In the name of God
the Merciful, the Compassionate

20th National and 8th International Congress of Biology,
22-24 August, University of Maragheh, Iran

The congress was held as four distinct conferences included:
Applied, Animal, Cellular and Molecular, and Plant Biology Conferences

Proceeding of
Applied Biology Conference
Welcome Message of Congress President

In the name of God

The history of biology goes back to long time ago, about 3.8 billion years, when the first living creatures began to exist on the earth as amoebas. Although biology was confirmed as an independent discipline in the 19th century, it actually originated in ancient medical science in Mesopotamia, China, India, and Egypt. Nevertheless, modern biology and its tendency to study the nature go back to ancient Greece. During the Renaissance and early modern era, biological thoughts underwent major changes due to development of an inclination toward empiricism and discovering many types of new living creatures.

It is a pleasure for us to welcome eminent scholars and researchers to the 20th National and 8th International Biology Congress in Iran, Maragheh. Maragheh encompasses the most comprehensive of all scientific, cultural and artistic treasures. Maragheh reminds us the memory of a "university as wide as a city" culture because it first presented it to the history and world of science. The development of the City-University of Maragheh, in its historical memory, commemorates the first girls' school (the dynasty of the Ilkhani), is undoubtedly based on the revival and stabilization of scientific, cultural and artistic School of Maragheh.

University of Maragheh is proud to hold this glorious scientific congress by the presence of honorable professors, researchers, students, teachers, and other guests interested in biology. Wishing you all a heartfelt welcome.

Mohammad-Ali Lotfullahi Yaghin

The Massage of Congress Chairman

In the name of Allah

We are very glad that the Iran’s 20th national and 8th international Congress of biology 2018 took place in the beautiful city of Maraghe with the estimable endeavor and cooperation of chancellor and faculty members of the University of Maragheh and the managing council of Iranian society of biology. This gathering was an opportunity for all Iranian biologists inside and outside the borders of Iran to obtain the basis of more progresses in this zone of mankind knowledge, with interchanging their information and achievements together and with scientists from different countries of the world like Italy, Canada, Turkey, Armenia and...

The society of biology is honored for obtaining the conditions for this affair, and is thankful to Maraghe university, the academic staff at department of biology and other collaborators of that university for their heartily host.

The endeavor of the scientific and administrative committees in this congress in creating scientific and heartily atmosphere during the period of celebration is definitely praiseworthy.

I deem it necessary for myself to heartily appreciate the endeavor of the respectful directorship, vice chancellors and specially the dear students of Maraghe university. And to wish grace and success for all my valuable colleagues in Iranian society of biology’s executive committee who endeavored in both administration and diplomacies of this congress.

And hope to see all of the participants from all over the world in the next congress.

Mohammad Nabiuni
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Congress Plenary Invited Lectures

**Prof. Khosrow Adeli** (University of Toronto, Canada)
RNA regulatory network in lipid metabolism: critical roles of micro RNAs and RNA granuls

**Prof. Sinerik N. Ayrapetian** (Head of UNESCO Chair in Life Science, Armenia)
The quantum-mechanical nature of cell signaling system

**Prof. Mohammad Ghannadi-Maragheh** (Institute of Nuclear Science and Tecnology, I.R. Iran)
Application of nuclear science and tecnology in biology and medicine

**Prof. Ali Akbar Moosavi Movahedi** (University of Tehran, I.R. Iran)
Biomimetics and lifestyle

**Prof. Luciano Sasso** (Sapienza University of Rome, Italy)
Pharmacological applications of modulators of oxidative stress
Lectures
Applied Biology Conference Invited Lectures

Prof. Mohamad-Bagher Rezaei (Research Institute of Forests and Rangelands, I.R. Iran) Natural environment saved by herbal of chemical composition (pest controlling)

Prof. Behzad Ghare Yazie (Agricultural Biotechnology Research Institute of Iran, I.R. Iran) Beyond genetic engineering for food security

Assoc. Prof. Saber Khodabandeh (Tarbiat Modares University, I.R. Iran) Marine bioactive compounds: How Persian Gulf puffer fish TTX control breast tumor growth?

Dr. Sadegh Rostamnia (University of Maragheh, I.R. Iran) A new trends on design and preparation of nanoporous & Fe₃O₃ based hybrid supports for covalently immobilization of enzymes: Dithiocarbamate instead of glutaraldehyde

Assoc. Prof. Mohammad Amin Hejazi (Agricultural Biotechnology Research Institute of Iran, I.R. Iran) Molecular mechanisms behind CO₂ adaptation in microalgae

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Designing and construction of appropriate genetic elements for production of glyphosate tolerant transgenic rice

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Rice (Oryza sativa L.) is one of the most important food products and the primary source of food for more than a third of the global population. Weed management is an important factor in production of rice in order to achieve high yield and high quality crop. Glyphosate (2-phosphonomethyl-glycine) is a potent non-selective herbicide with a wide range and is widely used in farms. This herbicide inhibits EPSPS, a key enzyme in the shikimate pathway. Development of herbicide-tolerant transgenic plants is the ultimate strategy to reduce the herbicide effect. In this study, the target gene (epsps) that isolated from the Pseudomonas bacteria after screening of soil, it has been shown that this epsps gene has produced an acceptable resistance in bacteria. We needed ideal genetic construct to transfer this gene to plants, so we decided to design this structure. First the epsps gene of the pJET28 vector was transferred using the opportune restriction enzymes into the pBI221 vector. This structure required an appropriate promoter for gene expression into monocotyledonous plants such as rice. We needed ideal genetic construct to transfer this gene to plants, so we decided to design this structure. First the epsps gene of the pJET28 vector was transferred using the opportune restriction enzymes into the pBI221 vector. This structure required an appropriate promoter for gene expression into monocotyledonous plants such as rice. We selected the pAct1-D promoter, based on the studies, carried out by the restriction enzymes from the pAct1 carrier to the target vector at the upstream end of the epsps gene. The Intended vector was prepared and transferred into Ecoli.Dh5α bacterial. First the plasmids were extracted from the bacterium, to confirm the cloning of the gene and the promoter in the vector, a polymerase chain reaction was initiated with primers designed for the epsps gene as well as the promoter pAct1-D. Secondly, the presence of a cloned gene and promoter using appropriate restriction enzymes and double digestion enzyme was confirmed. These results of this study can be use in transferring useful and commercially genes to recalcitrant plants.

