has various treatment options such as surgery, radiotherapy, and temozolomide chemotherapy but the prognosis is still poor and the patients are faced with bad quality of life throughout the disease with median survival of 16 to 19 months. Therefore, Novel therapeutic approaches are required to improve outcome for patients with GBM

Results: The therapeutic potential of vaccine immunotherapy for malignant glioma should not be underestimated. There are investigations regarding therapies which target tumor growth factor receptors and downstream pathways, angiogenesis, modulation of cancer stemlike cells, cell cycle regulation, oncolytic viruses, novel radiotherapy methods, and immunotherapy, including vaccines and immune checkpoints modulation like programmed cell death 1 and cytotoxic T-lymphocyte antigen 4. In addition to novel agents, there are developments in methods to get past the blood-brain barrier to facilitate central nervous system drug exposure. However, first generation tyrosine kinase inhibitors of EGFR and PDGFR, rapamycin mammalian target, Akt, and PI3K inhibitors have failed in clinical trials. The monoclonal antibodies ipilimumab and nivolumab inhibit CTLA-4 and PD-1, based on their efficacy observed in melanoma are now being tested in recurrent glioblastoma. Vaccines for glioblastoma are being investigated as another way to increase the activation of the patient's immune system. Just as with vaccines against infections, tumor vaccines are used to prime the immune system against a specific antigen. Two types of vaccines are being explored: peptide based and cell based

Conclusion: A data eruption in the genetics, molecular biology, and immunology of glioblastoma has happened. Novel therapies may need cautious selection of patients depending on their mutations and protein expression of the tumor.

Keywords: Vaccine Therapy; Glioblastoma; Tumor; Immunotherapy

Vitamin D3 Is Well Correlated with Anti-Helicobacter Pylori Immunoglobulins and Could Be a Well Biomarker for Immunity Competence Against the Disease

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Background and Aim: *Helicobacter pylori* (HPY) provokes gastrointestinal disorders and gastric cancer. We supposed that the HPY disrupts the 25-OH vitamin-D3 (Vit.D3) absorption. We evaluated the association between Vit.D3 and anti-HPY immunoglobulins, the Vit.D3 potency as a predictive biomarker for HPY infection.

Methods: Methods: 603 patients' raw data were gathered from a private clinical laboratory. Anti-HPY immunoglobulins including serum IgG, IgA and IgM, in addition to HPY-stool antigen (STHP) were assessed by the immunoassay methods. Vit.D3 was determined by the high-pressure liquid chromatography (HPLC). Correlations, ordinal comparisons, cut-off points (COP) and odd's ratio (OR) were estimated.

Results: Age mean $\pm$ SD= 39.83 $\pm$ 18.426 for female and 38.82 $\pm$ 16.937 for male participants ($p=0.521$). Significant correlations existed after age and gender-adjustment between Vit.D3 serum levels and the HPY IgG ($R=0.298$) and IgA ($R=0.271$) but not for IgM ($R=-0.103$). Approximately, 48% of male and 36% of female had insufficient/deficient Vit.D3 serum levels (male/female OR: 1.65; 1.16-2.33; $p=0.0051$). After age- and gender-adjustment, the best cut-off point of Vit.D3 to predict a HPY IgG positive patient was Vit.D3 >32.80 ng/mL with 66.23% diagnostic accuracy (DAAC), 30.43% specificity (SPC), 90.41% sensitivity (SEN). For the HPY IgA the values were Vit.D3 >37.83 ng/mL, DAAC= 60.45%, SPC= 58.82%, SEN= 64.20%. For HPY IgM the values were Vit.D3 >37.32 ng/mL, DAAC= 58.97%, SPC= 57.33%, SEN= 100%.

Conclusion: Vit.D3 had good association with anti-HPY immunoglobulins and may be a good biomarker for immunity competence against HPY infection if the patient's age and gender are considered when interpreting the laboratory results.

Keywords: Biomarker; Cholecalciferol; Chromatography; *Helicobacter pylori*; Immunoglobulins



Medical Genetics



LZTFL1, A Novel Gene Responsible for Doubling the Risk of Death from Covid-19

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Background and Aim: The human species has lost many lives since the coronavirus disease 2019 (COVID-19) pandemic beginning from Wuhan, China. Different ethnic groups have shown a divergent relationship between the risk of serious disease and death. Social-economic inequalities have been suggested as an explanation which obviously is not thorough. LZTFL1 is an understudied gene identified by genome-wide association studies as the possible casual gene which likely doubles the risk of COVID-19 respiratory failure associated with 3p21.3. Through a combined multiomics and machine learning approach, rs17713054G>A SNP, a gain-of-function risk allele was recognized as a potential causative variant. In this study, out of 122 primary articles searched in PubMed and Google Scholar databases from 2020 to 2021, 20 articles with LZTFL1, SARS-CoV-2, COVID-19 and SNP keywords were selected and studied.

Results: Through gene expression analysis and chromosome conformation capture it was indicated that rs17713054-affected enhancer up-regulates leucine zipper transcription factor like 1 (LZTFL1). LZTFL1 encodes cytosolic leucine zipper protein which is associated with the epithelial marker E-cadherin and has a role in releasing of several signaling molecules. LZTFL1 is highly expressed in pulmonary epithelial cells specifically ciliated epithelial cells that is one of the major cellular targets of SARS-CoV-2 infection. Regarding malignancy, LZTFL1 inhibits epithelial–mesenchymal transition (EMT) which has a role in healing wounds and immune responses. Increased levels of LZTFL1 could hinder the beneficial effects of an acute EMT response, suppressing reduction of ACE2 and TMPRSS2 levels and/or slowing EMT-driven tissue repair.

Conclusion: The potential role of LZTFL1 and EMT in pulmonary pathogenesis needs further studies. It can be concluded that pulmonary epithelial cells that go through EMT are probably accountable for the 3p21.31-associated risk and not the immune cells. LZTFL1 could be a potential therapeutic target as the 3p21.31 effect is result of a gain-of-function.

Keywords: LZTFL1; SARS-CoV-2; COVID-19; SNP

A New Method for Screening 8 Types of Cancer

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Background and Aim: Floating tumor DNA mutations in the bloodstream can be highly specific markers of cancer. The use of a combination of selected biomarkers for the early diagnosis of cancer has the potential to change the way cancer is screened, and based on the same reasons, a combination of drugs can be used to treat cancer. The CancerSEEK test is a unique, non-invasive test that can detect the levels of 8 types of cancer proteins and the presence of mutations in ovarian, liver, stomach, pancreas, esophagus, colorectal, lung and breast cancers in DNA floating in the blood.

Methods: First, a liquid biopsy was performed on people who had recently been diagnosed with one of eight types of cancer. People who had cancer spread to other parts of the body were excluded from the study to focus on the early stages of the disease. First, several hundred genes and 40 protein markers were selected and then their number was reduced to 16 genes and 8 protein markers. The test was performed on 812 healthy controls and showed only 7 false positive results and was more than 99% specific for cancer. This high specificity is critical because it exposes patients to false positive and unnecessary follow-up tests to confirm the presence of cancer.

Results: The effectiveness of CancerSEEK method was very different depending on the type of cancer. The test diagnosed 98% of ovarian cancers, compared with 32% of breast cancers. The test was also able to identify the organ from which the disease originated in 63% of people. However, the test results were higher in the higher stages of cancer than in the early stages, and in stage III, the disease showed 63% versus 43% of stage I tumors.

Conclusion: This test detected cancer in about 70% of cases in more than 1000 people who were diagnosed with cancer, and the next step was to change the focus of cancer from higher to early stages. Which is vital for reducing long-term cancer mortality.

Keywords: new method; cancer; screening.

An Expression Signature of lncRNA SPRY4-IT1 in the Breast of Apparently Clinically Healthy Women is Associated with Aging and Menopausal status

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Background and Aim: Revealing the mechanisms of action of breast cancer risk factors seems to be a key priority in today's medical research, especially their early molecular footprints before the clinical onset of tumors. Previous studies have shown that breast cancer-related factors are related to the function and expression of long noncoding RNAs (lncRNAs). On the other hand, several pieces of evidence support the occurrence of several early expression lesions in the breasts of apparently clinically healthy women that link to some breast cancer risk factors. In the current study, we assessed the expression levels of the two-breast cancer-related lncRNAs, including SPRY4-IT1 and H19, in relation to various breast cancer risk factors in breasts of apparently clinically healthy women.

Methods: The population of the current study was 123 cancer-free women who had undergone cosmetic mammoplasty. The expression of lncRNAs SPRY4-IT1 and H19 was measured by quantitative real-time PCR (qRT-PCR).

Results: We indicated that the expression of SPRY4-IT1 was significantly higher in women with age more equal forty compared to women aged less than (P= 0.011). In the same way, the linear multivariate regression models showed a significant association between the high expression of SPRY4-IT1 and increasing age in women (multivariable β = 0.043, P= 0.016). Furthermore, the elevated expression of SPRY4-IT1 was also observed in postmenopausal women than that in premenopausal women (P= 0.015). However, significant difference was not obtained regarding the expression levels of lncRNA H19 in participant subjects.

Conclusion: These observations suggest that the dysregulation of breast cancer linked SPRY4-IT1 may be as an early lesion that occurs due to the effects of aging and postmenopausal factors in apparently clinically healthy women, although further investigation is needed.

Keywords: lncRNA SPRY4-IT1 expression; ageing; postmenopausal status; clinically healthy breast tissue

Association of Vitamin D Receptor Gene Polymorphisms and Clinical/Severe Outcomes of COVID-19 Patients

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Background and Aim: Growing evidence documented the critical impacts of vitamin D (VD) in the prognosis of COVID-19 patients. The functions of VD are dependent on the vitamin D receptor (VDR) in the VD/VDR signaling pathway. Therefore, we aimed to assess the association of VDR gene polymorphisms with COVID-19 outcomes.

Methods: In the present study, eight VDR single nucleotide polymorphisms (SNPs) were genotyped by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) in 500 COVID-19 patients in Iran, including 160 asymptomatic, 250 mild/moderate, and 90 severe/critical cases. The association of these polymorphisms with severity, clinical outcomes, and comorbidities were evaluated through the calculation of the Odds ratio (OR).

Results: Interestingly, significant associations were disclosed for some of the SNP-related alleles and/or genotypes in one or more genetic models with different clinical data in COVID-19 patients. Significant association of VDR-SNPs with signs, symptoms, and comorbidities was as follows: ApaI with shortness of breath ($P < 0.001$) and asthma ($P = 0.034$) in severe/critical patients (group III); BsmI with chronic renal disease ($P = 0.010$) in mild/moderate patients (group II); Tru9I with vomiting ($P = 0.031$), shortness of breath ($P = 0.04$), and hypertension ($P = 0.030$); FokI with fever and hypertension ($P = 0.027$) in severe/critical patients (group III); CDX2 with shortness of breath ($P = 0.022$), hypertension ($P = 0.036$), and diabetes ($P = 0.042$) in severe/critical patients (group III); EcoRV with diabetes ($P < 0.001$ and $P = 0.045$ in mild/moderate patients (group II) and severe/critical patients (group III), respectively). However, the association of VDR TaqI and BglI polymorphisms with clinical symptoms and comorbidities in COVID-19 patients was not significant.

Conclusion: VDR gene polymorphisms might play critical roles in the vulnerability to infection and severity of COVID-19, probably by altering the risk of comorbidities. However, these results require further validation in larger studies with different ethnicities and geographical regions.

Keywords: COVID-19; Vitamin D receptor; Single nucleotide polymorphisms (SNPs); Genetic predisposition; Clinical outcomes

CFTR Modulators and Gene Therapy as The Superior Path of Cystic Fibrosis Treatment

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Background and Aim: Cystic Fibrosis (CF) is an autosomal recessive genetic disorder caused by mutation in CFTR gene which encodes a protein channel in epithelial cell membranes and thus, results in severe damage in several organs. This study aims to examine the treatment methods of CF patients.

Methods: In this study, out of 30 articles searched in Google Scholar database in 2021, 18 articles with related keywords were selected and studied.

Results: Since the description of disorder until a decade ago, only Symptomatic treatment was available which needs continuous use. About a decade ago a more targeted treatment emerged called CFTR modulators that showed significant improvement in CFTR channel performance. However, some patients cannot tolerate this method of treatment due to factors like age, drug side effects or different or unknown genotypes and thus, need a more effective treatment such as gene therapy. Gene therapy could be done by various methods such as delivering the healthy copy to cell, modifying the mutated gene in the body using novel methods like CRISPER, genetically modifying cell outside the body and introducing it inside the body. Needless to say, these methods come with challenges such as lack of an effective gene delivery system in long term, difficulty finding the precise location for modifying gene and unclear levels of immunity in transplanted cells in out of body cell modification.

Conclusion: Symptomatic treatment has played a significant role in improving patient's condition but due to temporary nature of the treatment, the focus should be on the more targeted treatments. Challenges in the way of these novel treatments have not been resolved yet although studies and researches are trying to. Taken together, a better future for treatment of CF patients will soon arrive.

Keywords: Cystic Fibrosis; gene therapy; CFTR modulators

COVID Diagnosis Through Molecular Methods: An Overview into The Nucleic Acid Based Testing Methodologies

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Background and Aim: Molecular detection of COVID-19 relies on the amplification and detection of RNA of the SARS-CoV-2 virus. Overall, Reverse transcription polymerase chain reaction (RT-PCR) is the most common detection method which enable us robustly detect the viral RNA. The most favorable viral genes which are used to detect COVID-19 are RNA dependent RNA polymerase (RdRP), nucleocapsid (N), envelope (E), and spike (S) proteins of the virus. Although RT-PCR tests have been widely used, but these detection methods solely cannot meet the growing need of pathogen detection around the world and nowadays many alternative procedures have been established to overcome the pitfalls of TR-PCR. The main challenge in molecular diagnosis of COVID-19 is the time and also the test sensitivity. Availability of a rapid method with higher sensitivity is a golden weapon to fight against the SARS-CoV-2. Here we highlight the main issues surrounding molecular diagnosis of COVID-19, including false negatives from the detection of viral RNA, temporal variations of viral loads, selection and treatment of specimens, and limiting factors in detecting viral proteins.

Methods: We review all current methods which have been used to detect COVID-19 and discuss their advantages and pitfalls. Apart from conventional methods, other techniques, including isothermal nucleic acid amplification techniques, clusters of regularly interspaced short palindromic repeats/Cas (CRISPR/Cas) based approaches or digital PCR methods are currently used in research contexts or are awaiting approval for diagnostic use by competent authorities.

Results: Apart from conventional methods, other techniques, including isothermal nucleic acid amplification techniques, clusters of regularly interspaced short palindromic repeats/Cas (CRISPR/Cas) based approaches or digital PCR methods are currently used in research contexts or are awaiting approval for diagnostic use by competent authorities

Conclusion: Human being has learned too many lessons from COVID-19 pandemy and its molecular diagnosis which shed light on our way to be prepared for the potential coming pandemies in the future.

Keywords: COVID-19; molecular diagnosis; nucleic acid base methodologies

Evaluation of ABCC6 Expression Oscillations in A549 Cell Line

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Background and Aim: Circadian clocks play a critical role in coordinating organisms with circadian cycles. The central clock, known as the master clock, is made up of 20,000 neurons and placed in the suprachiasmatic nucleus (SCN) of the brain. The main components of the circadian clock are transcription factors which regulate the expression of their downstream genes by binding to E-box (enhancer box) elements. ABCC6 transporter belongs to the ATP-binding cassette (ABC) family and is involved in drug extrusion from the cells and resistance to chemotherapeutic agents. In this study, due to the presence of E-box sequences in the ABCC6 gene, we hypothesized that ABCC6 may be controlled by clock genes and therefore has an oscillating expression over a 24-hour period.

Methods: A549 Cell line was prepared and cultured under standard conditions. Stimulation of the clock genes was done by horse serum shock method. Then, RNA extraction and cDNA synthesis were performed. In the next step, alterations of ABCC6 expression were assessed by real time PCR. The PCR products were checked by gel electrophoresis.

Results: According to bioinformatics analysis, ABCC6 gene has several canonical and non canonical E-box elements. Our findings disclose that ABCC6 exhibit oscillatory expression pattern within 24 hours.

Conclusion: Given the presence of E-box elements in the ABCC6 sequence and its expression oscillation, it can be suggested that this gene is probably controlled by the clock genes. Therefore, due to the importance of ABCC6 in drug resistance, especially resistance to chemotherapy, more studies are needed in this field.

Keywords: Circadian clock; ABCC6; Oscillation; E-box elements

Evaluation of CircFOXO3 Gene Expression in Cancer Stem-Like Cells in A Mouse Model of Breast Cancer

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Background and Aim: Breast cancer is the most common malignancy in among females globally. Breast cancer stem cells (BCSCs) are reported to have a very important role in tumorigenesis, disease recurrence and metastasis and are resistant to current cancer therapies. The spheroid formation is a reliable laboratory method for assessing the presence and ability of CSCs. Recent evidence has shown that circFOXO3 is involved in the tumorigenesis and development of some cancers. The main purpose of this study was to evaluate the expression of circFOXO3 in spheroid-forming tumor cells in breast cancer.

Methods: After the development of an animal model of triple negative breast cancer (TNBC), the heterogeneous population of breast cancer cells were isolated from the tumor mass. The spheroid formation was then conducted among these breast cancer cells, and the qRT-PCR was performed to analyze the expression of circFOXO3 in the spheroids.

Results: Our data revealed that, compared with primary breast tumor cells, the expression of circFOXO3 was significantly downregulated in spheroid-forming tumor cells.

Conclusion: This study showed that circFOXO3 expression changed in BCSCs and targeting these circRNAs could be used for designing targeted therapeutic strategies in combat with these cells.

Keywords: circFOXO3; circRNA: Triple-negative breast cancer; cancer stem cells

Expression Analysis of An Inflammation-Related LncRNA in HLA-DRB1*15:01-Negative Patients with Multiple Sclerosis: A Probable Cause for Gender Differences in Multiple Sclerosis Susceptibility?

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Background and Aim: Multiple sclerosis is a disease that affects male and female patients differently. Several studies have been performed to explain the gender differences in MS susceptibility, but the genetic causes underlying gender differences remain unknown. The association between multiple sclerosis and the HLA-DRB1*15:01 haplotype has been confirmed to be female-specific. We hypothesized other immunological components such as lnc-DC may be gender-specific among multiple sclerosis patients, especially when MS patients are negative for the HLA-DRB1*15:01 allele. Therefore, the current study, considering the results of previous studies, aimed to evaluate the expression level of the lnc-DC gene in HLA-DRB1*15:01-negative female patients with relapsing remitting MS (RRMS).

Methods: A total of 50 MS female patients and 50 female healthy controls were enrolled in this observational case-control study. HLA-DRB1*15:01, as a critical risk factor for MS, was ruled out in all patients. The peripheral blood mononuclear cells were obtained from all patients and total RNA was isolated and cDNA synthesis was carried out. The gene expression of lnc-DC was evaluated by real-time quantitative PCR.

Results: Our results have shown that lnc-DC expression level was significantly higher in total MS female patients compared with female controls ($P = 0.0044$). In addition, the correlation between lnc-DC with disease duration, EDSS, and age at onset did not reach a statistical significance in our study ($r = 0.0336$, $P = 0.817$; $r = 0.0914$, $P = 0.5278$ and $r = 0.0743$, $P = 0.6083$, respectively).

Conclusion: Our results give further evidence that lnc-DC may play a gender-dependent role in MS pathogenesis.

Keywords: Long Non-coding RNA; HLA-DRB1*15:01; Multiple Sclerosis; Gender Differences

Expression Evaluation of APOBEC3B, COL1A1, And HLF Genes in HNSCC

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Background and Aim: Squamous cell carcinoma of the head and neck has been identified as the sixth most common cancer worldwide. APOBEC3B may be involved in cancer development because of its ability to increase genomic instability. COL1A1 is closely linked to tumors and other diseases and can regulate multiple cancers by acting as an oncogene or tumor suppressor. Some studies have suggested a vital role for HLF in regulating certain types of cancer. This study aimed to measure the relative expression of three genes (APOBEC3B, HLF, and COL1A1) in tumor tissue and healthy adjacent samples with head and neck squamous cell carcinoma and to determine their potential capacity for utilizing them as a diagnostic marker.

Methods: Processed expression data (RNA-seq) and clinicopathological data of tumor tissues of 520 patients with HNSCC and 44 healthy individuals from TCGA-HNSCC cohort were achieved for bioinformatics study and then 45 fresh tumor samples of head and neck squamous cell carcinoma and 45 healthy adjacent samples were collected. Then, RNA extraction and cDNA synthesis was conducted. Finally, the relative expression of three genes (APOBEC3B, HLF, and COL1A1) was measured by Real-Time PCR.

Results: Our findings disclosed that while APOBEC3B and COL1A1 were significantly upregulated, and the expression of HLF was decreases in HNSCC tissues comparing to normal ones. Moreover, the area under curve value for APOBEC3B, COL1A1, and HLF genes were 0/747, 0/861, and 0/767, respectively, and in a combination manner, as a three-gene panel was 0/982.

Conclusion: Our research was the first to investigate the expression of the APOBEC3B and COL1A1 genes in Iran and HLF worldwide in HNSCC. Besides, the acquired panel showed a promising diagnostic value for HNSCC.

Keywords: Head and neck squamous cell carcinoma; APOBEC3B; COL1A1; HLF

Familial Hypercholesterolemia: A Pedigree-Based Genetic Study

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Background and Aim: Familial hypercholesterolemia (FH) is a leading risk factor for premature atherosclerotic cardiovascular disease. Here, we report genetic underlying cause in a family clinically diagnosed with hypercholesterolemia.

Methods: Two affected proband from non-consanguineous parents with increased LDL-C level and multiple subcutaneous xanthomas compatible with probable FH, were referred for genetic counseling. Genomic DNA extracted and analyzed using whole-exome sequencing followed by bioinformatics. To validate the candidate variants, Sanger sequencing was done followed by PCR using specific primers. Lipid profile showed that proband and her affected brother had elevated LDL-C as 410 and 386 mg/dl, respectively. Parents had serum LDL-C and total cholesterol in borderline range. Whole exome data analysis showed two different pathogenic mutations c.443G>A and c.1592T>G in exons 4 and 11 within the LDLR gene as compound heterozygote. Both variants were co-segregated in parents, affected and healthy offspring.

Conclusion: According to the site of mutations, study patients had different phenotype severity in clinical presentations. Cascade familial screening based on genetic findings can be considered in clinical practice to identify and treat those individuals with unrecognized FH. Additional development in the understanding of the molecular basis of hypercholesterolemia will provide opportunities for the development of targeted therapy soon.

Keywords: Familial hypercholesterolemia; LDLR; Genetic; Xanthoma

Importance of DQB1 Gene and Amino Acids in The Pathogenesis, Molecular Diagnosis and Risk Assessment of Narcolepsy

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Background and Aim: Narcolepsy is a disabling neurological and a genetic disorder of autoimmune origin, which is characterized by excessive daytime sleepiness, cataplexy (sudden loss of muscle tone); sleep paralysis, and disturbed nocturnal sleep. Although there are several diagnostic tests for Narcolepsy such as MSLT (mean sleep latency test), polysomnography and low range of hypocretin in cerebrospinal fluid (CSF), but sensitivity and specificity of these techniques are not sufficient enough. Thus, methods with higher sensitivity and specificity for the accurate diagnosis and confirmation of the disease are required.

Methods: Concerning to the infrequent prevalence of narcolepsy disease, we planned a case-control association study with 20 narcoleptic patients and 150 healthy controls in a high-resolution HLA typing procedure utilizing PCR technique with sequence specific primers (PCR-SSP) and sequencing.

Results: Results: Our study reveals that the DQB1*06:02 allele presents the highest susceptibility with absolute risk of 0.13%, for Narcolepsy ($P=1 \times 10^{-14}$, $RR=60.5$, $PcPPV=0.13\%$), while, HLA-DQB1* 03:05 allele offers protection to Narcolepsy ($P= 1 \times 10^{-4}$, $PcPPV=3.19 \times 10^{-4}\%$). Moreover, for a first time, the amino acids (AA) analysis demonstrated that AA serine182 and threonine185 located on epitope of DQ β 1 chain receptor (DQB1Ser182, Thr185) provide significant susceptibility for Narcolepsy disease ($Pc= 87.03 \times 10^{-13}$, $PcPPV=0.024\%$) while, asparagine182 located on epitope of DQ β 1 protein receptor (DQB1Asn182) presents the highest protection against development of Narcolepsy ($Pc= 2.16 \times 10^{-5}$, $PcPPV=0.0012\%$). Therefore, this can be suggested that the polymorphic differences in the epitope of the HLA receptor could contribute to their differential association with the Narcolepsy in Iranian population.

Conclusion: Conclusion: Our risk assessment analysis using PcPPV formulae indicate that DQB1*06:02 allele provides the absolute risk of 0.13%, for Narcolepsy ($PcPPV=0.13\%$) in Iranian. Furthermore, our finding proposes that AA serine at position 182 and threonine at position 185 located on epitope of DQ β 1 chain which is functionally very important sit of antigen binding groove of HLA DQ β 1 receptor, play a crucial role in the development of Narcolepsy. While AA asparagine at position 182 located on epitope of DQ β 1 chain provides strong protection against the Narcolepsy disease.

Keywords: Sleep; Narcolepsy; HLA-DQB1; Polymerase chain reaction; sequence-specific primers

Investigating the Expression of EGFR Gene in Patients with Lung

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Background and Aim: Lung cancer is an illness that is caused by genetic and epigenetic variations and activates oncogenes and inactivates tumor suppressor gene. The goal of this education is to quantitative estimate of EGFR gene expression level in blood samples of lung tumor in associate with normal people examine the character of these gene as biomarkers during lung cancer judgment and screening.

Methods: This case-control study was performed on 100 blood samples of lung tumor patients related with 100 normal controls in hospital ardabil. Total RNA from Blood samples were extracted and cDNA is synthesized. In this study using from Real Time PCR.

Results: Significant increase was experiential in the expression of target biomarkers in cancer patients associated to control population. Results showed quantitative increase of EGFR gene with 4.36 respectively for lung cancer in compare with normal samples ($p=0.003$). Also, there was a significant relationship among grade of the disease and biomarkers expression level, so that with increasing the stage and degree of severity of cancer, the expression of biomarkers increased ($p=0.003$).

Conclusion: Based on this study results we could forecast the expression level of (EGFR) gen in suffered patients quantitatively which could use as biomarker indicator during screening of lung cancer samples.

Keywords: EGFR- Real Time PCR- Lung Cancer

Investigating the Relationship Between A Population Gene Pool Diversity And That Population Resistance to Coronavirus

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Background and Aim: In recent times, a new lineage of betacoronavirus; Severe Acute Respiratory Syndrome Coronavirus2 which is known as SARS-CoV-2, and as a highly contagious pathogen, has spread rapidly around the world. Under the influence of the immune system, the virus causes mutations in humans. SARS-CoV-2 uses ACE2, also a receptor for SARS-CoV for entry into the host cell. Some SNPs located in the regions encoding the ACE2 gene show different allelic frequencies in different populations.

Methods: To write this article, we used research sites such as PubMed and science direct. And we used SARS-CoV, coronavirus, betacoronavirus, gene diversity and infection keywords. And we got our data from medical and biological studies of the last few years.

Results: Variation of allelic in ACE2 receptor can be an important factor distinguishing the susceptibility to SARS-CoV-2 infection. ACE2 can be a host receptor for SARS-CoV-2. The function of the ACE2 gene is to encode the angiotensin-converting enzyme-2, which has been shown to be a receptor for the SARS-CoV and the human respiratory coronavirus NL63. Differences in AFs of several types of ACE2 coders between various populations indicate that a diverse genetic basis may affect ACE2 performance among populations. SNPs are the most common mutational events that have been identified globally and in Asia.

Conclusion: In this article, we conclude that RNA viruses have a high mutation rate owing to the lack of proofreading activity of polymerases, Due to insufficient polymerase revise activity. This immense diversity can potentially increase the adaptability of the viral population and make it difficult to eliminate. We also found that the East Asian populations have much higher AFs in the eQTL variants related to higher ACE2 expression in tissue, which may indicate different sensitivities or responses to SARS-CoV-2 from different populations under similar conditions.

Keywords: Coronavirus; SARS-CoV-2; ACE2; gene diversity; population; AFs

Microarray Datasets Analysis of Hallmark Genes in Hepatocellular Carcinoma; The Association Between Obesity and Hepatocellular Carcinoma Occurrence And Recurrence

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Background and Aim: Hepatocellular carcinoma is one of the most important leading causes of cancer-related death along the world. This study explored the potential biomarkers of hepatocellular carcinoma occurrence and recurrence, and obesity, along with the involved molecular mechanisms.

Methods: Three microarray data sets, namely GSE18897 (for genetic suppressor elements associated with obesity), GSE25097 (tumor), and GSE36376 (recurrence) were downloaded from pubmed database (Gene Expression Omnibus) and the expression of differentially expressed genes in hepatocellular carcinoma and obesity have been explored. Database for Annotation Visualization and Integrated Discovery used for identification of the functional and pathway enrichment analysis of these differentially expressed genes. The STRING online tool and Cytoscape software was used for the protein-protein interaction network analysis.

Results: We screened 160 common differentially expressed genes and found that these genes correlated to certain pathways as well as metabolic, adipocytokine signaling and terpenoid backbone biosynthesis pathways. Ten genes which are including RPS16, RPS7, CCT3, HNRNPA2B1, EIF4G1, PSMC4, NHP2, EGR1, FDPS, and MCM4 were identified in the mentioned subnetwork. According to our findings, in the GSE18897 dataset HNRNPA2B1 and RPS7 genes, in the GSE25097 dataset RPS16, RPS7, CCT3, HNRNPA2B1, PSMC4, NHP2, FDPS, and MCM4, and RPS16, RPS7, CCT3, HNRNPA2B1, EIF4G1, PSMC4, NHP2, FDPS genes, and in the GSE36376 dataset MCM4 gene showed significant positive fold changes.

Conclusion: These differentially expressed genes could be considered as diagnostic biomarkers in hepatocellular carcinoma, and so pertaining to both hepatocellular carcinoma occurrence and recurrence, and obesity.

Keywords: Obesity, Differentially Expressed Genes (DEGs); Hepatocellular Carcinoma (HCC); Occurrence; Recurrence; Microarray Datasets Analysis

Non-Invasive Prostate Cancer Diagnosis

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Background and Aim: Prostate cancer is the second most common cancer in men in the world after lung cancer. This cancer is usually silent in the early stages and does not show specific clinical symptoms until metastasis, so rapid and early diagnosis can be very important in controlling the disease and choosing targeted treatments. ctDNA is one of biomarkers has been used to early detection of prostate cancer by evaluating its specific genes.

Methods: In this research, by referring to the available data and articles, the common methods of diagnosing prostate cancer and the importance of using ctDNA and its uses in prostate cancer are discussed.

Results: The results of this discussion show that ctDNA evaluation in the near future promises promising more effective diagnostic and therapeutic methods for prostate cancer.

Conclusion: All the conventional methods that are widely used at the present in the diagnosis and screening of prostate cancer in the clinical phase have limitations that make their use difficult. Tissue biopsy is a definitive diagnostic method for prostate cancer. But in addition to the fact that solid biopsy is an invasive and dangerous procedure and increases the chances of developing urinary tract infections, it can't be used to track the response to treatment and track the progression of the disease. In the treatment process, we can't take tissue biopsies from the patient continuously because it is a painful, costly and time-consuming procedure for the person. Evaluation of ctDNAs can provide accurate and complete information about prostate cancer tumors and is a good alternative to sequential and dangerous biopsies in the diagnosis of this cancer.

Keywords: Circulating tumor DNA; Liquid biopsy; prostate cancer; Biomarker; Early cancer detection; Non-invasive

RNA-Based COVID-19 Vaccines; Advantages and Disadvantages

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Background and Aim: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is an emerging virus that is highly pathogenic and has caused the recent worldwide pandemic. Currently, considerable efforts have been put into developing effective and safe drugs and vaccines against SARS-CoV-2. Gene-based vaccines use genetic instructions, in the form of DNA or RNA, for a SARS-CoV-2 protein that stimuli an immune response and have shown promising results encompassing both humoral and cell-mediated immune responses in previous studies, supporting their implementation for COVID-19 vaccine development. In this review article, we decided to survey the advantages and disadvantages of RNA-based COVID-19 vaccines.

Methods: This systematic review was conducted to outline comprehensive studies published in PubMed, Scopus, and Google scholar databases from 2020 to 2021 by using 4 keywords 22 articles were screened, and 9 were totally included.

Results: The overwhelming advantages of mRNA vaccine over traditional vaccines include versatility and rapid development and its synthetic nature and sequence-independent manufacturing allow extremely fast and flexible vaccine design and production. Besides, the mRNA vaccine is more stable, easily translatable, rapidly producible, and usually economical. These vaccines possess much higher biosafety than DNA-based vaccines as the translation of the antigens takes place in the cytoplasm rather than the nucleus, thus much less possible for the mRNA to integrate into the genome. Low temperature and pH are vital to the preservation of DNA integrity in the long term. In contrast, RNA products are very sensitive to temperature and should always be kept at -70°C during storage and distribution. The presence of ribonucleases can destroy the RNA vaccine product; therefore synthesis, purification, and storage of the product must be done in absence of enzymes to elongate its shelf-life.

Conclusion: Despite the many advantages over the small disadvantages of the RNA-based vaccine platform, this system can be more efficient than others in producing vaccines for various diseases, especially COVID-19.

Keywords: RNA-based vaccines; COVID-19; Advantages; disadvantages

Study of EGFR Gene in Association Among Polymorphism And MiR-214 Binding Site in Lung Cancer in East Azerbaijan

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Background and Aim: Lung tumor is the most common cause of cancer-associated deaths worldwide and non-small cell lung cancer (NSCLC), particularly adenocarcinoma, is the most common kind of lung malignancy. Most cases of adenocarcinoma will happen Due to KRAS mutations or EGFR mutations and amplifications. The polymorphism rs884225 which is related with augmented EGFR expression is placed in the 3'UTR of the EGFR within the miR-214 binding site. This study goals to examine the relationship among rs884225 and lung malignancy in East Azerbaijan population.

Methods: This case-control education was showed on genomic DNA from the blood samples of 100 healthy subjects and 100 patients. After finishing the primer design, the genotypes were determined using the Tetra Primer ARMS- PCR technique.

Results: The statistical investigation was performed on the Power Marker software, and the SISA website. The frequency of the allele's C/T at rs884225 was practically similar between the East Azerbaijan population with and without Lung malignancy, and a significant relationship among this polymorphism and lung malignancy wasn't observed (CC: OR=2.28, $p>0.05$). The Odd Ratio (OR) of this polymorphism in changed kinds of lung tumor was also determined in the population that were not statistically significant (CC in NSCLC: OR=4.9, $p>0.05$).

Conclusion: It seems that no rs884225 is related with lung tumor, and for more precise conclusions, it is suggested to study in larger statistical societies.

Keywords: Lung malignancy; EGFR, polymorphism

The Fibroblast of The Association Among FGFR2 Gene Rs2981582 Polymorphism and Breast Cancer Risk in Ardabil

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Background and Aim: Breast tumor is the most common cancer kind and the leading source of tumor induced deaths in women, international. The Fibroblast Growth Factor Receptor 2 (FGFR2) is a tyrosine kinase receptor that shows a vital character in the development, invasion, and angiogenesis of cancer. Several single nucleotide polymorphisms have been originated in the intron 2 of the FGFR2 gene associated with a high risk of breast tumor. Genetic difference in this receptor is a novel risk factor for breast tumor. The present education aimed to evaluate the relationship of single-nucleotide polymorphism rs2981582C/T in women with breast tumor.

Methods: In total, 120 women with breast cancer and 120 healthy women (controls) were selected from Ardabil Hospital, Iran to contribute in this investigation. Polymorphism rs2981582 was examined to study its relationship with breast tumor. DNA extraction from blood samples was performed using a kit. The presence of these single-nucleotide polymorphisms was determined by Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR - RFLP). Statistical analyses were performed by SPSS using Chi-squared test at $P \leq 0.05$.

Results: Significant alterations were detected in the frequency of rs2981582 polymorphism in the FGFR gene among the control and patient groups ($P=0.000$). In the patient group, the TT genotype was significantly related with the risk of breast cancer ($P=0.001$; OR=3.685). On the other hand, allele C indicate a protective role against the disease ($P=0.000$)

Conclusion: The attained information exposed a significant association among rs2981582 C/T polymorphism and the risk of breast cancer; thus, this single-nucleotide polymorphism could be used as a biomarker to forecast breast cancer

Keywords: breast cancer- FGFR2

The Potential Use of GFR Signaling Against COVID-19 as A Novel Therapeutic Strategy

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Background and Aim: The coronavirus disease 2019 (COVID-19) pandemic caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is still holding on worldwide. The infection caused by SARS-CoV-2 may damage several organs. Growth factor receptor (GFR) signaling has been suggested to have a significant role in some virus infections. Epidermal GFR (EGFRs) are thought to have a role in entry of the virus and suppressing the immune response of the host. This study aims at finding a new therapeutic strategy for treatment of COVID-19. In this study, out of 48 primary articles searched in PubMed and Google Scholar databases from 2020 to 2021, 15 articles with EGFRs, SARS-CoV-2, COVID-19, GFRs keywords were selected and studied.