Keywords: Rice, Glyphosate Tolerant, Promoter, Transformation

The study of OeACP2 polymorphism in some Iranian native olive cultivars

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Olive (Olea europaea) genus from Oleaceae family has about 20 shrub species which have been spread around the world. Olive oil is valuable not only for its natural resource but also for unsaturated fatty acids content, particularly oleic acid, which made it more stable than other vegetative oils. Desaturase enzymes such as Stearoyl-ACP desaturase, desaturates the saturated fatty acids. Acyl-carrier protein is one of the most protected proteins and is considered as hydrocarbon chains transporter in vivo. Most recently six ACP coding cDNAs were characterized from Olea europaea, which two of them, OeACP2 and OeACP3, showed the most occurrence in the fruit, leaf and flowers, respectively. Conversely, results demonstrated that OeACP2 and OeACP3, less than the former, alleles were involved in fatty acids biosynthesis and triacyl glycerol accumulation in fruits. In this research the quality and quantity of oil of Iranian olive germplasm evaluated by Firestone (1989) method and after recognition of the eligible genotypes (Mary and T18 as qualified and Shengeh and T20 as less qualified) OeACP2 gene was isolated from them and sequenced. Then the nucleotides sequence and possible efficient polymorphisms in coding regions was studied by bioinformatic softwares. The proposed proteins was studied according to their structural stability and function, bioinformatically. We found 19 nucleotide polymorphisms in exons 2 and 3 of OeACP2 that 11 cases were related to the high quality oil varieties and no significant structural changes were observed in the protein analysis.

Keywords: Olive, Fatty acids, ACP, Polymorphism
Qualitative optimization in DNA Extraction from Date tissue-culture Seedlings by comparing two different methods

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The palm propagation is very tedious and expensive because the number of saucers in each tree is limited, so that nowadays tissue culture in palm is commonly used. Tissue culture and micropropagation technique plays a significant role in increasing the production of palm seedlings, however sometimes genetic variation which is called somaclonal variation occurs. The molecular markers are among the biological elements used as laboratory probes to find and identify an individual, tissue, cell, cell nucleus, chromosome or gene and are introduced in various forms of the allele. In order to determine the genetic stability of these plants, we must first get access to their DNA. Therefore, the first step is to determine a DNA extraction method that is fast, safe and cost effective. Various methods have been reported by various researchers to minimize DNA extraction processes. The aim of this study was to optimize DNA extraction from palm seedlings. First, the Doyle& Doyle (1990) method and then the modified Margaritopoulos (2003) method were examined. The quality and quantity of DNA extracted were measured by spectrophotometric and gel electrophoresis methods. Also for extraction of DNA quality, the SSR technique was performed using a random primer called mPdCIR15. Finally, it can be stated that Margaritopoulo (693.85ng µl−1) method is recommended in case of using RNase, Proteinase K and increasing the washing steps to reduce the amount of contamination as a convenient and cost effective method for DNA extraction in this plant.

Keywords: DNA extraction, Genetic stability, Dates

Role of most cultivated woody plants in Tehran city to reduce air pollution

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Particulate matter is an air contaminant in urban and especially industrial areas that affects human health, ecosystem health. In recent years the role of plants are more concerned. Urban cultivated trees and shrubs can improve air quality through the direct removal of air pollutants. In present study, the ability of five most cultivated shrubs in Tehran for removing particulate matters (PM10 and PM2.5) was compared to each other. So, Ligustrum vulgare, Nerium oleander, Berberis thunbergii, Cotoneaster horizontali, and Euonymus japonicas Were selected. The results showed that Berberis is the most effective and Nerium was least effective taxa to reduce particulate matters. However, the pollution level of the sites has direct effects on the rate of PM deposition on leaves.

Key words: Air polluteon, Particulate matter, Urban green space
Green Algae: Bioindicator of Water Quality in the Rivers, Neyshabur, Iran

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In recent years, as water scarcity has been exacerbated, the focus on water resources management has been twofold. The optimal use of fresh water resources in each country requires the development of appropriate methods for monitoring and maintaining water quality. Industrial and agricultural activities are the main commercial activities that endanger the health of water resources due to the release of hazardous wastewater. Considering the limitations of common methods applied to assess the amount of water contamination, development of alternative approaches to measure the physicochemical properties of water has always been considered. Paying attention to the potential of algae communities in declaration of the level of pollution in water resources, they can be served as suitable biological indicators. In the present study, the release of garden wastewater to the river of Bojan village from the city of Neyshabur was carefully investigated. Effects of excessive nutrient supplementation such as Sequesterene resulted in 7-fold increase in the concentration of Fe\(^{2+}\) ion in comparison with the Control. It were significantly correlated with the changes in algal communities and their frequency of observation. The results of this study showed that the use of algal species as a suitable biochemical indicator could be useful for investigation the presence and concentration of pollutants such nutrients and iron ion, in particular. The results of this research could be used in better recommendations for gardening and more accurate management of wastewater entering the riverside.