Results: GFR has a role in regulating a vast range of cellular tasks such as proliferation, adhesion, or differentiation. Reports show that numerous viruses utilize EGFR as an entry receptor. There are indications that many respiratory viruses could induce EGFR activation which results in suppression of interferon regulating factor-1 (IRF1)-dependent interferon λ that compromises antiviral defense in airway epithelium. Moreover, it has been reported that SARS-CoV-2 abnormally increases bioenergetic of mitochondria and activates epidermal growth factor receptor (EGFR)-mediated cell survival signal cascade to ensure survival of the SARS-CoV-2. Furthermore, SARS-CoV-2 also activates EGFR signaling cascade which is related to increased pulmonary fibrosis when over-activated.

Conclusion: Taken together, these data show the significance of GFR signaling in SARS-CoV-2 infection which could be of therapeutic use in treating COVID-19.

Keywords: EGFRs; SARS-CoV-2; COVID-19; GFRs



Physiology and Pharmacology



Anti-Inflammatory Effect of Nectaroscordum koelzi Methanolic Extract on Acute and Chronic Inflammation in Mice

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Background and Aim: Inflammation is a vital biological process and response of the immune system to maintain homeostasis. In this defense mechanism, complex events and various mediators are involved in inducing, maintaining or intensifying the inflammatory response. The present study deals with the effect of Nectaroscordum koelzi fruit extract on acute and chronic inflammation.

Methods: A total of 84 NMRI mice were used in this study. The effect of the extract on acute inflammation was analyzed by increasing vascular permeability via acetic acid and xylene induced ear edema among mice. The extract was evaluated in terms of effects on chronic inflammation by means of the cotton pellet test among mice. For the assessment of inflammation degree, the mice paw edema volume was measured by the plethysmometric test.

Results: The findings showed that the extract was effective on acute inflammation induced by acetic acid in mice. In the xylene ear edema, N. koelzi extract indicated a significant activity in mice. In the cotton pellet method, the methanol extract produced a significant reduction in comparison with the control and dexamethasone. Mice paw edema volume decreased with the extract.

Conclusion: In general, the data from the experiments indicated that the methanol extract of N. koelzi has an anti-inflammatory effect on acute and chronic inflammation. However, the exact contributing mechanisms have not been investigated for the pharmacological effects.

Keywords: Nectaroscordum koelzi; anti-inflammation; edema; extract; formaldehyde; immune deficiency

Diagnosis of Female Reproductive Diseases by Extracellular Vesicles: A review

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Background and Aim: Extracellular vesicles (EVs) play an important role in inter-cellular communication and carry various regulatory molecules, physiology and pathological conditions of reproduction.

Methods: EVs in the oviduct, uterine and follicular fluids are considered as vehicles to regulate follicular development, oocyte maturation, embryo implantation and pregnancy. EVs also play an important role in female reproductive pathologies, such as polycystic ovary syndrome (PCOS), endometriosis, and pregnancy complications.

Results: In fact, these vesicles carry different molecules that make the diagnosis of various diseases, including reproduction, without the need for biopsy. Moreover, a notable advantage of exosomes is that have higher amounts of materials for further analyses. Today, EVs might serve as diagnostic biomarkers and therapeutic targets in PCOS, endometriosis, ovarian and endometrial cancers. However, these current findings are still quite exploratory and need to be validated in clinical trials with standard protocols prior to their routine use in the clinic because of the different methods for extracting extracellular vesicles. Today, several kits have been commercialized for better and easier separation, including ExoQuick.

Conclusion: From this study, EVs may be used as biomarker to improve the early diagnosis and treatment of related diseases.

Keywords: Exosome; Ovary; Cancer; Diagnosis

Effect of Resveratrol on The Expression of TGIF- β , COL1A, α SMA Genes (fibrogenic genes), And P-Smad 2C in Palmitic Acid-Activated LX-2, A New Remedy for Hepatic Fibrosis

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Background and Aim: Background: Liver fibrosis is the result of chronic liver injury and a major cause of morbidity and mortality in the world. The present study was done to investigate the protective effect of resveratrol on liver fibrosis, focusing on the possible involvement of TGF- β signaling through Smad2. Hepatic stellate cells (HSCs) activate during chronic liver injury, express more transforming growth factor-beta (TGF- β), Collagen (COLA1), and actin-alpha smooth muscle (α SMA) that lead to hepatic fibrosis. Aim: This study investigated the antifibrotic effects of resveratrol on palmitic acid-activated human HSC and its mechanism in vitro.

Methods: Methods: First, the human HSCs (LX-2) were treated with palmitic acid (125 μ M) for 24 hours and then with resveratrol for 24 hours. Total RNAs were extracted, reversely transcribed into cDNA, Quantitative Real-time PCR and western blot were performed.

Results: Results: The mRNA expression levels of TGF- β , COL1A, α SMA genes, and the level of smad2 phosphorylation in palmitic acid-activated LX-2 treated with resveratrol were significantly reduced compared to palmitic acid-activated LX-2.

Conclusion: Conclusion: Resveratrol is effective in reducing the expression of fibrogenic genes in the activated LX-2 treated with palmitic acid through downregulation of the TGF- β /smad2 signaling pathway. Therefore, resveratrol possesses significant antifibrotic properties in hepatic fibrosis

Keywords: Keywords: Hepatic fibrosis; LX-2, Resveratrol; TGF- β /Smad2

Effect of Troxerutin (vitamin P4) on Cerebral ischemia in Male Mice

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Background and Aim: Stroke is a neurological disease and one of the leading causes of death worldwide. It is also an important cause of disability in people over age 65. It may be possible to reduce the incidence of cerebral ischemia using antioxidants, followed by inhibition of oxidative and inflammatory mechanisms. Troxerutin is a natural bioflavonoid with antioxidant and anti-inflammatory properties. However, its neuroprotective effects have not yet been studied. The present study aimed to investigate the effect of troxerutin on ischemic brain damage in male mice.

Methods: In this experimental study, 39 male mice (30-25 grams) were used. The animals were divided into three groups: 1. Sham group 2. Ischemic brain group 3. The ischemic brain group, receiving troxerutin drug. Cerebral ischemia was induced in the second and third groups of mice. Five hours later, group 3 received 300 mg/kg troxerutin intraperitoneal injection. After 24 and 48 hours, the effect of troxerutin on sensory and motor neurological disorders, cerebral edema, and infarct volume was investigated. Analysis of variance test was performed by SPSS software (21) using one-way and two-way.

Results: The results indicated that troxerutin reduced stroke volume (P less than 0.05) and cerebral edema (P less than 0.05) compared with the control group. Troxerutin also improved sensory-motor function (P less than 0.0001) and balance function and muscle strength (P less than 0.05).

Conclusion: Treatment with troxerutin can have beneficial effects on complications and disorders caused by cerebral ischemia.

Keywords: Cerebral ischemia; Troxerutin; Antioxidant

Evaluation of The Effect of ketamine on Expression of Tumor Necrosis Factor-Alpha and Interleukin-10 Genes in Leukocytes

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Background and Aim: Ketamine is primary used in clinical settings for induction and maintenance of anesthesia. At sub-anesthetic doses, this drug is also a promising agent for pain management and treatment-resistant depression. Results from recent studies indicate that ketamine can affect the immune system. The aim of this study was assessing the effect of ketamine on gene expression of tumor necrosis factor-alpha and interleukin-10 in leukocytes.

Methods: Ten Wistar rats were reared in an isolated animal house. Leukocytes were isolated from spleens of rats and cultured in RPMI1640 culture medium supplemented with fetal bovine serum. Ketamine was added to cells at different concentrations. Phosphate buffered saline was added to the control cells (control group) instead of ketamine. To evaluate gene expression levels in leukocytes, mRNAs were extracted from leukocytes after 48h of culture. Then, cDNA was synthesized from mRNAs using reverse transcriptase enzyme. Gene expression levels were measured by SYBR green-based real-time RT-PCR assay.

Results: Evaluation of RNAs extracted from leukocytes using the Nano Drop spectrophotometer showed that RNAs were extracted with appropriate quality and quantity. Real time RT-PCR data analysis showed that tumor necrosis factor-alpha gene expression in ketamine-exposed leukocytes at concentrations of 100 and 500 ng/ml was decreased 0.91-fold ($p>0.05$) and 0.3-fold ($p<0.05$) and decreased by 0.79 times ($p>0.05$) in leukocytes cultured in the presence of ketamine with concentration of 100 and 500 ng/ml, respectively.

Conclusion: These findings suggest that ketamine can have anti-inflammatory effects by reducing the gene expression of tumor necrosis factor-alpha in leukocytes. Ketamine can also affect interleukin-10 gene expression in leukocytes in a dose-dependent manner.

Keywords: Ketamine; Leukocytes; Gene expression; Tumor necrosis factor-alpha; Interleukin-10

Gut-Brain Axis Modulation with *Lactobacillus Acidophilus*: A Novel Therapy in Animal Model of Autism

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Background and Aim: Autism Spectrum Disorder (ASD) is recognized by difficulties with social interaction along with restricted and repetitive behaviors. Influences of microbiome on brain development and behavior is called microbiome-gut-brain axis. Gut microbiota has shown alternations in ASD patients and their mothers. Modulation of gut microbiota with probiotic administration is very renowned in treatment and improvement of diseases. The aim of our study was to investigate serum biochemical parameters, anatomical and behavioral changes in animal model of autism and estimating the effects of different doses of *Lactobacillus acidophilus*, as a probiotic therapy, on the forenamed parameters

Methods: *Lactobacillus acidophilus* was cultured daily and was administered orally at low and high doses to adult female rats, which had received Valproic acid on 12.5 prenatal day, from first day of pregnancy until post-natal day 25 to measure the effects of probiotic on improvement of autistic abnormalities in their offspring. After 50 days of administration, the rats were weaned and behavioral, biochemical and anatomical tests were performed and compared with control groups.

Results: Significant behavioral, biochemical, and anatomical improvements were observed in autistic litters of probiotic receiving dams with high doses during pregnancy and lactation periods in comparison to the control group.

Conclusion: Probiotic administration may be considered as an attenuating or therapeutic factor for autistic patients.

Keywords: Autism; Valproic acid; Probiotics; *Lactobacillus acidophilus*

Oxidative Stress and Animal Model of Autism: Is There A Link?

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Background and Aim: Autism spectrum disorder (ASD) is characterized by impaired social interactions as well as stereotype sensory–motor behaviors. The valproic acid (VPA) animal model of autism is one of the most broadly used animal models in terms of simulating ASD symptoms. Intraperitoneal injection of valproic acid on day 12.5 of gestation in rats will cause autism-like symptoms in newborns. The aim of this study was to investigate oxidative stress profile in animal model of Autism.

Methods: pregnant rats were divided equally into control and treatment groups. On day 12.5 of conception, the treatment group received 600 mg/kg valproic acid (dissolved in saline at a concentration of 250 mg/ml) while the control group received normal saline. After weaning of newborns (20 rats), sera samples were isolated. As indicators of oxidative stress, Superoxide dismutase (SOD), Catalase (CAT) and Malondialdehyde (MDA) levels in sera samples were evaluated in both groups using commercial kits

Results: The results of the present study revealed that serum levels of SOD and CAT is significantly lower in VPA group whereas MDA level is higher.

Conclusion: The results of the present study revealed that serum levels of SOD and CAT is significantly lower in VPA group whereas MDA level is higher.

Keywords: Autism; Animal Model; Oxidative Stress

Renal Clearance and Urine Output Reduced by Angiotensin 1-7 Receptor Antagonist in Hypertensive Estradiol Treated Rats

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Background and Aim: Backgrounds: Renin-angiotensin system (RAS) is one of the most essential systems to maintain blood pressure and body fluid volume. Angiotensin (Ang) 1-7 antagonizes vascular and tubular actions of Ang II and exerts its vasodilatory, natriuretic, and diuretic effects by activating the Mas receptor (MasR). In animal models of hypertension such as two-kidney-one-clip hypertensive (2K1C), Ang 1-7 prevents the development of hypertension. It is known that the RAS component may up-regulate by estrogen, and intratubular Ang converting enzyme 2 (ACE2)/Ang 1-7/ MasR axis contributes to protecting female rats from hypertension. The main objective of this study was to investigate the role of MasR antagonist (A779) on renal function in normotensive and 2K1C rats with or without estradiol therapy.

Methods: Method: A total of 32 female 7-9 weeks Wistar rats underwent ovariectomy, based on received high-dose estradiol therapy (500 µg/ kg/ weekly/ IM) or placebo and subjected to 2K1C or non-2K1C, were divided into 4 groups (each group was divided into the two subgroups which received A779 or its vehicle). Four weeks later, the rats were anesthetized, and the trachea was cannulated. Jugular vein, carotid, and femoral arteries were catheterized for drug infusion, mean arterial pressure (MAP), and renal perfusion pressure (RPP) measurement, respectively. A catheter was placed in the bladder to collect urine. Subsequently, creatinine clearance (CrCl) and urine flow (UF), were determined.

Results: No significant differences in renal perfusion pressure (RPP) were detected in the base and 30 min post A779 administration in each group. However, in both normotensive and hypertensive rats either treated with estradiol or vehicle, CrCl, and UF decreased after the administration of MasR antagonist.

Conclusion: The MasR agonist might be applied as a new pharmacological agent to increase UF and CrCl in normotensive and hypertensive subjected with and without estradiol therapy.

Keywords: Renin-angiotensin system (RAS); Two kidney-One Clip (2K1C); Mas receptor; Estradiol; Renal function

Renin Angiotensin System Induced Secondary Hypertension: The Alteration of kidney Function and Structure

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Background and Aim: Long-term hypertension is known as a major risk factor for cardiovascular and chronic kidney disease (CKD). Renin angiotensin system (RAS) plays a key role in hypertension pathogenesis. Angiotensin II (Ang II) enhancement in Ang II dependent hypertension led to progressive CKD.

Methods: Based on a variety of sources including PubMed, Google Scholar, Scopus and Science-Direct, in current review we will discuss the role of RAS induced secondary hypertension on the alteration of renal function.

Results: In two kidney one clip model (2K1C) more renin is synthesized in the principal cells of collecting duct than juxtaglomerular cells (JGC). An increase of renal Ang I and Ang II levels, and a decrease of renal cortical and medullary of Ang 1-7 occur in both kidneys of 2K1C hypertensive rat model. Also change the balance in production of renal ACE, ACE2 and PrCP expression. Intra-renal angiotensinogen (AGT) expression and urine AGT (uAGT) excretion rates in unclipped kidney are greater than clipped kidney in 2K1C hypertensive rat model. The enhancement of Ang II in clipped kidney is related to renin secretion, while the elevation of intrarenal Ang II in unclipped kidney is related to stimulation of AGT mRNA and protein in proximal tubule cells by direct effect of systemic Ang II level. Ang II dependent increases cytokines, which stimulate AGT synthesis in proximal tubules. Accumulation of inflammatory cells in the kidney aggravates hypertension and renal damage. Moreover, Ang II dependent hypertension alters renal Ang II type 1 & 2 receptors (AT1R & AT2R) and Mas receptor (MasR) expression, and the renal interstitial fluid bradykinine, nitric oxide, and cGMP response to AT1R, AT2R, or BK B2-receptor antagonists.

Conclusion: Ang II dependent hypertension affect local renal RAS components and function.

Keywords: Renin angiotensin system; Hypertension; Renal function

Sex Differences in The Renal Vascular Functions of Mas Receptors in Two-kidney-One-Clip Hypertension

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Background and Aim: Hypertension is a complex condition which induces abnormalities such as renal dysfunction. The prevalence and severity of hypertension is gender dependent with more symptoms in males than in females. Also, the activity of renal renin angiotensin systems (RAS), are gender related. In this study we investigated sex differences in renal vascular responses to angiotensin II (Ang II) administration without Ang II type 1 and Mas receptors antagonists in the 2K1C rat model of renovascular hypertension.

Methods: Male and female 2K1C rats were divided into 4 experimental groups (2 of each sex) treated with losartan or A779+losartan. Responses of mean arterial pressure (MAP), renal blood flow (RBF), renal vascular resistance (RVR) to Ang II were determined.

Results: No gender differences were observed when the AT1R was blocked. While The RBF response to Ang II in A779+Losartan groups was gender related. With the stronger RBF response to Ang II in A779+Losartan 2K1C female treated groups.

Conclusion: The stronger RBF response to Ang II in A779+Losartan 2K1C female treated groups related to greater distribution and function of ACE2/Ang 1-7/MasR arm of the RAS in female 2K1C rats. Therefore, rather than blocking the ACE/AngII/AT1R axis, activating the ACE2/Ang 1-7/MasR axis of the RAS may be more effective in improving the renal and systemic side effects of Ang II dependent hypertension in the female but not in male.

Keywords: Renal blood flow; angiotensin II; renovascular hypertension; gender



Toxicology



Caster Bean Ingestion and Hepatic Enzymes: A Case Report

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Background and Aim: Ricin is a protein toxin derived from the castor bean plant *Ricinus communis*. (1) Ricin poisoning has been reported after inhalation, parenteral injection, and oral ingestion. Successful absorption of ricin after oral ingestion is determined by multiple variables. (2). > 1000 ricin poisoning cases secondary to intentional castor bean consumption have been re-reported in the literature since the late 1800s. (3)

Methods: Case report: We present a case, A 27-year-old man, was referred to the toxicology emergency department, that highlights ingestion of seventeen castor beans together 3 hours before admission. On admission, the physical examination found normal and the patient had symptom of nausea and vomiting. His primary blood tests showed in normal range, chest x-ray and ECG were also normal. Taken to him early management (oral charcoal therapy, symptom therapy, hydration, an antiemetic therapy and a supplementation of hydro electrolytic losses), Within the first 24 h after admission, the patient resumed his normal state, the discharge was possible. Two days later the patient was readmitted with abdominal pain and in blood test, hepatic enzyme was elevated. In physical examination, vital signs were in normal range, abdominal examination was normal, without tenderness or guarding. Treatment with intravenous (dose of 300 mg/kg/24 h) NAC (N-acetyl cysteine) as an anti-oxidant for hepatic intoxication was started and after 72 hours the patient discharged with normal range of hepatic enzyme and without any abnormal symptom.