Keywords: Bioindicator, Biodiversity, Drainage, Fertilizer

Relationship between Halocnemum, Salicornia and Suaeda halophyte plants and soil in Rahmanloo port of Ajabshir

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The southeast saline lands of Urmia Lake have more than 85,000 hectares extension. For study the relationship between soil nutrients and three halophyte species (Halocnemum, Salicornia and Suaeda) in Rahmanloo port of Ajabshir, the land unit maps were prepared and Random sampling was done from selected plants. Soil samples collected from rhizosphere, the physical and chemical properties and nutrients (calcium, magnesium, sodium, potassium and chloride) analyzed. Species selection for sampling with coverage percentage and presence of species in each unit was done using Kriging scale. The results showed that micronutrients content in roots was higher than shoots, whereas inverse is true about macronutrients except for calcium. Soil properties such as moisture, pH, carbonate calcium, organic matter and soil texture have been effective in different plant species according to vegetative conditions. Halocnemun was found mainly in light textured soils (sandy loam) compared to other selected plants. The soil of Suaeda plant can tolerate higher electrical conductivity and pH than Salicornia, also Salicornia ratios compare to Halocnemun. Chloride and sodium accumulation indicates that chloride and sodium content decreased significantly from Suaeda to Salicornia and Halocnemum, respectively. Because of low Sodium Adsorption Ratio (SAR) content due to high presence of calcium and magnesium, adverse effects of sodium decreased, that is a characteristic of calcareous soils. There was a significant positive correlation (\(P<0.05\)) between available soil nutrients and nutrient contents in studied plants. Therefore, due to relationship between plants and soil characteristics, these results can be used for sustainable land management in saline regions.

Keywords: Margin of Urmia Lake, Salinity, Nutrients, Land management
Evaluation of air pollution stress on physiological parameters in Tree species (*Ailanthus altissima* (Mill.) Swingle) in two seasons and two areas of Tehran city, Iran

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Environmental stresses such as air pollution cause different types of changes such as morphological, physiological and biochemical levels in plants. To resist these tensions. The persistence of environmental stresses that leads to oxidative stress in these plants makes them antioxidant compounds which reduces the effects of stress on the plant and allows the plant to survive under such conditions. This research was carried out to investigate and evaluate some of the physiological and biochemical parameters of the *Ailanthus* tree under the stress of air pollution during two seasons in Tehran. In order to deal with these stresses, plants showed different responses at different levels, including some physiological parameters such as enzymatic and non-enzymatic antioxidants to oxidative stress that reduces the effects of stress on the plant. The purpose of this research is to investigate some of the physiological and biochemical parameters of the *Ailanthus* tree under the stress of air pollution during two seasons in Tehran. In this research, some physiological parameters of the species of the tree species were evaluated during spring and summer (air pollution is increasing gradually) in 2017 in Punak (fairly clean area) and Azadi (polluted area) (respectively 5 and 9 of Regional Tehran residential districts in Tehran from where sample leaves were collected. Carotenoids contents, Catalase, Ascorbate peroxidase, and PAL activities. The results of this study showed that air pollution in area 9 (full of pollutants) in the spring and summer season significantly increased the enzymatic and non-enzymatic antioxidants in comparison with the area 5 in this tree. Which indicates its potential in resisting the air pollution stress. Therefore, it could be planted in polluted areas of Tehran.  
**Keywords:** Air pollution, *Ailanthus altissima*, Physiological parameters, Antioxidant properties, Tehran city

Effect of endophytic fungus *Piriformospora indica* on Pb partitioning in safflower (*Carthamus tinctorius* L.) under Pb toxicity in soil

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Root endophytic fungi play a crucial role in improving plant performance and participate in fungus-assisted phytoremediation in their hosts. The present experiment investigated the influence of root endophyte fungus *Piriformospora indica* on Pb accumulation of root and shoot in *Carthamus tinctorius* L. (safflower) cv. Sina in soil contaminated with four levels Pb (0, 400, 800 and 1200 mg Pb/kg soil) using Pb (NO₃)₂ solution under greenhouse conditions. By increasing Pb concentration in the soil, Pb accumulation in root and shoot were significantly enhanced. A significant increase in Pb accumulation in root and a significant reduction on shoot Pb amount was observed under fungal symbiosis. Also, *P. indica*-inoculated plants had lower translocation factor (TF) and bioaccumulation factor of Pb in the shoot (BFS), whereas higher bioaccumulation factor of Pb in the root (BFR), in comparison to non-inoculated ones. The results from our study illustrated that *P. indica* can be used as an important contributing factor for the Pb immobilization of host roots in phytoremediation process.  
**Keywords:** Endophytic fungus, Heavy metal stress, Safflower, Lead accumulation
MicroRNA-34a promote apoptosis in T-cell acute lymphoblastic leukemia cell line (Jurkat)

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T-cell acute lymphoblastic leukemia is a lymphoid malignancy affected by oncogenic transformation of immature T-cell progenitors. Acute lymphoblastic leukemia currently accounts for 20-25% of adults and 10-15% of pediatric cases. Clinically, T-ALL patients represent high white blood cell counts. MicroRNAs are a class of small, highly conserved non-coding RNAs containing ~22 nucleotides that interact with the mRNAs of coding genes to direct their post-transcriptional repression. MiR-34a is a tumor suppressor with lost or reduced levels of expression in many cancers, including T-ALL. Jurkat T-cell acute lymphoblastic leukemia (T-ALL) cells were maintained in RPMI 1640. MiR-34a mimic was transfected using jetPEI in vitro DNA transfection reagent. Cell viability was assessed with 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. Total RNA was extracted from the cells by using TRIzol reagent. Then, the flow cytometry assay was exploited to measure cell death and apoptosis stage. qPCR analyses showed that in Jurkat cells after transfection with miR-34a mimic (at the concentration of 5nmol) the expression of miR-34a mRNA was effectively increased compared with the control group. MTT assay results of microRNA were dose-dependent and at the 5nmol concentration of miR-34a, cell viability was less than their value in the control group. According to the flow cytometry assay result, in the transfected cells, miR-34a mimic (at the concentration of 5nmol) was able to induce apoptosis in Jurkat cell line. Our results suggest that the miR-34a effectively decreases the viability of T-cell acute lymphoblastic leukemia cells, induces apoptosis in this cell line, and therefore could be considered as a potent adjuvant in T-ALL therapy.