Conclusion: The toxicity of ricin is dependent on the dose delivered and the route of the exposure. Supportive care and correction of laboratory disorders is the mainstay of treatment. As shown in our case, early management is crucial in symptomatic patients and management with intravenous NAC is beneficial for a good outcome in cases with elevated liver enzyme.

Keywords: castor bean; ricin; intoxication; poisoning; emergency; liver enzyme

Clinical And -Epidemiological Evaluation of Patients with Pesticides Poisoning: A Five Years Cross Sectional Study in The Referral Poisoning Center

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Background and Aim: Epidemiological information is important to prevent the potential risks of poisoning. The aim of this study was to evaluate the epidemiological pattern of pesticide poisoning by collecting and reviewing statistics and preliminary information.

Methods: This retrospective study was conducted in poisoning referral center in khorshid hospital, affiliated with Isfahan University of Medical Sciences, Isfahan, Iran. The data of 1567 cases of pesticide poisoning (fungicides, herbicides, insecticides, acaricides, rodenticides and unknown pesticides) who hospitalized between 2016 to 2020 including demographic, toxico-clinical information, and outcome variables (survive, complication and death) was collected. All information was analyzed based on outcome variables.

Results: Mean age of the patients was 31.38 ± 13.76 . 867 (55.3%) cases were men. Insecticides (51.3%), and rodenticides (30.3%) poisoning were the most common. 90.4% patients recovered without complication. There was a significant relationship between gender, job, type of exposure, endotracheal intubation with outcome of the patients (P value < 0.05). The insecticides (P value, $=0.01$), herbicides and fungicides (P , 0.03) and rodenticides (P , 0.006) based on the gender showed a significant correlation with the outcome. This relationship was also observed in patients with a history of psychiatric problem (P value < 0.05). In addition, clinical manifestations with respect to cardiovascular in insecticides poisoning (P , 0.040) and respiratory problems in herbicides (P , 0.042), and in rodenticides (P , 0.004) had a significant correlation with outcome.

Conclusion: Some demographic factors such as gender, job, type of exposure and clinical manifestations especially cardiovascular and respiratory systems had a significant relationship with outcome of patients with pesticide poisoning.

Keywords: Poisoning; pesticide; fungicide; herbicide; insecticide; rodenticide; outcome

COVID-19 And Nicotine: A Review of The Evidence

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Background and Aim: The ongoing COVID-19 epidemic has focused on infectious diseases and their associations with host factors and underlying conditions. Taking into account the role of cigarette smoking in the pathogenesis of lung diseases and the effect of nicotine on expression of the entrance receptor for extreme acute respiratory syndrome coronavirus 2 (SARS-CoV-2), it is significant to determine the implications of smoking in COVID-19. This review examines recent data on a potential relationship between smoking and COVID-19.

Methods: Articles describing the association of tobacco use with COVID-19 were searched on PubMed and Google. This review covers the relevant studies on the subject published from January 1, 2020 to October, 2021.

Results: From analysis of the available studies to date, it seems that tobacco use in all forms, is significantly linked with severe COVID-19 outcomes. Pre-existing comorbidities in tobacco users such as cardiovascular diseases, diabetes, respiratory diseases and hypertension were found to further exacerbate the disease demonstrations. Considering the limitations of the study, the results should be construed with great caution and be viewed just as a initial report to motivate related basic and clinical researches.

Conclusion: Nicotine exposure is associated to cardiopulmonary vulnerability to COVID-19 and tobacco use can be a potential risk factor for not only getting the viral infection but also its severe demonstrations as a result Smoking is an independent risk for having progression of COVID-19, including mortality.

Keywords: COVID-19; Nicotine; Smoking; Coronavirus Disease

Cytotoxicity of Crude Venom from *Cassiopea Andromeda* Against Human Breast Adenocarcinoma Cell Line (MDA-MB-231)

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Background and Aim: *Cassiopea Andromeda* is a venomous animal from cnidarian species called the upside-down jellyfish. Venoms have long been traditionally used among people. Researchers have done extended studies on venoms to find Active and functional compounds to apply in new therapies. MDA-MB-231 is an invasive breast cancer cell line that is negative for estrogen receptor (ER), progesterone receptor (PR) expression, and HER2 (human epidermal growth factor receptor). In this study, the Cytotoxicity of *Cassiopea Andromeda* crude venom against MDA-MB-231 was evaluated.

Methods: Cells were cultured in 96-well plates (6000 cells/ well) containing 100 μ l medium. After 24hrs the cells were treated with different concentrations of crude venom for 72 hrs. The metabolic activity of the cells was determined by the MTT assay. After removal of 100 μ l medium, MTT dye solution was added (15 μ l / 100 μ l medium) and the plates were incubated at 37°C for 4 hrs. After that, 100 μ l of DMSO were added to each well and mixed to dissolve the dye crystals. The absorbance was measured at 570 nm with a reference wavelength of 630 nm.

Results: The 50% inhibition concentration (IC₅₀) is a measure to determine the concentration which reduced cell growth by 50% in treated cells compared to untreated. The IC₅₀ value obtained for the crude venom of *Cassiopea Andromeda* was approximately 625 μ g/ml.

Conclusion: According to the results of the present study, *Cassiopea Andromeda* venom was cytotoxic in MDA-MB-231 cell line and it can be used in subsequent pharmaceutical studies.

Keywords: *Cassiopea Andromeda*; MDA-MB; Cytotoxicity

Descurainia Sophia Attenuates Gentamicin-Induced Renal Insufficiency by Reducing Inflammation, and Apoptosis in Rats

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Background and Aim: Recent studies have tried to suggest an efficient agent to prevent gentamicin (Gm) induced renal failure. In this study, we evaluated the potential renoprotective effects of Descurainia sophia against Gm-induced nephrotoxicity in rats.

Methods: Forty rats were categorized in five groups (n = 8 per group). Control group was treated with normal saline. In four experimental groups, the rats were initially treated with normal saline (A), 800 (B), 1600 (C) and 2400 (D) mg/kg Descurainia sophia respectively for four weeks. Then, the animals of experimental groups were administered with Gm (80 mg/Kg) for 7 consecutive days.

Results: Serum BUN level significantly increased in group 1. However, pretreatment with either 800, 1600 or 2400 mg/ kg of Descurainia sophia dose-dependently prevented elevation of BUN following Gm treatment in Wistar rats with the most efficient dose that was 2400 mg/Kg. The rats in group 1 also had significantly increased serum creatinine level while those received various doses of Descurainia sophia showed significantly lower creatinine level with the lowest level observed in 2400 mg/kg group. The rats received normal saline showing significant increase in urinary Na⁺ concentration and fractional excretion of sodium (FeNa⁺). On the other hand, pretreatment with either 800, 1600 or 2400 mg/Kg of Descurainia sophia significantly decreased urine Na⁺ concentration and FeNa⁺ at all the studied doses. Serum BUN, creatinine, cholesterol, and triglycerides level, as well as urinary excretion of Na⁺ significantly increased in group A. Furthermore, Gm induced inflammatory cells infiltration, apoptosis, and renal cells injuries in rats were pretreated with normal saline. However, in the animals pretreated with Descurainia sophia extract there were significant and dose-dependent reductions in blood and urine markers, apoptosis rate, and inflammatory cells infiltration in renal tissues.

Conclusion: Overall, Descurainia sophia showed significant renoprotective effects against Gm-induced AKI by alleviating biochemical and histological markers of renal toxicity.

Keywords: Gentamicin; Nephrotoxicity; Inflammation; Apoptosis

Determination of Aflatoxin B1 in Animal Feed in Dairy Farms of Sabzevar District (Northeast Iran) During 2020

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Background and Aim: Aflatoxins are a class of carcinogenic mycotoxins produced by *Aspergillus* fungi and are known to contaminate a large portion of the world's food supply. Aflatoxin B1 (AFB1) is the most potent of these compounds and has been well-characterized to lead to the development of hepatocellular carcinoma (HCC) in humans and animals. This study was designed and performed to determine the aflatoxin B1 in livestock feed of farms around the Sabzevar.

Methods: There are about 360 farms around the Sabzevar that were divided into two groups: large (industrial) and small (traditional) farms. The sampling was performed from different food groups (concentrate, dry bread and dry forage). In order to measure aflatoxin B1 in animal feed, tests were performed by ELISA aflatoxin B1 kit (R-Biofarm, Germany) according to the manufacturer's protocol.

Results: The average amount of aflatoxin B1 in the feed of industrial and traditional farms was 14.9 ± 2.99 ng / g and 13.6 ± 2.72 ng / g, respectively.

Conclusion: The results of this study showed that all samples of concentrate, dry forage and dry bread collected from traditional and industrial farms were contaminated and the level of contamination was higher than the national standard of Iran (5 ng / g).

Keywords: *Aspergillus*; Aflatoxin B1; Animal feed

Evaluation of Secondary Infections in Patients with COVID-19

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Background and Aim: Coronavirus is single-stranded, enveloped RNA virus that belongs to the beta group Covid-19, which attacks the body's respiratory system and causes infection. Common symptoms generally include fever, cough, fatigue, muscle aches, gastrointestinal disorders, olfactory system, heart and etc. The purpose of this article is to review secondary infections in patients with Covid-19.

Methods: In this study, the relevant information was searched from Pubmed, Scopus, Google Scholar and Magiran databases. Data analysis was performed qualitatively.

Results: Studies show that Covid-19 has long-term effects on the patient's body. Secondary infection such as pneumonia is one of the most important viral complications associated with this disease. Research shows that a small number of patients develop secondary bacterial infection. Factors that contribute to the exacerbation of secondary infection, especially bacterial ones, include Corticosteroids, weakened immune systems, and prolonged hospitalization

Conclusion: Patients with Covid-19 are generally less likely to develop secondary bacterial infection. Disinfection of the environment plays an important role in reducing the incidence of infection caused by drug-resistant pathogens; In addition, antibiotics cause changes in the mucus, so antibiotics should be used with caution during treatment.

Keywords: Covid-19; secondary bacterial infection; antibiotics.

Evaluation of The Thrombocytopenia in Patients with Acute Pesticides Poisoning

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Background and Aim: Most deaths in the world due to pesticide poisoning have been reported in developing countries, and due to the declared hematotoxicity and thrombocytopenic properties of these toxins. In this study, we investigated the causes of thrombocytopenia in patients with acute poisoning with pesticides.

Methods: In this retrospective cross-sectional study, 51 patients with acute pesticides poisoning (insecticides, herbicides, and fungicides, rodenticides, acaricides) who were admitted to the emergency department of Khorshid Hospital during 2017 to 2021, and had thrombocytopenia during hospitalization were evaluated. All of the information including demographic information, clinical, paraclinical, treatment protocols and their outcome (survived or death) were recorded in data gathering forms.

Results: fifty-one patients (36 men and 15 women) with a mean age of 33.47 ± 13.67 years [min-max: 16-77] years were studied. Most cases of poisoning occurred in autumn (17%). The route and type of exposure was orally (49%), and intentionally (45%). The most common pesticides were insecticides (21%), followed by herbicides and fungicides (15%) and rodenticides (14%), respectively. Thrombocytopenia was presented most often in patients with moderate severity of insecticide poisoning. (15.7%) There was a significant relationship between the administration of N-acetylcysteine, vitamin C, vitamin E, corticosteroids, norepinephrine, dialysis procedure, and ICU treatment with the outcome. The mean platelet count at admission was 167.47 ± 48.64 . There was no significant relationship between the trends of patients' blood platelets during hospitalization with their outcome.

Conclusion: Thrombocytopenia was presented most often in patients with insecticide poisoning. There was no significant relationship between the trend of patients' blood platelet during hospitalization with their outcome.

Keywords: pesticides; thrombocytopenia; poisoning; outcome; insecticide

Evaluation The Cytotoxicity of Recombinant ML1-STxB Protein in Free and Encapsulated Chitosan Nanoparticles on Skbr3 Cell Line

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Background and Aim: Targeted toxins are a new class of therapeutic proteins that consist of an antibody, a growth factor or a ligand conjugated to either a plant toxin (Ricin, Mistletoe lectin) or a bacterial toxin. Ribosome inactivating proteins (RIPs) are toxic proteins with characteristic N-glycosidase activity. They remove a specific adenine (depurination) from the 28S rRNA of the eukaryotic ribosome, preventing its binding with elongation factor, and thereby inhibit translation. One of these proteins is called ML1 (Mistletoe lectin 1) which is a member of RIP 2 family. These combinations can be used as active biological factors and anticancer because of their ability in destroying cells by stopping protein production. StxB (Shiga toxin B-subunit) connects to its own cell receptor, globotriaosylceramide (Gb3). There is Gb3 On the surface of many cells in our body. The expression of Gb3 on the surface of cancer cells is abundant. The use of chitosan nanoparticles is in the delivery of peptide drugs. The use of peptides is expanding because of its smaller size, less toxicity and easier passage through blood vessels. We use this nanoparticle for the above reasons in this study to reduce antigen cytotoxicity

Methods: In this experimental study, the recombinant ML1-STxB protein was expressed in E.coli and purified by affinity chromatography on nickel column. ML1-STxB was then encapsulated using ionic gelation method in chitosan nanoparticles. Finally, the ML1-STxB cytotoxicity and the obtained nanoparticles were performed using standard MTT assay on skbr3 cell line.

Results: The results of mtt assay at 20, 40 and 60 ng/μl dilutions showed lower toxicity of recombinant antigen loaded in chitosan nanoparticles than free recombinant antigen. Finally, there was a reverse relationship between the antigen concentration and the nanoparticles used and the viability of the test cells.

Conclusion: According to the results of this study, which shows the cytotoxicity of the recombinant ML1-STxB protein can be reduced by encapsulating this protein in the nanoparticles, thus we can use the chimeric as the anticancer drug candidate.

Keywords: toxins; cytotoxicity; ML1-STxB;skbr3

Investigation of Heat Effect on Production of Polycyclic Aromatic Hydrocarbons During the Production of Infant Formulae

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Background and Aim: In the present study, we aimed to investigate effect of heat on generation of four priority polycyclic aromatic hydrocarbons (PAH4) during the production of infant formulae. Benzo (a) anthracene (BaA), chrysene (Chr), benzo (b) fluoranthene (BbF) and benzo (a) pyrene (BaP) are the most important PAHs, and their sum has been defined as PAH4. It has been shown that PAHs may produced by pyrolysis of organic substances. The International Agency for Research on Cancer has categorized PAH4 as carcinogen (BaP) and possible carcinogen (BaA, Chr, and BbF).

Methods: Identification and quantification of PAH4 were accomplished by use of GC analyses with a Agilent 6890 N (Agilent, Waldbronn, Germany) gas chromatograph, coupled to a Agilent 5975 quadrupole mass selective spectrometer. Level of PAH4 was determined in raw milk, pasteurized milk, feed tank, mixing tank, and final products. Samples were obtained from production line of company.

Results: findings revealed that pasteurization process not affected level of PAH4. However, homogenization led to significant increase in PAH4 level (p value less than 0.01). Also, it was observed that the level of PAH4 diminished during the spray drying of final product (p value less than 0.01)

Conclusion: heat treatment during the homogenization process increased the PAH4. In addition, direct steam injection led to decrease in PAH4 level.

Keywords: PAH4; Infant formula; Heat; Milk powder; Gas chromatography mass spectrophotometry (GC-MS).

Manufacture And Characterization of Curcumin And Quercetin Nanosystem in Gastric Cancer

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Background and Aim: Gastric cancer is the most deadly gastrointestinal cancer. The side effects of current treatments are considered the main challenge. The two natural compounds curcumin and quercetin show multi-functional anti-cancer activity, but with low efficacy in the aqua environment. Therefore, this project was performed to investigate the toxic effects of nano-system containing curcumin and quercetin on gastric cancer cells.

Methods: Initially, nano capsulation of these two compounds was made. Then, the effect and toxicity of nano-system containing curcumin and quercetin on gastric cancer cell line were performed. The 50% lethal dose was calculated for nano-composites containing curcumin and quercetin, to check the gene expression of Wnt.

Results: The IC₅₀ level for the Nanosystem containing curcumin and quercetin in gastric cancer cells was 54.27 micromolar / ml. Gene expression was also reduced in the treated group.

Conclusion: The results of our study showed the efficacy of this nano system in the induction of cell death in gastric cancer cell lines with two combinations of curcumin and quercetin.

Keywords: gastric cancer; curcumin; quercetin; IC₅₀

Potential Drug-Drug Interactions in Patients with COVID-19

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Background and Aim: Patients with corona virus 2019 (COVID-19) are significant examples in terms of drug-drug interaction (DDI) and the effects of poisoning (leading to a range of toxicity to death or reduced efficacy). Many therapists in the treatment of this disease, especially in the beginning, have resorted to a variety of experimental drugs (polypharmacy). Also, most of the hospitalized patients include the elderly with various underlying diseases and as a result receive various medications. The aim of this study was to evaluate the risk of poisoning due to drug-drug interactions in patients with Covid-19.

Methods: Drugs were evaluated for DDI using drug references (UpToDate and Katzung 2021 Clinical Pharmacology). A wide variety of herbal medicines, blood products and laboratory studies were eliminated. In addition, potential drug interactions due to known pharmacokinetics and overlapping toxicities have been evaluated in studies since the beginning of the pandemic at Pubmed and Scopus sites.

Results: According to studies, the average number of drugs prescribed for patients with COVID is 5 drugs. Patients who have been prescribed at least 5 medications are at higher risk for DDI. About two-thirds of patients are exposed to at least one potential DDI (mainly by lopinavir / ritonavir and hydroxychloroquine). Victims of some of the metabolic effects (P450 cytochrome and changes in drug metabolism) as well as the causes of QT prolongation are most at risk. Hyperglycemia, gastrointestinal disorders, skin reactions, and increased sedative effect were the main clinical complications (prevalence less than 10%).

Conclusion: Drug and drug interactions are common in COVID-19 patients due to both comorbidities and polypharmacy. By considering these DDIs and managing the administration of drugs and the use of alternative drugs or dose adjustment, efforts can be made to minimize the risk of these potentially harmful drug combinations.

Keywords: COVID-19; Drug-drug interaction; Poisoning; Risk factors

Protective Effects of Natural Compounds Against Paraquat-Induced Pulmonary Toxicity: The Role of Nrf2/ARE Signaling Pathway

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Background and Aim: Paraquat (PQ) is a quaternary nitrogen herbicide that is highly toxic to humans. Once absorbed, PQ predominantly accumulates in the lungs. It has well documented that generation of reactive oxygen species (ROS) is the main mechanism of PQ toxicity.

Methods: Oxidative damages of PQ in lungs are represented as generation of cytotoxic and fibrotic mediators, interruption of epithelial and endothelial barriers, edema, collagen deposition and inflammatory cells infiltration. Up to now, no effective treatment for PQ has been introduced.

Results: Cumulative studies have shown that natural compounds (NCs) such resveratrol and silymarin reveal protective effects against PQ-induced pulmonary toxicity, due to its antioxidant, and anti-inflammatory effects.

Conclusion: Based on various studies, NCs function as protective agent through stimulation of nuclear factor erythroid 2-related factor 2 (Nrf2)/antioxidant response element (ARE) signaling pathway. Nrf2/ARE signaling pathway is the most important endogenous antioxidant pathway that discovered to date. Thus, we aimed to review the role of Nrf2/ARE signaling pathway in protective effects of NCs against PQ-induced pulmonary toxicity.

Keywords: Paraquat; Natural Compounds; Protective; Toxicity; Nrf2



Medical Physics



Investigating The Correlation Between HbA1c, Dyslipidemia And liver Enzymes in Healthy, Prediabetic, Diabetic and Uncontrolled Diabetic Individuals

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Background and Aim: Diabetes mellitus (DM) characterized by chronic hyperglycemia, contributes to a high risk of metabolic disorders. Glycemic hemoglobin (HbA1c) reflects average plasma glucose over the previous 8–12 weeks. The liver plays an important role in maintaining glucose homeostasis. We investigated the relationship between HbA1c, serum lipid profiles, and liver enzymes in healthy, pre-diabetic, diabetic, and uncontrolled diabetic individuals.

Methods: Thirty hundred controls with HbA1c lower than 5.7%, 300 prediabetics (HbA1c from 5.7 to 6.4%), 300 types 2 diabetic (HbA1c >6.5% and 300 uncontrolled diabetics (HbA1c >8%), were studied. The sera were analyzed for (FBS), high-density lipoprotein cholesterol (HDL-C) and low-density lipoprotein cholesterol (LDL-C), total cholesterol (TC), triglycerides (TGs), alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), lactate dehydrogenase (LDH), γ -glutamyltransferase (GGT). Then were analyzed by SPSS.

Results: our study showed that there were significantly lower levels of HDL and Significantly higher levels of TC, TG, LDL, FBS, GGT, and ALT in diabetic and uncontrolled diabetics compared to healthy and prediabetic individuals.

Conclusion: Our results indicated significant increases in lipid parameters, ALT, GGT with increasing HbA1c in diabetic and uncontrolled diabetic, so HbA1c can be used as a good parameter for predicting dyslipidemia. Diabetic patients should do routine monitoring of blood glucose and serum lipid profiles because controlling blood sugar at early stages can reduce the risk of metabolic diseases.

Keywords: Keywords: Diabetes mellitus; HbA1c; serum lipid profiles; dyslipidemia; liver enzymes

Study of Radioprotective Effect of Ergocalciferol in Peripheral Blood Lymphocytes Using Annexin v Apoptosis Assay

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Background and Aim: The wide use of radiation therapy for cancer patients in the recent years has led us to the fact that one of the most vulnerable cells to radiation is in fact lymphocytes and this may seriously impact and impair the immune system and immune response of patients after radiation therapy. A few radioprotection agents have been suggested to reduce the overall damage to lymphocytes. Over the last years studies have shown the effectiveness of cholecalciferol (vitamin d3) as a radioprotective agent in lymphocytes which resulted in the increased radiation tolerances and reduction of DNA damage. However, the effects of ergocalciferol (vitamin d2) as a radioprotective agent remains unknown. Ergocalciferol is widely available through supplements and prescribed for hypoparathyroidism.

Methods: herein we investigate radioprotective effect of ergocalciferol via annexin v test after 2Gy radiation based on the fact that vulnerability of the lymphocytes to the radiation will cause higher apoptosis rate in the cells.

Results: It appears that ergocalciferol could potentially impact the radio resistance of the lymphocytes and immune response of radiation therapy patients.

Conclusion: If the radio resistance of the ergocalciferol will be proved it will be a huge help for radiation therapy patients as an available agent.

Keywords: Radiation therapy; Radiation resistance; Vitamin D



Trace elements



Aluminum Toxicity Impact on Serum Phosphorous and Neutrophil Count in End-Stage Renal Disease Patients Hospitalized in A City in Markazi Province

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Background and Aim: Aluminum (Al) toxicity is a complication of renal failure in end-stage renal disease (ESRD) patients. But we have not precise information about Al status among different Iranian population due to underestimating the importance of trace element's role in patient's health and well-being. We aimed to evaluate the Al toxicity in hemodialysis patients hospitalized in a university hospital in Khomein city.

Methods: Twenty-nine hemodialysis patients were evaluated for serum Al levels before and after dialysis. We tested the serum samples using inductively coupled plasma (ICP) method. We have checked blood chemistry tests that are routinely evaluated for hemodialysis patients. Paired samples t-test and correlation analysis were the statistical methods with 95% confidence interval.

Results: From 29 participants, 24 (82.8%) had toxic levels of serum Al before hemodialysis. Anyhow, 28 (96.6%) patients had toxic levels of serum Al after hemodialysis. Al serum levels before hemodialysis (Mean±SD= 25.61±15.61 µg/L; range= 2.8-69.7 µg/L) were increased after dialysis (Mean±SD= 30.1±15.19 µg/L; range= 9.1-79.3 µg/L), significantly (p= 0.033). Serum levels of Al before hemodialysis were positively correlated (R= 0.765; p= 0.000) and were increased significantly, after hemodialysis (p= 0.033). The level of Phos was reversely correlated with Al before dialysis (R= -0.547; p= 0.008). But, after dialysis Al was correlated reversely with neutrophil count (R= -0.468; p= 0.033) and NLR (R= -0.495; p= 0.023).

Conclusion: Al toxicity is clearly evident in hemodialysis patients in Khomein city. Increased level of Al after hemodialysis is related to the hemoconcentration event. In addition to present work, our previous studies in south and north parts of Iran confirm that hemodialysis patients should be monitored for Al toxicity. This toxicity seems to affect reversely on Phos and neutrophil counts. Treatment of such toxicity helps to enhance the life style of hemodialysis patients.

Keywords: Aluminum; hemodialysis; end-stage renal disease; phosphorous; neutrophil count

Arsenicosis Is a Frequent Disorder Among Hemodialysis Patients; A Situation That Needs Treatment

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Background and Aim: Toxic trace elements are increased in end-stage renal disease (ESRD) patients due to weak excretion. Although kidney eliminates the arsenic element, arsenicosis is an unknown circumstance in ESRD patients hospitalized in Iran and so Khomein city, in Markazi province.

Methods: Twenty-nine hemodialysis patients were evaluated for serum As levels before and after dialysis using inductively coupled plasma (ICP) method. Complete blood count (CBC) and ultrafiltration volume were tested before and after hemodialysis intervention. Statistical methods included: paired samples t-test. Kaplan-Meier survival test, and Pearson correlation.

Results: As serum levels before hemodialysis (Mean±SD= 7.58±1.99 µg/L; range= 1.56-10.6 µg/L) were increased after dialysis (Mean±SD= 8.62±1.83 µg/L; range= 5.68-13.58 µg/L), significantly (p= 0.025). The minimum and maximum As values before and after hemodialysis were equal to 1.56 µg/L and 10.6 µg/L, respectively (As > 1µg/L was considered toxic). Accordingly, all patients had arsenicosis. Before and after hemodialysis the mean±se of survival time was equal to 4.3±0.642 years (median= 3; 95% CI= 1.5 to 4.49 years). The As serum levels before dialysis were only correlated with WBC count (R= 0.482; p= 0.027) and neutrophil count (R= 0.443; p= 0.044). After dialysis the As serum level was associated significantly with ultrafiltration volume (R= 0.485; p= 0.016).

Conclusion: Although As is water soluble and excreted from nephrons, its concentrations are increased in ESRD patients. We suggest that arsenicosis is due to inability of nephrons to its excretion. Anyway, as the hemoconcentration after hemodialysis increases the As serum levels, we supposed that the toxic effects of As are enhanced, consequently. Therefore, As monitoring, or treatment using chelating agents are of choice. In addition to ultrafiltration, WBC and neutrophil counts were associated with As serum levels. Therefore, As roles in the inflammation, water balance and immunologic responses need to be elucidated.

Keywords: Arsenicosis; inflammation; end-stage renal disease; survival

Assessment of Nail Levels of Some Heavy Metals in Type 2 Diabetic Patients and Healthy Individuals

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Background and Aim: Trace elements as components of biological activity play important role in the metabolism of carbohydrates. In the present study, we aimed to investigate the nail levels of some heavy metals in Type 2 diabetes and healthy subjects.

Methods: In the present study, the subjects were selected among the newly diagnosed T2DM patients (n=124) admitted to the Amirkola Health and Ageing Project (AHAP) and healthy individuals (n=73). T2DM was confirmed by fasting blood glucose (FBG) ≥ 126 and hemoglobin A1C (HbA1c) $> 6.5\%$. The nails samples of T2DM patients were obtained from the newly diagnosed patients prior to any treatment. Nail levels of selected trace elements (Se, Cr, Cu, Cd, Pb, and Sr) were quantified using an atomic absorption spectrophotometer (AAS).

Results: The nail levels of Pb and Cd were significantly higher in the T2DM patients compared to the healthy ones. Besides, the Se, Cr, and Cu levels in the nail were reduced in T2DM patients relative to the normal subjects.

Conclusion: Se, Cr, Cu, Pb, and Cd play a critical role in the pathophysiology of T2DM. It seems that low nail levels of Se, Cr, and Cu and high levels of Pb and Cd are remarkably associated with T2DM. Probable causes should be assessed in other factors such as urine, blood, hair, and intervention factors in absorption and utilization and individual conditions.

Keywords: Diabetes; Trace element; Selenium; Cadmium; Copper; Chromium

Improving The Endothelial Dysfunction in Type 2 Diabetes with Chromium and Vitamin D3 Byreducing Homocysteine and Oxidative Stress

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Background and Aim: Atherosclerosis is one of the main causes of mortality in T2DM. Endothelial dysfunction has recently been under special investigation as the primary cause of biochemical processes which ultimately lead to atherosclerosis. Oxidative stress (OS) and hyperhomocysteinemia have prominent roles in the pathogenesis of endothelial dysfunction. Over the recent years, many studies have concentrated on pathogenic mechanisms and their therapeutic interventions. Chromium picolinate (CrPic) and vitamin D3 are known as two antioxidant micronutrients. In the present study, we examined the effects of the coadministration of chromium picolinate (CrPic) and vitamin D on serum Hct, Malondialdehyde (MDA) as an indicator of lipid peroxidation, total thiol groups (SHs) as innate antioxidants, total antioxidant capacity (TAC) as a marker of antioxidant status, and VCAM-1 and PAI-1 as biomarkers of endothelial dysfunction.

Methods: Subjects (n = 92) in this randomized, double blind, placebo-control study were randomly assigned to receive oral placebo (group I), D3 (group II: 50,000 IU/ week), chromium picolinate (CrPic) (group III: 500 µg/day), and both vitamin D3 and CrPic (group IV) for four months. Fasting blood samples were drawn at study baseline and following intervention to determine Hct, MDA, total antioxidant capacity (TAC), total thiol groups (SHs), vascular cell adhesion molecule- 1 (VCAM-1), and plasminogen activator inhibitor-1 (PAI-1).

Results: After intervention, MDA significantly decreased in groups II and IV; TAC significantly increased in group IV, and SHs significantly augmented in group III; Hct was significantly reduced in groups II, III, and IV; and VCAM-1 significantly decreased in groups III and IV and PAI-1 was significantly reduced in groups II, III, and IV.

Conclusion: Our findings suggest that through reducing homocysteine and oxidative stress and improving endothelial dysfunction, chromium and vitamin D3 co-supplementation might be predictive and preventive of cardiovascular diseases associated with T2DM.

Keywords: Vitamin D3; Chromium picolinate; Type 2 diabetes mellitus; Oxidative stress; Endothelial Dysfunction; Homocysteine

Iron Chelator or Iron Supplement Consumption in COVID-19? The Role of Iron with Severity Infection

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Background and Aim: Iron is a trace element that is used to replicate the virus and has a role in the vital functions of the body and the host's innate immune system. The mechanism of iron in COVID-19 severity is still not well understood. The aim of this study was to evaluate the association of iron with COVID-19 severity.

Methods: A case-control study was performed on 147 patients with a positive PCR test result and 39 normal individuals admitted to the Persian Gulf Martyrs Hospital in Bushehr, Iran. The iron profiles and related tests were measured along with hematological analytes.

Results: Hemoglobin (Hb), Fe, and saturated transferrin decreased in all the groups compared to the controls, but ferritin increased in the patient groups. After adjusting for age and sex, we found that increased ferritin levels augmented the odds ratio (OR) of the disease in the moderate (OR = 2.95, P = 0.007), severe (OR = 6.1, P < 0.001), and critical groups (OR = 8.34, P < 0.001). The decreased levels of Fe reduced the OR of the disease in the mild (OR = 0.96, P < 0.001), moderate (OR = 0.96, P < 0.001), severe (OR = 0.95, P < 0.001), and critical (OR = 0.98, P = 0.001) groups. Fe (AUC = 85.95, cutoff < 75.5 µg/dL, P < 0.001) and ferritin (AUC = 84.45, cutoff > 157.5 ng/dL, P < 0.001) have higher AUC for disease prognosis, but only ferritin (AUC = 74.89, cutoff > 261.5 ng/dL, P < 0.001) has higher AUC for disease severity assays.

Conclusion: It could be concluded that the use of iron chelators to reduce iron intake can be considered a therapeutic goal. In addition, measuring Fe and ferritin is beneficial for the diagnosis of the disease and determining its severity.

Keywords: COVID-19; Fe; ferritin; disease severity; Iron chelator

Trace Elements Against Covid-19

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Background and Aim: Covid 19 is a highly infectious disease that has infected a great number of people all around the globe. Although the majority of the population has been vaccinated, a look at trace elements and their effect on the prevention of this disease seems necessary. Therefore, this paper reviews recent studies exploring this matter. Literature review showed that trace elements such as copper, zinc, and selenium play an important role in strengthening the immune system to fight against the virus since all these trace elements were higher in survivors and healthy samples compared to non-survivors. It can be concluded that intake of supplements enriched in these trace elements may be useful to the immune system and somehow control the rise of pandemic in the near future.

Methods: review

Results: During the post-corona period, some studies have tried to find out the role of trace elements in lowering the chance of being affected by covid-19. For instance, the findings of a study that was conducted on covid patients in Iran indicated that the amount of zinc in those who suffered from covid 19 was more than healthy clients. The amount of Mn though seemed to be effective mostly for those older than 55 years old. Another research done in a village located in India concluded that the amount of selenium was fairly higher in healthy samples compared to those who were ill. Similarly, research done in Hubei province of China, demonstrated that selenium has a prominent effect on the rate of mortality, since Enshi, a city with soil enriched in selenium reported less death rate than Heilongjiang, a city with a population usually deficit in selenium. Furthermore, some studies have investigated the effect of copper on immunity against covid-19, for instance, a study exploring the effect of copper on immunity against covid-19 in German patients found that the amount of copper in the sample of the survivors was higher than non-survivors.

Conclusion: It can be concluded that a group of trace elements have a major effect on the immune system and therefore, have the ability to assist the human body against covid 19. Selenium, which is found as selenoproteins in the body, seems to play a huge role in immunity since it was fairly more in both survivors and healthy samples compared to non-survivors, it can be considered that the intake of selenium can lower the risk of becoming sick. Zinc was also considerably more in survivors and healthy samples compared to those who died; however, it can't be eventuated that additional intake of zinc has a positive effect on prevention of covid 19. Copper was also higher in survivors and healthy samples, and it was also shown that selenium and copper have a positive correlation, however, since none of the samples suffered from copper deficiency, it can't be fully demonstrated that intake of copper supplements can prevent covid 19 from happening.

Keywords: trace Elements; zinc; selenium; copper; covid19

A Systematic Review and Meta-Analysis of The Effects of Curcumin or Turmeric on Polycystic Ovary Syndrome

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Background and Aim: Polycystic ovary syndrome (PCOS) is the leading cause of anovulatory female infertility. We aimed to systematically review the available animal and human studies on the effects of curcumin or turmeric on PCOS and performed a meta-analysis of the randomized placebo-controlled clinical trials (RCTs). We hypothesized that curcumin/turmeric would improve reproductive and metabolic aberrations of PCOS.

Methods: A systematic search was performed in September 2020 in PubMed, Scopus, Embase, and Web of Knowledge databases, and Google Scholar. All original articles evaluating the effects of curcumin/turmeric on PCOS were eligible. The meta-analysis of RCTs was conducted using a random-effects approach. The effect sizes were expressed as mean difference (MD) and 95% confidence intervals (CI).

Results: Out of 73 articles, seven animal studies and four RCTs assessing the effects of curcumin on PCOS met our inclusion criteria. The reviewed studies indicated the beneficial effects of curcumin on the management of hormonal and metabolic disturbances of animals with PCOS. The meta-analysis demonstrated significant changes of fasting plasma glucose (MD=-3.33; 95% CI: -5.49 to -1.16), fasting insulin (MD=-1.60; 95% CI: -2.82 to -0.38; p=0.01), homeostatic model assessment for insulin resistance (HOMA-IR) (MD=-0.43; 95% CI: -0.73 to -0.12; p=0.01), quantitative insulin-sensitivity check index (QUICKI) (MD=0.01; 95% CI: 0.00 to 0.02; p=0.02), total cholesterol (MD=-11.32; 95% CI: -22.36 to -0.28; p=0.04), and body mass index (BMI) (MD=-0.20; 95% CI: -0.39 to -0.01; p=0.04) following curcumin supplementation.

Conclusion: In conclusion, animal studies underlined the favorable effects of curcumin on PCOS. Our meta-analysis also revealed significant improvement of glycemic control, cholesterol levels, and BMI after curcumin supplementation in overweight or obese patients with PCOS. The effects of bioavailability-improved formulations of curcumin on PCOS should be investigated in future studies.

Keywords: Polycystic ovary syndrome; Curcumin; Curcuma

Comparing The Effect of Cinnamon, Ginger, And Metformin on Glycemic Control, Lipid Profile, Anthropometric Indices and Sexual Hormones in Women with Poly Cystic Ovary Syndrome: A Randomized Single-Blinded Controlled Clinical Trial

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Background and Aim: Polycystic ovary syndrome (PCOS) is a prevalent endocrine disorder in women. Insulin resistance, hyperinsulinemia, and hyperglycemia are the main findings in women with PCOS. The objective was to determine the effects of cinnamon, ginger, and metformin on glycemic control and lipid profile in women with PCOS.