Keyword: Cancer, MiR-34a, Apoptosis, Jurkat cell line

Preparation, evaluation and characterization of biomimetic DBM scaffold

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Bone tissue engineering is a combination of 3D, three-dimensional, scaffolds and cells for repairing damaged or defective bone tissue. Therefore, extracellular matrix (ECM) derived scaffolds are a useful tool for providing a (micro) Environment for precisely evaluation of cellular interactions in 3D. The prepared fresh cow bones were converted into small pieces, and Freeze dried after physical and chemical decellularization. After processing by mixer mill under liquid nitrogen, derived nano powdr, were used for production of the DBM derived biomimetic electrospun nanofibrous scaffolds. Radiographic methods, electrolyte analysis and calcium oxalate tests were used to prove calcification. The nanofibrous scaffolds have been verified for surface properties, biodegradability and porosity. The results from the scanning electron microscopy, revealed the appropriate morphology and surface properties of them. After the biocompatibility evaluation of scaffolds for the human umbilical cord derived mesenchymal stem cells, the cells growth, proliferation and adhesion to the scaffold were confirmed using cytotoxicity evaluation via MTT-assay and microscopic studies. Therefore, this biomimetic scaffold may be used as a suitable candidate in regenerative medicine and bone tissue engineering.

Keywords: Nanofibrous scaffolds, DBM, Biomimetic, Stem Cell, Biocompatibility, MTT
The Role of circulatory microRNA in metabolic alteration of endometriosis patients

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Endometriosis is clinically described as the growth of endometrial glands and stroma outside the uterus and inside the peritoneum cavity. This disease often leads to dysmenorrhea, dyspareunia, episodic abdominal pain, and bowel symptoms and sometimes it linked with symptoms unconnected to the reproductive tract, for instance, women with endometriosis have been shown frequently to have subordinate body mass index (BMI) than those without the disease. Although there is not enough information regarding the pathophysiology of this disease may be dysregulation of expressed genes can be the principal reason for these clinical symptoms. Regulations of expressed genes are related to circulating microRNA and are known to be differentially expressed in the sera of women with endometriosis compared to the healthy group. Here we sought to determine whether endometriosis-related differential miRNA expression could induce changes to normal metabolic gene expression in women with endometriosis. These findings may explain the clinically-observed low body mass index of patients with endometriosis and contribute to our understanding of endometriosis as a complex and systemic disorder.

Keywords: Endometriosis, BMI, MicroRNA

Cyanobacteria: a new bioorganic fertilizer for medicinal plants, with emphasis on chamomile (Matricaria chamomilla L.)

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Chamomile or Matricaria chamomilla L. is one of the most important medicinal plants which grow in many parts of the world naturally. The essential oil of flower head has many therapeutic effects. In this investigation the effect of several cyanobacteria suspension (Nostoc spongiforme, Anabaena vaginicola, Nostoc punctiforme) on chamomile growth and essential oil was investigated. These cyanobacteria species were isolated from medicinal plants bed and then purified. Application of cyanobacterial suspensions for pot plants were conducted every month. After three months of planting, the growth parameters of plants such as root and shoot height and fresh weight, root and shoot dry weight, flower head diameter and weight of flowers were measured. Hydrodistilled essential oil composition of the flower heads in control and treated samples was assessed using GC-FID and GC-MS. Statistical analysis showed that there was a significant difference in the studied parameters compared to the control at the 0.05 level. Addition of some cyanobacterial suspensions had a positive effect on quantity as well as the quantity of some essential oil constituents.

Keywords: Essential oil, Cyanobacteria, Compositae, Medicinal plant
Extraction of inulin from Jerusalem artichoke (*Helianthus tuberosus* L.) by using High-intensity ultrasound in the north of Iran

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Inulin a polysaccharide that is a mixture of fructose oligomers and polymers. This is a prebiotic sugar, which is found in more than 36000 types of plants. In this research, *Helianthus tuberosus* L. is wild and tuberous, from the sunflower family, which is a source of inulin sugar. In the present study, inulin is extracted from *H. tuberosus* populations of Guilan, Golestan and Mazandaran provinces and then the best population in terms of inulin amount is introduced. After collection of tubers, 0.05 gr of each tuber was measured after that methanol extract was prepared by using 1.5 ml of potassium phosphate buffer. The samples were moved to ultrasonic waves for 10 minutes. In addition, for checking the sugar amount, Anthron method was used. Then the samples for 8 minutes in a water bath of 100 °C were placed. Finally, the amount of total inulin and fructose were measured by spectrophotometry at wavelengths of 520 and 625 nm. The results showed that the levels of the sugar in studied populations are not similar and same. The highest amount of inulin absorbed in Ramsar (361.87 DW), Gorgan (336.37 DW) and Astaneh Ashrafieh (319.12 mg/g dry weight) populations. Several ecological factors including light intensity, number of sunny days per year, moisture content, and altitude and soil acidity have significant effects on creation of different amounts of inulin in all studied populations.

**Keywords:** Inulin, *Helianthus tuberosus*, Ultrasound, Spectrophotometry, North of Iran

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Excessive, improper Acanthophyllum harvesting

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Sesame or Tahini halvah, known in Iran as Halva Ardeh, is known in many European and Asian countries. It is produced and consumed in various forms from the Balkans and Poland to the Mediterranean region, the Middle East, Turkey and the Caucasus. Soapwort is one of the ingredients traditionally used in this halva in Iran. Some species of Caryophyllaceae contain significant amounts of saponin in their roots whose extracts are used in the chemical, pharmaceutical and health industries. In different countries, this substance is produced by boiling wooden roots of various species. In Mediterranean countries the root of *Saponaria officinalis* is used, in Turkey some *Gypsophila* perennial species and *Ankyropetalum* are used. However, in Iran it is derived from the root extract of several species of *Acanthophyllum* (such as *Acanthophyllum laxiusculum* and *A. squarrosum*). In Iran, halva factories usually buy Soapwort from traditional producers (herbal distributors). Most traditional sweet factories are located in the central regions of Iran, especially Yazd province. Picking wild *Acanthophyllum* species in this province has limited the population of this plant, therefore, plant picking in this province is currently banned. Due to limited monitoring around the Torbat-e-Jam and the Afghanistan border, herbal distributors now usually obtain their *Acanthophyllum*’s roots from these areas. Since plant harvesting in these areas will soon lead to the extinction of these valuable species in terms of biodiversity, it is suggested that in the Yazd province or adjacent provinces establish industrial farms for different usable species. By producing enough plants, while preventing plant picking from natural habitats, this substance can also be used in the production of other products.

**Keywords:** Biodiversity, Conservation biology, Tahini
CXCL10 is a member of the CXC chemokine family. It is secreted from a variety of cells in response to IFN-γ and stimulates a range of inflammatory responses via binding to its receptor, CXCR3. CXCL10 has a pivotal role in the pathogenesis of various infectious diseases, cancers, inflammatory and autoimmune diseases. It has been put forward as a potential biomarker and therapeutic target in diagnosis and treatment of these diseases. In the present study, production of camel heavy chain antibodies (HCAbs) specific to the CXCL10 is reported. In this regard, recombinant CXCL10 was used for immunization of camel and subsequently, the CXCL10 HCAbs were obtained. Afterward, three subclasses of IgG were separated using protein A and protein G affinity columns, characterized by SDS-PAGE and confirmed for specific binding to the CXCL10 using ELISA. These IgG subclasses successfully recognized CXCL10 and a strong and specific reactivity towards CXCL10 were observed. Therefore, the selected HCAbs and their corresponding expression library could be used to develop a recombinant variable domain of these HCAbs (nanobody or VHH) as a new possible strategy for treatment of multiple sclerosis and other autoimmune diseases.

**Keywords:** CXCL10, Polyclonal antibody, Heavy chain antibody, Nanobody, Multiple sclerosis, Chemokine
Study of biodegradation of oil pollution in laboratory grade

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The effects of oil pollution on human health have led to the provision of solutions. Identification of oil-degrading bacteria can convert heavy hydrocarbons into simpler derivatives. Soil samples collected from 5 polluted areas. One gram soil was inoculated with 100 ml of synthetic medium containing at least 1% oil as the source of carbon and incubated at 20 rpm for 5 days. After observing the turbidity in the broth medium, 100 μl was transferred to agar medium containing 1% oil. For purification single colonies were cultured in an agar medium containing 1% oil. The morphological characteristics of the strains were described by Gram staining and, for the analysis of the rate of degradation, the residual oil was separated by diethyl ether. The weight of oil was decreased in comparison to the initial weight of oil. Eleven strains of gram-negative and 1 strain of fungi were identified. Out of 12 isolated strains, 2 strains showed the highest degradability, which included strains screened from Shahre refinery (64%), Jayrood repairer (57%), wastewater from the Moon-Urban refinery (52%), Samen Al-Haj Square Bus Terminal (47.57%), ethanol purifier flood in Shah Refinery (44.7%). These isolates are Gram-negative bacilli. The strains can play a role in potential the reducing of pollution at the field scale. This method of studying is easier, less costly and faster than spectroscopy and chromatography. **Keywords:** Biodegradation, Liquid petroleum, Oil-degrading bacteria, Oil pollution.

Green synthesis of Fe₃O₄/Ag nanocomposite by Spirulina platensis cyanobacteria extract and evaluation of their effect on norA and norB efflux pump genes expression in ciprofloxacin-resistant strain of Staphylococcus aureus

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Today, the overuse of antibiotics has led to the emergence of antibiotic-resistant strains of bacteria, and a worldwide crisis in the battle against them. Accordingly, the discoveries, production, and prescribing of new antimicrobial compounds have become one of the most important concerns of researchers. The antibacterial activity of silver nanoparticle, as well as Fe₃O₄ nanoparticles, had been previously studied and widely described. Synthesis of nanocomposites from the combination of two types of nanoparticles with antibacterial effect increases the performance of both materials. There are several methods for the synthesis of nanoparticles among which, green techniques that refer to the synthesis of metal nanoparticles by natural sources were developed and considered as an eco-friendly and cost-effective alternative for chemical and physical methods. Here, we report the biosynthesis and characterization of Fe₃O₄/Ag nanocomposites by Spirulina platensis cyanobacteria and their impacts on the expression of efflux pump genes in ciprofloxacin-resistant strains of Staphylococcus aureus. S. aureus is one of the most common pathogens responsible for nosocomial infections that can acquire resistance to antibiotics by different mechanisms including drug extrusion through efflux pumps such as NorA and NorB. The physical properties of biosynthesized nanocomposites measured and confirmed by ultraviolet-visible spectroscopy, Fourier-transform infrared spectroscopy, X-ray diffraction, scanning electron microscopy and transmission electron microscopy. Minimum inhibitory concentration (MIC) of ciprofloxacin in antibiotic-resistant S. aureus strains determined in the presence of Fe₃O₄/Ag nanoparticles by broth microdilution method. The effect of Fe₃O₄/Ag nanoparticle on the expression of norA and norB genes were evaluated by Real-time PCR. The UV–Visible spectroscopy exhibited the presence of absorption peak at 390 nm that clearly indicates the formation of Fe₃O₄/Ag NPs in the solution. SEM and TEM microscopy of biosynthesized Fe₃O₄/Ag NP showed a spherical shape with an average size of 10 ± 2 nm that are well dispersed with the concentration of 2.3 × 10⁵ particles/mL. XRD patterns in agreement with the results of electron microscopy and confirmed the crystalline structure of the synthesized Fe₃O₄/Ag NPs. The results of FT-IR analysis confirmed the stability and biocompatibility of biosynthesized nanoparticles by capping them with biomolecules in the cyanobacterial biomass extract. The MIC values for ciprofloxacin and Fe₃O₄/Ag NPs were 32 μg/mL and 64 μg/mL, respectively. Moreover, reduction of the antibiotic MIC in the presence of nanoparticles to at least one-quarter of the initial value clearly evidenced synergistic effect of two antibacterial agents to effective inhibition of bacterial growth. In the presence of Fe₃O₄/Ag NPs, the expression of norA and norB genes were significantly increased that may be the reflection of the bacterial effort for survival. In conclusion, the green synthesized Fe₃O₄/Ag NPs enhanced the antibacterial effect of ciprofloxacin against ciprofloxacin-resistant S. aureus. Therefore, the Fe₃O₄/Ag nanocomposites can be used as effective inhibitors of antibiotic resistance in medicine. **Keywords:** Antibacterial activity, Fe₃O₄/Ag nanocomposite, Efflux pump, NorA, NorB, Spirulina platensis cyanobacteria.
Synthesis of magnetic carboxymethyl cellulose nanoparticle for immobilization of Laccase enzyme