Methods: In this four-arm, double-blinded, randomized, placebo-controlled trial, 100 eligible participants were recruited. The participants were randomly assigned into one of the following four subgroups: Control Group: receiving rice flour capsule as a placebo, three times a day, Ginger Group: receiving capsules containing 500 mg of ginger, three times a day, Cinnamon Group: receiving capsules containing 500 mg of cinnamon, three times a day, Metformin Group: receiving 500 mg of metformin, three times a day.

Results: Consumption of Metformin and Cinnamon decreased insulin resistance (HOMA-IR) significantly in comparison to the placebo and ginger groups (p-value less than 0.05). It was indicated that consumption of Ginger decreased the FSH (follicle-stimulating hormone) and LH (Luteinizing hormone) level in comparison to the placebo group. While Metformin and Cinnamon decreased the Testosterone level (p-value 0.001, less than 0.001), none of the groups showed a significant change in relation to DHEA (Dehydroepiandrosterone).

Conclusion: In women with PCOS Ginger and Cinnamon can decrease body weight as well as Metformin. While Cinnamon can decrease Insulin resistance, Testosterone level and FAI (free androgen index) to the same extent of Metformin, Ginger only could decrease LH level. Over all Cinnamon and Ginger can be used as complementary treatment with Metformin in PCOS treatment.

Keywords: Polycystic ovary syndrome; metformin; cinnamomum zeylanicum; ginger.

Evaluation of Cortisol Level in Patients with Coronavirus and Comparison with Healthy Subjects

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Background and Aim: Coronavirus 2019 (COVID-19) is caused by a new virus, the acute respiratory syndrome of Coronavirus 2 (SARS-CoV-2), which first occurred in December 2019. There is a part of the human brain called the hypothalamus, which receives signals from all around, and in other words, the hypothalamus sends electrochemical messages to the pituitary gland by perceiving events in the body. The pituitary gland then begins to secrete various hormones, one of which is the adrenal cortex. When the adrenal gland is activated, corticosteroids, including cortisol, are released. High levels of cortisol indicate stress in the body, so the body will continue to be alert and coping and will try to meet the challenge of stress. Therefore, facing this severe challenge will cause a great loss of physical and mental energy, and if the body cannot cope, it will reach the stage of erosion. The beginning of the erosion process coincides with the manifestation of diseases such as high blood pressure, anxiety, mild and severe depression, aggression, fear, heart attack, and etc. With the perception of stress in the body (mental and physical stress), the hormone cortisol increases dramatically. It should be noted that cortisol is normally needed to regulate hormones and

Methods: In this study, serum cortisol levels were measured in 110 patients with coronavirus and compared with data from 50 healthy individuals who were matched with their age and sex. Data were analyzed using SPSS software and statistical tests.

Results: In this study, we measured cortisol levels in 110 patients with covid -19 disease identified by PCR, as well as cortisol levels in 50 healthy individuals. In this test, the mean age of the sick was 57 ± 2.13 and the female was 61 ± 4.5 , and the mean age of the healthy male was 49 ± 3.18 and the female was 53 ± 4.19 . It was done in the morning blood sample using Mono kit (ELISA Method). The normal range of cortisol according to the kit is 5-23 ug/dl. In the experiments, the level of cortisol in patients was 24.3 ± 5.6 and in healthy individuals was 16.8 ± 3.21 .

Conclusion: Studies have shown that high levels of cortisol are associated with an increased risk of death from COVID-19. Research has shown that patients with Covid 19 with very high levels of the stress hormone cortisol in their blood quickly get worse. Researchers suggest that cortisol levels can be used to identify patients who are likely to need intensive care. The findings of this study showed that serum levels of cortisol in patients with coronavirus were significantly increase compared to the control group.

Keywords: COVID-19; Cortisol; PCR



Other



Close Your Eyes, How Do We See the Invisible Bubble, & Burst It

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Background and Aim: This research can lead us to see other aspects of the covid-19 problem in society & resolve it. Let's go reduce the side effects of covid-19 in society while it pushes the people to us or even no before It. The rule of bubbles has also drawn to the consulting room. How dare they do! Is it because we advertised for products more than training? Or because we scared them more than we wanted sympathize with them? Or worse! Because we could not detect their invisible bubble! i Remember, I could not feel, they too! In fact, we could not, none of us! And it had nothing to do with the mask or oxygen! On the contrary, something was added to the space! i closed my eyes to the faces, and tried to track the visible clues in what was remain of in their body. And i listened carefully to the silence, what was hidden between the mask's rustle and the sound of breathing. And finally, i found it. There was an alive bubble there. i saw it, accepted without any condition, normalized it.

Methods: I use of ACT, EFT, Mind fullness, and we can continue with YOU via Correct Conscious Training with Empathy.

Results: If we can recognise the covid-19 side effects & normalise it with specific methods, we can reduce or eliminate its effects.

Conclusion: There is much elements that should change in our behave with both of covid-19's side effects and the society people, specifically in our training, which can lead us to the normal life same before in society with calm. so, close your eyes, and see the invisible bubble, & burst it!

Keywords: Covid-19; way to deal with COVID-19; Clinical consultation and consultation;covid-19 side effects;bubble

A Comparative Study of Exosome Isolation Using Ultracentrifugation and Commercial Reagents

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Background and Aim: Exosomes are extracellular endosomal nanoparticles, which appear under complex processes during formations of multivesicular bodies. Exosomes are potentially used as crucial agents for cell-free therapy, but their isolation and characterization can be challenging. In the current study, we have evaluated the characteristics and morphology of isolated exosomes and compared their efficiencies through two different methods.

Methods: We isolated mesenchymal stem cells (MSCs) from abdominal adipose tissue of 6 weeks age BALB/c mouse. The adipose tissue was minced and digested with collagenase type I (Gibco). MSCs were then cultured in suitable flask with DMED-F12 (Bioidea) content 15% fetal bovine serum (FBS; Gibco) and incubated under the standard condition at 37°C with 5% CO₂ for several days. In the third passage, the condition media was collected in order to exosome purification after 48 hours in FBS free medium. The condition media was transferred to ultracentrifugation tubes and centrifuged at 150 000 × g (150 minutes at 4 °C). Upon centrifugation, the supernatants were removed (leaving suspension 0.5 cm above the pellets) and the exosome pellets were suspended with PBS. On the other hand, isolation of exosomes with the commercial kit (Cibzist, Tehran, Iran) was performed according to the protocol of the supplier on the collected condition media. For both isolation methods, transmission electron microscopy (TEM), dynamic light scattering (DLS), and bicinchoninic acid (BCA) assay had performed.

Results: The electron microscopy and DLS indicated the presence of exosomes. Moreover, the kit and ultracentrifugation (UC) isolates contained approximately similar amounts of protein measured by the bicinchoninic acid (BCA) assay.

Conclusion: Overall, two isolation methods were applicable. Although ultracentrifugation is used as a gold standard to exosomes isolation, but commercial kit has some advantages and can apply alternatively according to the cost-effectiveness and time saving.

Keywords: Exosome; isolation method; ultracentrifugation; commercial kit

Adherence to Personal Protective Equipment for Prevention of COVID-19 And Other Diseases Among Healthcare Workers

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Background and Aim: Occupational exposure to blood and body secretions poses a significant risk of COVID-19, HIV, HCV, and HBV among healthcare workers (HCWs). Assessment of this exposure is necessary for optimized planning and policy-making measures. This study aimed to assess the exposure to occupational risk factors among emergency HCWs.

Methods: This cross-sectional study was performed on HCWs working in 3 educational hospitals affiliated with Arak University of Medical Sciences. The study sample included all HCWs working in emergency wards who met inclusion criteria. The data were collected using a valid and reliable researcher-made questionnaire and analyzed through analytical tests in SPSS software.

Results: The 116 studied HCWs included 97 nurses and 19 physicians and medical specialties. The mean age was 31.06 with 4.7 years of work experience (207.8 hours per month) in working at the patient bedside. The results indicated that needlestick injuries have a significant positive and negative relationship with job history ($p=0.001$) and month-averaged working hours ($p=0.012$), respectively. 96.6% stated that wearing gloves is not necessary, 59.9% stated that they do not use protective glasses due to a decrease in their vision, 50% did not use gowns due to the lack of gowns in the ward. 63.8%, 57.8%, 50%, 63.8%, 56% and 54.3% of the participants expressed the shift work, a high number of hospitalized patients in the crowded ward, need for high-speed working, high working load, increase in working hours, and low working consent as the most important factors affecting the increment in blood transmitted diseases, respectively.

Conclusion: Lack of motivation among HCWs, especially nurses, has worsened the adherence to protective measures. Leading systems should design and administer plans to decrease occupational exposures. On the other hand, hospital management should strive to provide high-safety equipment. Educational programs on infection control and prevention of occupational exposures are among other necessary measures. HCWs should be given the opportunity to check their antibody titers for HBV, HCV, HIV, and perform other necessary tests. Regarding the negative effects of exposure to blood, body, and respiratory secretions, it is necessary to design systems at the hospital and national levels for the surveillance of suspected contacts, their reporting, and the required supervision for health policy-making.



Keywords: Occupational Exposure; Blood Transmitted Diseases; Emergency Ward



Anti-Inflammatory And Antioxidant Effect of Moringa Peregrine Leaf Alcoholic Extract on Uni-Lateral Compression Model of Spinal Cord Injury in Rats

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Background and Aim: Inflammation and stress oxidative are two major factors that lead to the spreading secondary phase of spinal cord injury (SCI), so reducing these factors is an effective way to control the SCI. This study intended to evaluate the protective effect of alcoholic extract of Moringa peregrine leaf on the spinal cord against inflammation and oxidative stress damage induced by SCI.

Methods: thirty-five adult male Wistar rats were accidentally separated into five groups: control, control+ M. peregrine (150 mg/kg for fourteen days), sham(laminectomy)+saline, SCI, and SCI+ M. peregrine (150 mg/kg for fourteen days). After the injury, until fourteen days all animals were assessed by BBB (Basso, Beattie, and Bresnahan) for hind limb motor function. For evaluation of antioxidant enzymes (Superoxide dismutase (SOD), catalase (CAT), and Glutathione peroxidase (GPx)), oxidative stress markers malondialdehyde (MDA), and Total antioxidant), inflammatory factors (interleukin-1 β (IL-1 β), interleukin-6(IL-6), interleukin-18(IL-18) and tumor necrosis factor- α (TNF- α), and nuclear factor kappa B (NF- κ B) in spinal cord tissue all animal were sacrificed on the fourteenth day.

Results: Intraperitoneal injection of M. peregrine alcoholic extracts considerably decreased the expression of pro-inflammatory cytokines such as TNF-a, IL_1b, IL_6, IL_18, attenuated level of MDA, raised levels of SOD, GPx, CAT, and total antioxidant in spinal cord tissue; Furthermore, the level of NF- κ B in the SCI+ M. peregrine group was significantly lower than SCI group. Therefore, motor function was improved in the treatment group compared to the SCI group.

Conclusion: This conducted study shows that alcoholic extract of M. peregrine leaf has a neuroprotective effect against inflammation and oxidative stress induced by SCI. as a result, motor recovery in the treatment group is better than the untreated group.

Keywords: Moringa peregrine; spinal cord injury; inflammation; oxidative stress

Association Between the Organic Cation Transporter 3 Methylation and Hepatocellular Carcinoma

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Background and Aim: The most common cause of hepatocellular carcinoma (HCC) is hepatitis B infection and has a high death rate in the world. This study aimed to examine the expression of organic cation transporter 3 (OCT3) mRNA and gene methylation profile in the patients with HCC in comparison with healthy people.

Methods: Participants in this case- control study were patients with HCC (n=81) and healthy control group (n=90). Expression of OCT3 was studied using total RNA isolation and quantitative real-time PCR analysis (qRT-PCR) method. Methylation profile was evaluated by genomic DNA using methylation specific PCR (MSP) method.

Results: The mean OCT3 expression in patients with HCC was 0.58 ± 0.311 (Median=0.50, 0.20-1.30) and in healthy individuals was 1.20 ± 0.355 (Median=1.23, 0.60-2.10) (P0.05). The relative methylation as well as complete methylation (UM + MM) significantly increased the risk of HCC in comparison with unmethylation (OR=0.379, 95% CI=1.171-2.839).

Conclusion: The present findings revealed a tumor promoting function of the OCT3 in HCC. OCT3 methylation may be a pathological valuable factor in identifying the pathway of HCC.

Keywords: hepatocellular carcinoma; Organic cation transporters 3; methylation; quantitative real-time PCR

Asymptomatic Parasitic Infections and Their Challenges on The Supply of Safe Blood and Blood Components in Iran: A Review and Analysis of Available Data

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Background and Aim: Transmission of parasitic agents through transfusion can endanger the availability of safe blood and blood components to patients in need. The aim of this research was to describe the current status of transfusion-transmitted parasitic infections (TTPIs) and give appropriate strategies to minimize their transmission risk in Iran.

Methods: This review included all studies that estimated the prevalence of TTPIs based on parasitological, serological and molecular techniques in Iranian blood donors. The current research was carried out without time limitation using medical subject headings (MeSH) terms in the 10 electronic databases. The extracted data were recorded in a pre-prepared checklist, and analyzed using SPSS software.

Results: Twenty-seven papers were eligible for inclusion. In total, 11681 blood donors were examined for malaria, visceral leishmaniasis (VL) and toxoplasmosis in non-endemic and endemic areas. The overall serological prevalence of malaria, *Leishmania infantum* and *Toxoplasma gondii* infections was 10.53%, 1.96% and 33.95% among blood donors, respectively. The results of molecular techniques were positive in 3.33%, 39.22% (only seropositive samples) and 5.96% for malaria, VL and toxoplasmosis, respectively.

Conclusion: Regarding the detection of parasitic DNA and the presence of IgM antibody against parasites causing malaria, VL and toxoplasmosis among Iranian blood donors, the transmission of these infections through blood and blood components transfusion cannot be ruled out, particularly in endemic regions. Therefore, appropriate preventive strategies should be adopted to minimize the risk of TTPIs and ensure the availability of sufficient and safe blood and blood components for patients in need.

Keywords: Parasitic infection; Blood safety; Malaria; Leishmaniasis; Toxoplasmosis

Biopsychosocial Factors in Children Health in The Covid-19 Pandemic

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Background and Aim: The Covid-19 pandemic has affected various aspects of children's lives and children are not immune to it. Therefore, it can lead to various injuries in order to reduce their health. Therefore, the aim of this study was to review the bio-psychosocial factors in children's health in Covid-19 pandemic.

Methods: The present study is a focused review. Searching for information in SID, Science Direct, PubMed and Google Scholar databases was done using Persian and English keywords Covid-19, children, school and related factors through Boolean and MESH term strategies. The inclusion criteria for children are social factors and psychological factors and related articles were collected from 2019 to 2021. Finally, out of 25 articles searched, 12 articles related to the goal were reviewed.

Results: Factors will be divided into three categories: physical, psychological and social: the first category, physical factors including (irregular or unhealthy diets) due to school closures and family poverty; in fact, lunch at school is associated with improved academic performance And lack of adequate physical activity), the second category includes psychological factors (fear, fundamental changes in their lifestyle, experience of social isolation and high levels of parental stress) and the third category includes social factors (insufficient parental supervision and lack of basic necessities). Child, including safety, child abuse (including sexual abuse, physical abuse, and neglect), lack of daily routine and normalcy, school closures, resulting in learning gaps between children, hours of social restrictions on services Necessary (for example, grocery stores), lack of a suitable place to do homework, lack of Internet access, lack of heating facilities in poor families and housing instability).

Conclusion: The results of studies showed that the Covid-19 crisis affects a variety of physical, psychological and social aspects of children; Therefore, knowing and being aware of these factors can be effective for midwives, pediatricians and pediatric nurses to improve these factors and educate parents.

Keywords: Covid-19; Children; School; Related factors

Challenges of Covid-19 Vaccination in Iran

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Background and Aim: Immunization of a vast majority of people with safe and effective vaccines is one of the global strategies for overcoming coronavirus disease 2019 (COVID-19) pandemic. After a month-long severe acute respiratory syndrome-coronavirus 2 (SARS-CoV-2) genomic sequencing process, efforts to develop prophylactic COVID-19 vaccines have begun, and have led to the development of more than 80 vaccines with different platforms in world to reduce the rate of mortality and morbidity from SARS-CoV-2 and subsequently reduce health and socio-economical problems resulting from COVID-19 pandemic. So identification of the challenges in COVID-19 vaccination in Iran may help to faster achievement to extensive immunization.

Methods: In this study, which was conducted in may 2021, We searched PubMed, Google Scholar, Scopus and Irandoc databases for keywords including "COVID-19", "vaccination" And "challenges" in published articles without time restriction.

Results: After searching the databases and extracting a large number of articles based on the title and abstract, 29 articles were reviewed and finally 9 studies were included in this review.

Conclusion: In Iran, sanctions are still the major challenges in the field of purchasing and producing effective vaccines, but operative works had been taken and led to the development of various Iranian anti-COVID-19 vaccines and the beginning of the vaccination of medical staff and people at augmented risk for having severe forms of COVID-19. On the other hand performing social acceptance for vaccination, maintaining the security of vaccination sites, providing enough Vaccinators, Planning to implementation of medical interventions for people suffering from vaccine's side effects and planning for management of vaccination wastes are challenges of COVID-19 vaccination in Iran and Cooperation of relative authorities is essential to should make the necessary plans to overcome these challenges in the field of attaining to soon extensive immunization against SARS-CoV-2.

Keywords: COVID-19; Vaccination

Co-Infection of Sexually Transmitted Infection and Human Papilloma Virus in Cervical Sample: A Multicenter Study in Iran

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Background and Aim: Human papilloma virus infection is unlikely to be sufficient to cause cervical cancer, and the contribution of other sexually transmitted infections (STIs) could be the determining factor for cervical lesion-progression. STIs have an influence on women's health, related with cervicitis, urethritis, pelvic inflammation, complications of reproductive health, and poor pregnancy consequences. The aim of this study was to estimate the prevalence of *Gardnerella vaginalis*, *Chlamydia trachomatis*, *Trichomonas vaginalis* associated with HPV-positivity in cervical samples from patients who underwent annual routine gynecological exams from different geographical regions in Iran.

Methods: The 533 women between 18 and 63 years and living at 14 different cities in Iran. Samples of cervical swabs were collected, nucleic acid was extracted from vaginal swab suspensions (Add bio, Korea) and *G. vaginalis*, *Chlamydia trachomatis*, *Trichomonas vaginalis* were detected by multiplex PCR and HPV genotyping by hybridization method (Operon, Spain). The data were recorded using Microsoft Excel 2007 (Microsoft Corp, Redmond, WA, USA) and analyzed with the SPSS software (version 16; SPSS Inc., Chicago, IL, USA). P value < 0.05 was considered statistically significant.