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An efficient and novel immobilization of Laccase has developed using a magnetic hybrid nanostructured material of iron oxide and carboxymethyl cellulose (Fe$_3$O$_4$–CMC) as support. The resultant magnetic hybrid nanostructured material was characterized by X-ray diffraction (XRD) and Fourier transform infrared spectroscopy (FTIR). For this purpose, at the first iron oxide nanoparticle was synthesized from the inorganic salts of Fe (II) and Fe (III) by co-precipitation method and then iron oxide nanoparticles modified with carboxymethyl cellulose. Laccase enzyme (0.025 g/ml) loaded into this magnetic hybrid nanostructured material by using 400 mM of N-Hydroxysuccinimide (NHS) and 20 mM of 1-Ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC) as the activator and incubation lasting 24 hr at aqueous state pH of 5.5 and room temperature. Under this optimum condition, the efficiency of laccase immobilization was at least 35%. According to our result reveal that Fe$_3$O$_4$–CMC is an appropriate and applicable matrix for Laccase immobilization and can be used in different processes.

Keywords: Magnetic nanoparticle, Carboxymethylcellulose, Laccase, Enzyme immobilization

The effect of multi-walled carbon nanotubes on morphological and physiological indexes of castor plant (Ricinus communis L.)

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Nanotechnology or using technology in the case of atoms and molecules is one of the most important techniques in the present century, which will affect the future of the world economy. This study was designed to examine the effects of multi-walled carbon nanotubes (MWCNTs) (0, 75, 100, 125, 250 and 500 μg ml$^{-1}$) on morphological and physiological indexes of Ricinus communis L. Measurement of root and shoots dry weight and length showed that increasing of MWCNTs concentrations have an adverse effect on them. Results show that there wasn’t significant difference (p<0.05) between MWCNTs concentration stem length, root length, shoots wet weight, Soluble sugars, and Catalase but there was significant difference (p<0.05) between MWCNTs concentration plant height, phenol, root dry and wet weight, shoots dry weight compared to control and its reason can be found in the effect of different results of MWCNTs in different concentrations on morphologic and physiologic of castor plant.

Keywords: Catalase, Phenol, Soluble sugars, MWCNTs, Ricinus communis L.
Combination of landscape ecology and population genetics as a new approach for the studies of animals' gene flow

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Combination of methods and concepts of population genetics, landscape ecology, and spatial statistics that create the approach of landscape genetics has emerged as a new research area in population ecology, wildlife management and conservation biology. It aims to study and predict how landscape features interact with micro-evolutionary processes, such as gene flow. In the context of habitat fragmentation, the current focus of landscape genetics is on assessing the degree to which landscapes facilitate the movement of organism (landscape connectivity) by relating gene flow patterns to landscape structure. Therefore, understanding of how landscape features affects dispersal and gene flow within and among populations and habitat patches is important to predict the effects of increased habitat modification and landscape changes on population persistence and processes of divergence. In order to researches in this field we should study two key stages. 1- Tracing of the spatial genetic distances using the high resolution of genetic markers, 2- relationship between the spatial genetic distances and landscape and environmental variables such as habitat patches and barrier that are calculated by geographical information system (GIS), using spatial statistics.

Keywords: landscape genetics; population genetic; landscape ecology; gene flow

Toxicological effects and subcellular partitioning of Cadmium in Aporrectodea rosea, Aporrectodea trapezoides and Eisenia fetida

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Recently, biomarkers in earthworms were widely used for assessing the effect of pollutants on the environment. There is necessary for using earthworms and their biomarkers based on sensitivity and their presence in field, but testing usually focused on Eisenia fetida. Therefore, after a year field study to identify and determine the dispersal of earthworm species in Hyrcanian zone (case study, chaloos), Aporrectodea rosea and Aporrectodea trapezoides were selected and put in front of E. fetida. The species were exposed to lethal (14 d) and sublethal concentrations (7, 14 and 28 d) of cadmium (Cd) nitrate in OECD soil. For this purpose, apoptosis, lipid peroxidation (MDA), Total Antioxidant Capacity (TAC), lethal toxicity (LC50), weight loss and subcellular partitioning were assessed as biomarkers to compare species sensitivities. Acute toxicity for Cd was E. fetida > A. trapezoides > A. rosea (p< 0.05). Cadmium caused significant inhibition in TAC and weight loss at upper treatments, and significant increase in apoptosis and lipid peroxidation in all three earthworm species (p< 0.05). Subcellular partitioning in all three species for Cd were: Cytosol> Granules> Debris (P< 0.05). Comparison of biomarker responses showed E. fetida was less susceptible to Cd exposure than A. rosea and A. trapezoides. Therefore, this study confirms the employment of A. trapezoides and A. rosea is more suitable than E. fetida for the soil Cd toxicity tests due to their sensitivity and dispersal in the field.