Results: The mean age of the participants was 34.94 years (age range: 18-63years). A total of 202 (mean range of 33.6 years) patients were found positive for HPV with 36.63% (n=74) infected with high-risk type, 33.66% (n=68) infected with low-risk type and 29.70% (n=60) infected with multiple types. Among the 533 women, there were 14.25% (n=76) *G. vaginalis* infection, 3.0% (n=16) *Chlamydia trachomatis* infection, 0.75% (n=4) *Trichomonas vaginalis* infection. Among 202 women with HPV infection 18.81% (n=38) suffered from *G. vaginalis*, 4.45% (n=9) *Chlamydia trachomatis* and 1.48% (n=3) *Trichomonas vaginalis*.

Conclusion: HPV infection is associated with a number of factors, including age, pregnancy, and impaired immune function. Recent studies have shown that, in addition to the aforementioned factors, vaginal infection may be associated with increased odds for HPV infection. *Gardnerella vaginalis* infection and HPV infection may have consistency or synergies. Most of the women who present HPV with *G. vaginalis* infection were younger than 41 years old. In current study, *G. vaginalis* rate was 14.25%. In HPV high-risk type, HPV31 showed the highest prevalence and in HPV low risk-type, HPV6 showed the highest prevalence. STI maybe a cofactor and a risk factor of HPV cause cervical cancer and neoplasia, however this relationship is unclear and a large number of epidemiological and molecular studies are needed.



Keywords: Molecular characterization; co-infection; Human papillomaviruses; Gardnerella vaginalis; Trichomonas vaginalis; Chlamydia trachomatis



Comparative Study of The Effect of Music Therapy with Auricular Therapy on Postoperative Orthopedic Patients: A Randomized Controlled Clinical Trial Study

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Background and Aim: Postoperative management is one of the concerns of the medical system. The aim of this study was to compare the effect of music therapy and auricular therapy on patients' pain after orthopedic surgery.

Methods: In this clinical trial, 90 patients' orthopedic surgery patients admitted to the orthopedic ward of Ali Ibn Abitaleb Hospital in Rafsanjan in 1397 were included in the study. Individuals were drawn into three groups: music therapy, soft music with natural sounds of 25 decibels using a pair of headphones, and auricotherapy group using special earrings for 24 hours in both patient ears at the shenmen point and the control group. The intervention was performed 20 minutes and 24 hours after surgery and pain was measured before and after the intervention with a visual pain scale. Chi-square, Kruskal-Wallis and Wilcoxon tests were used to analyze the data

Results: Out of 90 subjects, 51 (56.7%) were male and 39 (43.3%) were female. The two groups were not different in terms of demographic variables. In the music-therapy group, the mean score of pain scores before the first intervention was 47.20, which twenty minutes after the intervention reached 38.60 (p less than 0.001). In the auricotherapy group, the mean score of pain scores before the first intervention was 42.95, which reached twenty minutes after the intervention 35.43 (p less than 0.001). But in the control group, there was no significant change in the mean rank of scores in the first phase of the intervention ($p = 0.059$). The aim of this study was to compare the effectiveness of music therapy and auricular therapy, Cohen's effect size was used. The results showed that in the music therapy group, Cohen d was 1.68, more than D Cohen in the auricular therapy group (1.15), which indicated a greater change in scores in the music therapy group. Also, in the second phase of the study, that 24 hours after surgery, Cohen D was 1.48 more in the music therapy group than in the auricular therapy group (0.51).

Conclusion: The use of music therapy can significantly reduce surgical pain. On the other hand, auricular therapy was also effective in reducing pain after orthopedic surgery. Implementing these protocols and allowing nursing staff to use them may further reduce pain and surgical anxiety and reduce analgesia.

Keywords: Music therapy; auricular therapy; pain; orthopedic surgery

Design and Construction a Novel Humanized Biparatopic Nanobody-Based Immunotoxin Against Epidermal Growth Factor Receptor (EGFR)

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Background and Aim: Targeted therapy has been developed for increasing the efficacy of cancer treatment. Epidermal growth factor receptor (EGFR) is one of the tumor cell surface markers that overexpressed in many epithelial cancers and makes it a good target for targeted therapies. The unique properties of nanobodies made them an effective tool for targeted therapy. However, the immunogenicity of nanobodies limited them to the treatment of human disease. Humanization is a capable strategy to overcome this problem.

Methods: In this study, a novel humanized anti-EGFR immunotoxin developed consisted of humanized biparatopic nanobody and PE24 (hu7CG2-PE24). The recombinant IT (RIT) was cloned in pET-22b vector and expressed in Escherichia coli BL21 Plys S, and then the protein was purified. The cell-based ELISA was used to determine the binding activity of RIT to the EGFR and cytotoxicity evaluated by using MTT assay.

Results: After producing the immunotoxin (hu7CG2-PE24) in bacterial host and purifying by affinity chromatography, the results showed that the hu7CG2-PE24 was attached to its target with high affinity and cytotoxicity occurred in EGFR-positive cell lines in a dose-dependent manner but not in the EGFR-negative cell line.

Conclusion: In the study, a novel humanized anti-EGFR immunotoxin (hu7CG2-PE24) designed and developed. The hu7CG2-PE24 specifically inhibited the proliferation of EGFR-overexpression cell lines.

Keywords: Immunotoxin; Nanobody; EGFR; Cancer; Targeted therapy

Effect of Total Light Deprivation on GABAergic Receptors Expression of Adult Rats Colliculus: An Immunohistochemistry Study

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Background and Aim: Deprivation of light is effective on neural plasticity and intervenes with other senses to compensate for this exclusion. Deprivation of light decreases the visual neurons and reduction of GABAA receptors in the nucleus of the visual and auditory responses can be strengthened. The aim of this study was to evaluate the effect of light deprivation on GABAergic receptors expression of adult rat's colliculus.

Methods: Twenty-four male and female adult Wistar rats that received two months of Total light deprivation (TLD) were used in this study. The animals were randomly divided in four groups. To study possible sexual differences in neurogenesis male and female rats were separated. TLD was started one week prenatally and continued for seven weeks. IHC study and Nissl staining were used respectively for studying GABA receptors expression and normal cell count of adult rats colliculus.

Results: Immunohistochemical results showed that the level of GABAB receptors in the Inferior Colliculus of experimental groups in Comparison with control groups had a significant increase (p value is less than 0/05). between experimental and control groups of males and females There was a significant difference (p value is less than 0/05). as well as the amount of GABAB receptors in the Superior Colliculus of the experimental groups Compared to control groups showed a significant decrease (p value is less than 0/05).

Conclusion: Our research showed that the expression of receptors and cellular changes in different parts of the nervous system are linked to TLD. These are is sex-related changes, as the rate of change is higher in females. Deprived of a sense over time, the other senses compensate for the function of that sense and bring the brain to its maximum processing capacity

Keywords: Light deprivation; GABA receptor; quadratic colliculus

Elevated Fecal and Serum Calprotectin in COVID-19 Are not Consistent with Gastrointestinal Symptoms

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Background and Aim: Intestinal epithelial cell damage caused by SARS-CoV-2 infection was thought to be associated with gastrointestinal symptoms and decreased fecal consistency. The association of the gastrointestinal symptoms with the COVID-19-mediated inflammatory response triggered by the gastrointestinal immune system was investigated in this paper.

Methods: Intestinal inflammation marker fecal calprotectin along with serum calprotectin and other inflammatory markers were measured in COVID-19 cases with and without GI manifestations as well as healthy individuals. Analyses were performed to compare COVID-19 patient subgroups and healthy controls and examine the relationship between fecal and serum calprotectin levels with gastrointestinal symptoms and disease severity.

Results: COVID-19 patients (n = 70) were found to have markedly elevated median levels of fecal (124.3 vs. 25.0 µg/g; P less than 0/0001) and serum calprotectin (3500 vs. 1060 ng/mL; P less than 0/0001) compared with uninfected controls. Fecal and serum calprotectin levels were not significantly different between COVID-19 patients who displayed GI symptoms and those who did not. Compared with other acute phase markers, both fecal and serum calprotectin were superior in identifying COVID-19 patients who progressed to severe illness.

Conclusion: Although the progression of COVID-19 disease is marked by an elevation of fecal and serum calprotectin, gastrointestinal symptoms or diarrhea were not correlated with calprotectin increase level.

Keywords: Biomarkers; COVID-19; Calprotectin; Diarrhea; Gastrointestinal Diseases; Inflammation

Epidemiological and Etiological Analysis of Burns in Qom Province in a 4-Year Period

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Background and Aim: Burn is one of the public health problems and a major cause of death as well as many physical and psychological disabilities in Iran. The aim of this study was to investigate the frequency and etiology of burns in Qom province from 2016 to 2020.

Methods: This is a descriptive analytical cross-sectional study. Data were obtained from the burn data record center of Qom province for 490 patients. A designed checklist was used for data collection. Data analysis was performed using SPSS 22.

Results: The mean age of patients was 17.01 years. Most burns (in 28.6% of the patients) were caused by oil and gasoline. 65.5% of the patients had 25% to 50% burning rates. The main remedial measure taken for 44.1% of the patients was debridement.

Conclusion: Since burn is more prevalent in childhood and adolescence, it is necessary to provide policy interventions and required training for the at-risk groups to prevent burn incidence.

Keywords: Burn etiology; Burn incidence; Epidemiological; Health problems; Grades of burn; Qom

Evaluating The Effect of Bariatric Surgery on Branched-Chain Amino Acids' Levels: A Systematic Review and Meta-Analysis

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Background and Aim: It has been proven that bariatric surgery is an effective and long-term weight loss procedure. Studies suggest that post-bariatric surgery patients have lower levels of branched chain amino acids (BCAAs), which may promote metabolic improvement. Based on a systematic review and meta-analysis, the current study examined the effect of bariatric surgery on BCAAs levels.

Methods: In this study searches were conducted in PubMed, Web of Science (ISI), Embase, and Scopus (Elsevier) databases to identify articles related to the subject that have been published as of July 2019. Throughout this review, all clinical trials were included if they evaluated the effects of bariatric surgery on the levels of BCAAs including leucine (mmol/L), isoleucine (mmol/L), and valine (mmol/L) for more than one week. The standardized mean difference (SMD) and 95% confidence intervals (CI) were applied for explanation of the effect sizes. We extracted the mean and standard deviation (SD) of the BCAAs levels pre- and post-surgery.

Results: There were nine studies with 11 effect sizes in the analysis. Our results revealed that bariatric surgery had a significant reduction effect on the valine (standardized mean difference [SMD]: -1.89, 95% confidence interval [CI]: -2.79, -0.99, I² = 90.9%), leucine (SMD: -0.96, 95% CI: -1.48, -0.44, I² = 72.4%), and isoleucine (SMD: -0.58, 95% CI: -0.84, -0.31, I² = 66.3%) levels after surgery in comparison with before the surgery. Moreover, none of the subgroups (age, continent, and duration) indicated a significant change in BCAAs.

Conclusion: The levels of valine, leucine, and isoleucine decreased significantly after bariatric surgery when compared with before. To make a more informed decision about the generalizability of our findings, more large-scale and homogeneous trials are required.

Keywords: Amino acids; branched-chain; bariatric surgery; meta-analysis.

Evaluation Information Sources of Students in Kermanshah Province, Iran about Covid-19 Pandemic

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Background and Aim: Covid-19 is a lung disease that inflames and narrows the airways. Covid-19 Pandemic causes recurring periods of wheezing, chest tightness, shortness of breath, and coughing. Covid-19 affects people of all ages. The relationships between life style and Covid-19 pandemic. This study was aimed to elucidate the source of information of the 4 student groups in Kermanshah's school including preschool, elementary school, middle school and high school about Covid-19 pandemic.

Methods: This cross-sectional prospective questionnaire survey was conducted in schools of Kermanshah province of Iran and Knowledge about Covid-19 pandemic was evaluated. 500 students in grades preschool, elementary school, middle school and high school were included. Data were statistically analyzed through one-way ANOVA, followed by Tukey's post hoc test.

Results: The majority of study population %67 social media as their main source of information and %15 obtained information from television; while only %3 achieved information from schools. An important relationship between age and source type of information were observed (P less than 0.05).

Conclusion: Our result indicated that adjustment of the red lines existed in strategist sights and the media; social media will reduce the risk of Covid-19 Pandemic with improving Life Style, revision and improvement of source of information should be a priority for all schools especially in preschools.

Keywords: Information; Covid-19; Pandemic; Students; Kermanshah

Evaluation of Anti-Inflammatory and Antioxidant Effect of Alcoholic Extract of Moringa Peregrina Leaves on The Hippocampus of Hypoxia Rat

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Background and Aim: Hypoxia is a condition that occurs in various diseases including ischemia, heart disease, bleeding and leaves irreversible effects. This study was performed to investigate the effects of alcoholic extract of Moringa Peregrina leaf, which is a rich source of various antioxidants, on the hippocampal area of the hypoxia model. In this study, 40 Wistar rats were used, which divided into four groups.

Methods: A, Control group (no intervention). B, Moringa extract group (Moringa extract was given to rats by gavage). C, Hypoxia group (rats were exposed to hypoxia with 8% oxygen concentration for 30 days and 4 hours per day). D, Treatment group (After each episode of hypoxia, rats were given a specific dose of the extract by gavage). Finally, the hippocampus of rats was removed and the RT-PCR technique for IL-18, IL-1 β , IL-6 and TNF- α gene expression evaluation and the ELISA technique for the evaluation of MDA, GPx, CAT and SOD activity were used. HIF-1 α gene expression was also measured to confirm the hypoxia model.

Results: According to the results, the expression of inflammatory genes in the hypoxia group increased significantly compared to the control group and in the treatment group, the expression of these genes decreased significantly in comparison to the hypoxia group. Also, the activity of enzymes in the hypoxia group was significantly reduced compared to the control group and in the treatment group, the activity of these enzymes was significantly increased in comparison to the hypoxia group.

Conclusion: Alcoholic extract of Moringa peregrina leaves due to its high content of various antioxidants, can have a high effect on reducing oxidative esterification and significantly reduce the side effects of hypoxia.

Keywords: Hypoxia; Moringa peregrine; Hippocampus

Evaluation of The Analytical Performance of ELISA for Testing of Antinuclear Antibodies

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Background and Aim: Enzyme-linked immunosorbent assay (ELISA) has emerged as an alternative method for detecting antinuclear antibodies (ANAs) due to its simplicity and cost-effectiveness. However, information regarding the specificity and sensitivity of ELISA is still controversial. In this study, we investigated the performance of ELISA as an alternative for the indirect fluorescent antibody (IFA).

Methods: Serum samples (n= 97) were obtained. Samples were screened for ANA using both ELISA and IFA methods according to manufacturer instructions. The index was calculated for each sample in the ELISA method by dividing sample optical density (OD) to OD of commercially provided cut-off solution. The results equal to or greater than 1 were considered positive. The cut-off value for the IFA test was set at 1:80 dilution. IFA tests were repeated at 1:160 and 1:360 dilution series for samples that were positive at 1:80 dilution. Evaluation of IFA slides were performed using EURO STAR III plus microscope

Results: Using the IFA method, the distribution of cases with positive ANA was 56.7% (n=55). When the ANA positive samples were retested at 1:80, 1:160, and 1:320 dilutions, it was revealed that respective counts were 47, 5, and 3. ELISA showed that 35% (n=34) were positive for ANA. No inconsistency between IFA and ELISA was seen with respect to the positive. All samples with positive ANA results with ELISA had also positive ANA tests with IFA. High index cases with ELISA coincided with higher titer IFA. Area Under Curve (AUC), sensitivity, and specificity for the ELISA method were 0.84 [0.76-0.920], 60%, and 100%, respectively.

Conclusion: Our result revealed that ELISA can be primarily used for detecting ANA due to its cost-effectiveness and simplicity. However, because of its possible false-negative rate, negative results using ELISA should be re-tested using IFA if there is clinical suspicion. For positive ANA results with ELISA, re-testing with IFA provides some advantages including titration and pattern determination. In conclusion, the IFA method as a gold standard should not be entirely replaced with ELISA. In the frontline, ELISA is suitable for ANA testing, but it is better to use IFA as a confirmatory method. Future studies with a clinical perspective are necessary to confirm our results.

Keywords: Indirect fluorescent antibody; Enzyme-linked immunosorbent assay; antinuclear antibodies

EXPRESSION PATTERN OF p53 AND Ki-67 IN HBV-RELATED HEPATOCELLULAR CARCINOMA: A QUANTITATIVE REAL-TIME PCR AND IMMUNOHISTOCHEMICAL STUDY

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Background and Aim: The HBV-related hepatocellular carcinoma (HCC) is an important liver malignancy worldwide and carries a poor prognosis. In this regard an accurate diagnosis is necessary to enable successful treatment. The aim of the current study was to assess the relationship between the expression of certain molecular markers and HCC diagnosis in Iran.

Methods: Immunohistochemistry and quantitative RT-PCR techniques were used to evaluate the expression patterns of p53 and Ki-67 in liver tissues from 121 HCC and/or HBV-infected patients and 30 healthy volunteers.

Results: Patients with HBV+HCC demonstrated increased expression of both p53 and Ki-67 compared to patients with HBV only, highlighting correlation between the p53 and Ki-67 expression levels and HCC diagnosis. The prognostic value of p53 for the diagnosis of HCC was more reliable. The p53 demonstrated higher sensitivity compared to the Ki-67 (sensitivity and specificity, 77.3 and 76.4% for the p53, and 51.0 and 97.9% for the Ki-67, respectively). A panel containing two positive markers had higher specificity and comparable sensitivity to a panel with one positive marker regardless of which one (sensitivity and specificity, 94.8% and 97.9%, for two positive markers and 96.5% and 86.4% for one positive marker, respectively).

Conclusion: Taken together the combined analysis of p53 and Ki-67 expression provides a mean to increase the specificity and sensitivity of HBV-related HCC diagnosis to an acceptable level.

Keywords: p53, Ki-67; hepatocellular carcinoma; immunohistochemistry; quantitative real-time PCR

General Crises and How to Diagnose Them in The Corona Pandemic

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Background and Aim: Coronaviruses are a new type of virus that causes symptoms similar to the common cold to severe (respiratory) pneumonia and has not been seen in humans before. After the epidemic and the conflict, almost all countries are trying to plan to prevent and control the disease. The aim is to investigate the crisis of infectious diseases and ways to diagnose it in coronary heart disease

Methods: In this review study, articles published until December 6, 2021 were reviewed in scientific databases. PubMed, Scopus, Web of science, SID, Google scholar, Science direct. , A number of articles were selected. After reviewing the full text of the articles, a limited number of articles were reviewed

Results: Monitoring, early detection, and rapid response to the spread of emerging and re-emerging infectious diseases require policy-making, planning, training, support, and responsible action by countries and health systems. This, in turn, owes much to having a resilient, flexible, intelligent and agile health system to deal with any public health event. In Iran, due to the shortage and high cost of diagnostic kits, it is not possible to screen through public testing, and in order to screen suspects, follow-up is done by telephone from the head and family members, especially high-risk families.