Keywords: Earthworm, Heavy Metal, Soil, LC50
Posters
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Molecular markers: key biotechnology tools in the research and development of biofortified cereals

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Biofortification of cereals is a new approach to manage micronutrient deficiencies in developing countries. In many micronutrient-deficient regions, cereal grains as the dominant staple food are naturally low in nutritional minerals, increasing the risk of hidden hunger. Biofortification of cereals is a new approach to manage micronutrient deficiencies in developing countries. Predictive cost-benefit evaluates have proven that biofortification is important in controlling of malnutrition. Biofortification is the development of micronutrient-dense food crops using biotechnology and/or conventional breeding practices. Biofortified crop system is highly sustainable over years, cost-effective, target low-income households, and available in remote rural areas. Micronutrient enrichment traits are present in the genome of cereals that could permit substantial improvement in grain zinc, iron, Cu and so on without negatively impressing grain yield. The genetically fortified traits are stable across various climatic environments and soil types. Biotechnology tools in genomic, molecular markers, identifying nutrient absorption enhancers and inhibitor genes can provide better and more efficient complementary breeding tools. A significant development in the next few years will be the use of molecular markers associated with accumulation of nutritional minerals such as Zn, Fe, and Cu in cereals. The identification of major-effect molecular markers can speed up the development of high yielding biofortified cereals even in micronutrient-deficient soils of Iran. DNA markers allow screening for micronutrient-rich crops independently of the environmental variability or growth stage. If markers are close enough to a gene of interest, they can be directly used in marker-assisted selection (MAS).

Keywords: Biofortification, Micronutrient deficiency, Food quality, Molecular marker

DNA methylation changes in the root of barley under long and short-term salinity stress

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Salinity is one of the most crucial factors, which inhibits crop production. Increasing evidence has revealed that epigenetic mechanisms modulate the gene expression in plants under abiotic stresses. Obviously, by understanding epigenetic regulation of plant growth and response to stresses, a heritable variation could be developed for crop breeding. In this study, the DNA methylation alteration under salt stress was analyzed in two barley cultivars differing in salt tolerance, namely Sahara3771 and Clipper. Coupled Restriction Enzyme Digestion-Random Amplification (CRED-RA) was utilized to detect CCGG sequence methylation alteration in shoot and root of the plants growing under 0 and 100 mM NaCl treatment. Leaf and root samples for DNA extraction were harvested 24 hours, 3 weeks, and 5 weeks after salt treatment. The results revealed that under salinity stress in the root of Sahara3771, 9.92% and 14.69% of sites were hypermethylated and demethylated respectively compared to the control. The average number of hyper and demethylated sites in the root of Clipper was 11.34% and 12.76% respectively. In Sahara3771, the number of hypermethylated sites was 1.5 times higher than demethylated sites. In addition, number of methyl-unchanged sites in Sahara3771 was higher than Clipper. Methylation changes induced by increasing stress duration. Then, under long-term stress number of hyper and demethylated sites respectively increased in Sahara3771 and Clipper. These results revealed that alteration of DNA methylation in plants could be a key factor in adaptation and toleration of plants to salinity.

Keywords: Barley, Salinity stress, Methylation pattern, CRED-RA
The effect of different ratios of coco peat and expanded clay on rooting and growth of pothos (Epipremnum aureum) stem cuttings

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The esthetic and psychological enhancement of interior environments and purification of the indoor air have become catalysts in promoting foliage plant production and increasing their wholesale value. Pothos is one of these popular indoor plants which it has beautiful leaves and stems. The propagation method is by stem cuttings and the composition of the culture medium will affect the rooting and growth of the cuttings. Therefore, this study was performed in a completely randomized design with 5 treatments (different ratios of coco peat and expanded clay) to compare rooting and growth of the pothos stem cuttings. The compositions of these five potted mixtures were by volume: 1. 100% peat, 2. 100% expanded clay, 3. 50% coco peat +50% expanded clay, 4.75% expanded clay +25% coco peat and 5. 75% coco peat +25% expanded clay. The results showed that the highest root and leaf fresh and dry weight, leaf number, root volume and the longest root length related to 50% coco peat +50% expanded clay. Therefore, this pot mixture can be recommended for the propagation of this plant by stem cuttings.

Keywords: Expanded clay, Pothos, Rooting, Stem cutting

Identification of fig mosaic disease by NGS

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Fig mosaic (FMD) is a common disease in fig orchards in Iran with high economic importance its etiology is not fully clarified. Association of different viruses and variation of symptoms suggest that FMD may be caused by the synergistic effects in mixed-infection of several viruses. Although FMD has been seen in Iran since some decades ago, high current incidence in fig orchards of Fars province, as one of the main fig production regions, create a national alarm which necessitates a better understanding of the viral complex associated with the disease. Next-generation sequencing (NGS) offers a beneficial tool for studying the virome profile of diseases. The main challenge in identifying the virome profile of infected fig by NGS is that separation of RNAs of viruses from those of the host plant is difficult. Consequently, the virome metagenome needs to be analyzed with the host RNA (fig). Unavailability of the full genome of fig adds another level of complexity to the analysis. In this study, we developed a pipeline for detection of viral sequences based on RNASeq de novo assembly of short reads in healthy fig (to make reference genome). To this end, RNA samples were extracted from leaf pools of infected and healthy fig plants in three biological replicates and total RNA from leaves of healthy and fig-mosaic-infected plants were sequenced using Illumina HiSeq 2000 high throughputs sequencing (as 100 bp, paired-end short reads) to determine the pathogenic virome profile of FMD in Fars province. The obtained sequences from total reads (4,852 megabases) in infected plants were compared with those obtained from healthy fig (5,116 megabases). Then, viral + fig sequences were mapped to the constructed reference genome. Fig infecting viral sequences were collected from unmapped reads. The obtained viral short reads were used to construct contigs and were subjected to blast analysis against NCBI to identify the virome profile in infected figs. The results demonstrated the existence of fig fleck-associated virus (FFKaV), fig badnavirus-1 (FBV-1), and fig mosaic virus (FMV) in fig leaves of the infected plant. This method provides a rapid and cost-effective pipeline for detection of virome profile of infected fig using the mixed RNA sequences of viruses and fig.