Conclusion: Although the epidemic of Covid-19 disease is worrying and it is very important to formulate appropriate policies and implement them properly, Iran and other countries in the world are making great efforts in this direction. The threat of Covid 19 disease in Iran has provided the opportunity to create an integrated referral system, which now more than ever needs more coherent and practical measures to control the epidemic.

Keywords: Keywords: crisis; Covid 19; pandemic; infectious diseases

Identification of Remdesivir Analogues as Potential SARS-CoV-2 RNA-Dependent RNA Polymerase Inhibitors Based on In-Silico Studies

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Background and Aim: RNA-dependent RNA polymerase (RdRp) is an essential component of SARS-CoV-2, having a crucial function in the infectivity of this virus by catalyzing the replication of RNA. Due to this key role, RdRp is considered as a target of anti-SARS-CoV-2 therapeutics. Remdesivir (RDV) with the inhibitory effect against this enzyme is recently used in the treatment of COVID-19 infection. The present study aimed to detect further structures with the potential to inhibit SARS-CoV-2 RdRp using in silico methods.

Methods: Different chemical databases, including ZINC, Cactus, and Chemical Entities of Biological Interest (ChEBI), were explored to prepare a library of RDV analogues. Using virtual screening methods, more than 200 compounds were extracted and then optimized to the triphosphate form. Docking experiments were performed on the dataset of RDV analogues using PyRx-Vina software by applying the optimized SARS-CoV-2 RdRp (PDB ID: 7BV2) model as the target. The compounds with the most favorable binding energy (≤ -7.3 kcal/mol for the active form of remdesivir) were selected for subsequent analyses after evaluating ligand-protein interaction. To determine the potential pharmacokinetic properties of the top compounds, including drug toxicity and drug-likeness, a preliminary computational screening was also performed.

Results: In the present study, five RDV analogues indicated the lower binding energies to RdRp among the search results are reported. In addition, the triphosphate forms of the mentioned compounds demonstrated the lowest binding energies to RdRp (compared to their main structures and also the active form of RDV; GS-441524-TP). The compound (2R,3R,5R)-2-(4-amino-7H-imidazo[4,5-d][1,2,3] triazin-7-yl)-tetrahydro-5-(hydroxymethyl) furan-3-ol) with the lowest binding energy is the most suitable candidate to inhibit SARS-CoV-2 RdRp.

Conclusion: Our results represented new analogues of RDV as potential inhibitors of RdRp due to the best protein-ligand interactions. These compounds can be used in the drug improvement process against COVID-19 after performing further studies.

Keywords: COVID-19; RdRp; Drug discovery; computational chemistry; Remdesivir

Investigation on Percentage and Kind of Alcohols Used in Sanitizer Samples Sent to Drug and Food Lab of West Azarbayjan Province in 1398-1399

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Background and Aim: Introduction: Regarding epidemic covid-19 consumption of different shapes of sanitizers like sprays or gels has been increased a mony people. Hand sanitizers (with appropriate concentration 70% alcohol) are used as infections agent disinfection like bacteria and viruses. Unfortunately, because of industrial alcohol (methanol) low price, some profiteer producers use methanol instead of ethanol in sanitizer production. Contrary to ethanol which doesn't absorb by uninjured skin, methanol absorption through specially injured skins is very high and could cause severe poisoning and sight problems. Consequently, detection of alcohol type, especially presence of methanol in sanitizers is very necessary during pandemic conditions.

Methods: Measurement of alcohol was done using Gas Chromatography (GC) – Agilent 6890N model equipped to headspace sampler 7697A model and FID detector and using DB-WAX column (30m×32mm×0.5µm). In order to determination curve was drawn for mixture of some alcohols including methanol, ethanol, 1-propanol and 2-propanol. Then samples were diluted by deionized water and injected to Gas Chromatography.

Results: 219 samples were analyzed in order to determine percentage and kind of alcohol in drag and food lab during 1398-1399. Results showed that 7.3 percent of sanitizers were including methanol against from Ministry of health instructions, and 2.74 percent of sanitizers were including mixture of ethanol and 2-propanol, 89.95 percent of samples were including ethanol according to Ministry of Health instructions.

Conclusion: Duo to the amount of disinfectants production in the country and the continuous use of these products by all age groups, the need for a national monitoring by the dependent organs is completely inevitable to identify the grounds for fraud and deal with it effectively and try to produce and supply safe and healthy products.

Keywords: Alcohol; Gas Chromatography; Headspace and west Azarbayjan province

Maintenance Hemodialysis Diminishes Magnesium and Manganese Serum Levels; This Necessitates Trace Elements Supplementation in End-Stage Renal Disease Patients

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Background and Aim: Magnesium (Mg) and Manganese (Mn) are essential elements plying a critical role in growth and regeneration of tissues in human body

Methods: Twenty-eight and twenty-six hemodialysis patients were evaluated for serum Mg and Mn, respectively before and after hemodialysis (BAHD) using inductively coupled plasma (ICP) method. Mann-Whitney Test was used to compare serum Mg and Mn ranks between before and after hemodialysis with at least 95% confidence interval.

Results: Mean $\pm$ SD of serum Mg before hemodialysis was equal to 17.30 $\pm$ 3.45 μ g/L that was decreased after this intervention in to 15.51 $\pm$ 3.14 μ g/L ($p= 0.022$; BAHD correlation=0.296 and insignificant). Mn serum levels were equal to Mean $\pm$ SD= 18.02 $\pm$ 9.09 μ g/L before hemodialysis that were diminished significantly into 16.12 $\pm$ 8.32 μ g/L after it ($p= 0.039$; BADH correlation= 0.873 and meaningful).

Conclusion: Many cellular and molecular process are dependent to the metalloenzymes. Thus, trace metals insufficiency could be associated with organ malfunction and pathologic consequences. We have seen that Mn and Mg serum levels are decreased after hemodialysis. Mn and Mg are essential cofactors for metalloenzymes. Considering hemoconcentration after hemodialysis and decreased levels of studies trace elements after dialysis sessions, we think that supplementary regimens are necessary for patients under maintenance hemodialysis.

Keywords: magnesium; manganese; supplementation; metalloenzymes; maintenance hemodialysis

mRNA Expression and Epigenetic-Based Role of Chromodomain Helicase DNA-Binding 5 in Hepatocellular Carcinoma

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Background and Aim: Chromodomain helicase DNA-binding 5 (CHD5 gene), as a tumor suppressor gene, may be change the individual susceptibility to hepatocellular carcinoma (HCC). This study aimed to evaluate the methylation pattern of the promoter region of CHD5 gene and CHD5 mRNA expression in the HCC patients compared to healthy individuals.

Methods: In this case-control study, the expression of CHD5 gene and DNA methylation pattern were analyzed in 81 HCC patients and 90 healthy individuals by Quantitative real-time PCR and methylation specific PCR.

Results: distribution of methylation patterns was different in the two groups and there was a statistically significant hypermethylation of CDH5 gene in 61.8% of the HCC patients and corresponding 54.4% of the controls (OR=0.418, 95% CI=1.219-2.799, P=0.008). The mRNA expression level was statistically lower in HCC pateints compare to the controls.

Conclusion: hypermethylation of the promoter region of the CHD5 gene may change the expression of this gene and the incidence and severity of HCC. Methylation of CHD5 can also study as a prognostic factor in HCC.

Keywords: chromodomain helicase DNA-binding 5; hepatocellular carcinoma; methylation; quantitative real-time PCR.

Newly Identified COVID-19 Drug Candidates Based on Computational Strategies

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Background and Aim: As of November 2021, the 2019-new coronavirus disease (COVID-19) has brought more than 260 million cases worldwide, with nearly 6 million deaths. Although several vaccines have been approved and repressed the pandemic, very few drugs have shown efficient inhibitory effects to attack the virus. The current study aimed to identify novel drug candidates based on quinoline structure with the help of computational strategies, including virtual screening and molecular docking approaches.

Methods: A library of 548 substances was collected based on similarity search from chemical and biological databases, including Chemical Entities of Biological Interest (ChEBI), PubChem, ZINC, Sigma-Aldrich, Enhanced NCI Database, and the synthetic compounds of our laboratory. Molecular docking was performed subsequent preparation of proteins encoded by new coronavirus genes. The interactions of all compounds with three SARS-CoV-2 proteins, namely main protease (Mpro), spike receptor-binding domain in a complex with ACE2 (Spro-ACE2), and chimeric receptor-binding domain in a complex with human angiotensin-converting enzyme 2 (RBD/ACE2) were determined to find new candidate inhibitors and clarify their mechanism of action. In silico drug-likeness properties and ADME analysis were also evaluated.

Results: By exporting the docking calculation results in detail, molecules with the minimum binding energy of the formed interactions with the target proteins were considered as the best inhibitors. We reported the top five hits from the output of the virtual screening process for each protein. The compound 1-(6-chloro-2-methoxyacridin-9-yl)-2-(5-nitrofuranyl)methylenehydrazine showed suitable binding to SARS-CoV-2 main protease and spike protein sites. More remarkably, N-4-(6-methyl-3-pyridinyl) phenyl)-9-acridinamine represented high affinity to all targeted proteins.

Conclusion: Our results indicate that the identified compounds can inhibit the SARS-Cov-2 entry into the host cell. Compounds with multiple mechanisms of action can reinforce drug efficacy and could imply powerfully promising drug candidates against COVID-19 following further examination.

Keywords: COVID-19; Computational Biology; Virtual screening; Molecular docking; Quinoline

Paper Title “Hu7CG2: A Novel Humanized Anti-Epidermal Growth Factor Receptor (EGFR) Biparatopic Nanobody”

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Background and Aim: Targeted therapy with antibody is an effective approach with low side effects in cancer therapy compared with other treatment approaches. Epidermal growth factor receptor, EGFR, is a favorable biomarker as targeted therapy because it overexpresses in several cancers.

Methods: In this study, we designed, constructed, and evaluated a novel humanized anti- EGFR biparatopic nanobody, hu7CG2. The hu7CG2 was designed by grafting the complementarity-determining regions of two camelid anti- EGFR nanobodies known as 7C12 and EG2 to a universal scaffold and then connected with a glycine-serine linker.

Results: The hu7CG2 produced in bacterial host and purified by affinity chromatography. The results of antigen-binding activity and cell viability assays showed that the hu7CG2 inhibited the growth of EGFR overexpression tumor cells. The data showed that hu7CG2 might be a useful tool in the targeting and treatment of tumor cells.

Conclusion: In vitro analysis showed that hu7CG2 specifically inhibited the growth of EGFR overexpression tumor cells with low immunogenicity. The results showed that hu7CG2 is a suitable candidate for in vivo investigations for targeting EGFR overexpression tumors.

Keywords: Hu7CG2; EGFR; Biparatope; Humanized Nanobody; Targeted therapy

Prevalence of Laboratory Test Quality Control Indicator Deviations in Hamedan Province

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Background and Aim: Internal laboratory test quality control is performed to ensure the stability of the test system during laboratory runs. Establishment of quality control principles guarantees laboratory performance improvement and patients' confidence in the results of their tests. This study was conducted to determine laboratory test quality control indicator deviations in Hamedan province

Methods: In this cross-sectional and retrospective study, data of 184 laboratories (117 private and 67 public) was entered to SPSS version: 16 software, based on the completed quality system of medical laboratory evaluating check list, available in the reference laboratory of Hamadan province from 2016 to 2020. Data were analyzed using nonparametric equation of Kruskal-Wallis test by comparing the significance level of P-value

Results: The highest percentage of deviation belonged to the acceptable total error determination with an average of %75.88 (P-value = 0.002). Mean of laboratory test quality control deviations were %21.9 and %15.68 in serology and microbiology respectively which were not significantly different from each other, but there was a significant difference in comparison of each of them with all laboratory wards in Hamedan province (P-value = 0.022 and 0.038).

Conclusion: The assessment of different laboratory test quality control indicator deviations during 4 years showed that private laboratories had better performance than public laboratories in term of laboratory test quality control establishment.

Keywords: Medical laboratory; Quality control; Acceptable total error; Deviation

Right Atrial Clot and Pulmonary Embolism in A Patient with COVID-19: A Case Report

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Background and Aim: The infection caused by the novel coronavirus (COVID-19) immersed the globe into a widespread pandemic. The disease leads to acute respiratory disease syndrome, hyper-coagulation, and cardio-vascular diseases.

Results: In this case report, we presented an 80-year-old man with right atrial clot and acute pulmonary embolism, who was diagnosed with COVID- 19. The patient was isolated and transferred to the intensive care unit with a diagnosis of submassive pulmonary thromboembolism and right atrial clot following COVID-19 infection. Antibiotics and anticoagulants were administered, and the patient was referred for mechanical thrombectomy. He did not die and after recovery, was discharged with warfarin administration.

Conclusion: Preventing thromboembolic events seems to be the first priority in the management of COVID-19 patients. It is necessary to look for strategies to manage and prevent the early occurrence of thromboembolic events in these patients.

Keywords: COVID-19 Embolism Atrial clot ARDS Coronavirus

Survey on Equipment-Related Non-Conformities in Public and Private Medical Laboratories of Hamadan Province

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Background and Aim: Different equipment were used in clinical laboratories for test performance and diagnosis of diseases. Equipment-related nonconformities influence laboratory test results. These non-conformities were defined as does not meet the predetermined requirements. Hence, supervision of equipment non-conformities has critical role for laboratory improvement by reporting and monitoring of them continuously. The aim of this study was to determine equipment-related non-conformities in public and private medical laboratories of Hamadan province.

Methods: In this cross- sectional retrospective study, data of 184 medical laboratories, 117 private and 67 public were entered to SPSS version 16 software. All data was extracted from the 4-year-completed medical laboratory quality system assessment check lists. The information was presented using mean standard deviation and nonparametric equation of Kruskal-Wallis test and significance level by P value

Results: By evaluation of the defined equipment indicators in the mentioned checklist, the highest equipment-related non-conformities with an average of 68% (P Value=0.025) belonged to the installation quality (IQ) indicator and the lowest non-conformities belonged to the log book with an average of 25.5%. Public laboratories with an average of 9.15% (P Value=0.030) and private laboratories with an average of 16.12% (P Value=0.001) had significant difference compared to all laboratories, but there was no significant difference between public and private laboratory nonconformities.

Conclusion: The average percentage of reported equipment-related non-conformities in the laboratories of Hamadan province has decreased during 4 years period of time. Continuous performed audits and more supervision and could be helpful to reduce non-conformity rate.

Keywords: Medical Laboratory; Non-conformity; Equipment; Installation quality.

The Effects of Platelet-Rich Plasma on Cell Proliferation, Adhesion and Spreading of Human Adipose-Derived Stem Cells and Preserves Their Stemness

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Background and Aim: Human adipose-derived stem cells (hADSCs) are a promising autologous cell source in tissue engineering. However, the use of hADSCs requires ex vivo expansion, to obtain sufficient cell numbers and improvement of cellular behaviors such as cell adhesion and spreading. Nevertheless, xenogeneic supplements (FBS) face risks such as the risk of infection and immunological reactions. Platelet-rich plasma (PRP) as an autologous source, contains several growth factors that can stimulate cell proliferation and cellular behaviors. Therefore, in this study, the effect of PRP in enhancing cell proliferation, spreading, adhesion, and maintaining stemness in hADSCs was investigated.

Methods: hADSCs were isolated and characterized from human adipose tissue. PRP was collected from human peripheral blood and activated with autologous calcium chloride. hADSCs(P3) were cultured in DMEM supplemented with two concentrations of activated PRP (5% and 10%), against FBS supplemented media. Cell proliferation rate was analyzed using the MTT assay. The number of attached cells (flat and rounded) per surface area and the percentage of spread cells (flat and spread) (the number of spreading cells/the number of attached cells $\times$ 100) were counted randomly in multiple areas using a phase-contrast microscope. The gene expression of stemness marker (Nanog and Klf4) was analyzed using qRT-PCR.

Results: The results showed a significant increase in proliferation, adhesion, and spreading of hADSCs cultured in PRP compared to FBS. The changes proliferation over different concentrations and times in PRP can be seen in more detail in Figure B. Moreover, PRP-expanded cells compared to FBS-expanded cells showed statistically significant up-regulation of stemness genes expression.

Conclusion: Our data demonstrated that PRP as an alternative to FBS enhanced cell proliferation, adhesion, spreading, and maintained stemness of the hADSCs during ex-vivo expansion.

Keywords: Adipose-derived stem cells; platelet-rich plasma; cell proliferation; regenerative medicine

The Prominent Chest CT Findings in COVID-19 Patients: A Systematic Review and Meta-Analysis

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Background and Aim: Assessing the quality and manner of pulmonary involvement through imaging modalities seems to be useful in predicting the severity of Covid-19 and thus determining the best disease management approach. The present study summarized systematically the chest CT findings of patients suspected to Covid-19

Methods: Data abstraction was independently performed by two un-blinded reviewers on structure collection forms without divergences in data collection. The manuscript databases including Medline, Web of knowledge, Google scholar, Scopus, and Cochrane were deeply searched for all eligible studies based on the considered keywords. The risk of bias for each study was assessed using the criteria outlined in the Cochrane Handbook for Systematic Reviews of Interventions and also according to QUADAS-2 tool

Results: Overall, 22 studies (including 4052 patients, overall mean age of 48.19 years ranged 42 to 57 years, male/female of 2113/1939) were reviewed. The pooled prevalence rate of CT positivity was 89.5% (95%CI: 95.4% to 92.6%). The common CT findings in Covid-19 patients include GOO in 78.2%, interlobular septal thickening in 48.5%, air bronchogram pattern in 44.5%, consolidation in 37.4%, crazy paving in 21.9%, nodules in 12.1%, reticular pattern in 9.8%, and pleural effusion in 7.3%. Other patterns including bronchiectasis, cavitation, lymphadenopathy, or pericardial effusion were rarely seen

Conclusion: Pulmonary involvement can be revealed by chest CT scanning in about 90% of patients and the most prominent CT findings include GOO, interlobular septal thickening, air bronchogram pattern, consolidation, and crazy paving

Keywords: Chest CT findings; COVID-19; Patients