Keywords: Fig mosaic disease; Next-generation sequencing
Reducing the disease severity of *Macrophomina phaseolina* by *Pseudomonas fluorescens* in greenhouse conditions

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Macrophomina phaseolina, the agent of charcoal rot, is one of the most important soil-borne diseases of common bean, which decreases its yield significantly. Its chemical control, not only causes environmental problems but also it is difficult and sometimes ineffective. Recently, researchers have focused on applying the biocontrol micro-organisms against plant diseases. Fluorescent pseudomonads by producing antibiotics, siderophore, hydrogen cyanide, and protease, are the most important biological control agents of plant diseases. In this research, for studying the biocontrol ability of *Pseudomonas fluorescens* strains against *M. phaseolina*, plants were infected by *M. phaseolina*, which were isolated from bean fields of Khoram-Abad. Virulence of fungal isolates was evaluated in the greenhouse, then a fungus with the highest pathogenicity was chosen for next. The biocontrol ability of 8 *P. fluorescens* strains, which have been proven as biocontrol agents of other plant pathogens against the fungal pathogen already, was examined in vitro by dual culture and antifungal activity of bacterial volatile and nonvolatile metabolites. *P. fluorescens* UTPf125, which significantly inhibited the pathogen growth more than others, was selected for greenhouse tests. Results showed that UTPf125 causes a significant increase in fresh and dry weight of the plant in the greenhouse in addition to decreasing the disease severity in root and crown of infected plants from 75% to 25%. All experiences have been done in 3 replications. Statistical analysis was performed using SAS ver.9.2 and Duncan test was used to show significant differences among treatments.

**Keywords:** Charcoal Rot, Antibiotics, Disease severity

Investigating the correlation between endorhizosphere population of *Pseudomonas fluorescens* biocontrol agent of *Rhizoctonia solani* and number of nitrogen-fixing bacteria nodes in bean root

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Bean is one of the most important members of Fabaceae, which is in a symbiosis with rhizobia, a species of atmospheric nitrogen-fixing bacteria. These bacteria establish nodes in bean roots and it’s effective in soil fertility. Yearly, after bean harvesting, large quantities of nitrogen are added to the soil. Rhizoctonia solani is the causal agent of Rhizoctonia root and collar rot is one of the most important diseases of the bean. Because of being a soil-borne pathogen, a wide range of hosts and high degrees of aggressiveness, its managing is difficult. The use of antagonistic microorganisms, especially *Pseudomonas fluorescens*, against it has been investigated as a promising control method. In this study, infected bean seedlings with *R. solani* were collected from Khomein. Virulence of fungal isolates was evaluated in the greenhouse, then a fungus isolate with the highest pathogenicity was chosen for next experiments. The biocontrol ability of 8 *P. fluorescens* strains against *R. solani* was examined in vitro. *P. fluorescens* P13, which was the most effective in controlling the pathogen in all experiments, was selected for next. For greenhouse studies, roots of bean seedlings were treated with 108 cfu/ml of P13. The results showed that biocontrol bacteria caused a 50% reduction in disease severity of Rhizoctonia rot. The endorhizospher population of P13 and number of Rhizobium nodes were evaluated in 15, 30 and 45 days after inoculation. Although, as time passed by, the population of P13 in the endorhizosphere decreased, but in the 45th day, a high population of this antagonist, about 3.2 × 105 cells bacteria/gr fresh root, recovered in bean endorhizosphere, which compared to the control plant, caused about 72% reduction in the number of Rhizobium nodes. In this interaction, the number of Rhizobium nodes decreased significantly, but no significant difference was observed in the dry weight of the treated plants and controls.

**Keywords:** Nitrogen-fixing bacteria, Biocontrol, Endorhizosphere
Evaluation of the inhibitory effect of some fungicides on Fusarium wilt disease in greenhouse cucumber

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Fusarium oxysporum f.sp. cucumerinum, the causal agent of Fusarium wilt is one of the most important diseases, which results in extensive damages to greenhouse cucumber every year. This pathogen is able to infect plants at all growth stages, decreases plant quality, and subsequently leads to plant death. This investigation was conducted to propose an appropriate fungicide in an efficient dosage for managing cucumber Fusarium wilt. The efficiency of different dosages of biological, organic and chemical fungicides on fungal growth inhibition was evaluated in vitro. These fungicides were included Amistar Xtra (0.75, 1 and 1.25 l/hectare), Tachigaren (2, 2.5 and 3 l/hectare), Uniform (1, 1.5 and 2 l/hectare), Lamardor (1.5, 2 and 2.5 l/hectare), Thiophanate-methyl (1, 1.5 and 2 kg/hectare), Fusar (1.5, 2 and 2.5 l/hectare) and Trianum P.

The EC50 of all fungicides calculated first, then varying doses of fungicides were prepared and added to PDA culture. A 0.5-cm diameter PDA plug containing mycelia of the fungus was placed in the center of each plate. After a week of incubation at 26°C, the fungal growth in different treatments was evaluated and compared. The results showed that Amistar extra (1.25 l/hectare) had the most efficient inhibitory effect (more than 98%) on fungus growth in vitro. All experiences have been done in 4 replications. Statistical analysis was performed using SAS ver.9.2 and Duncan test was used to show significant differences among treatments. The efficient treatments that had the highest fungal growth inhibitory effects will select to apply in greenhouse condition.

Keywords: Fusarium wilt, Fungicide, Inhibitory effect

Zinc application alleviates drought stress in bread and durum wheat genotypes under dryland conditions

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Drought stress and zinc (Zn) deficiency are abiotic stress factors limiting crop production in Iran, especially in rainfed areas. In this study, the effects of Zn application and drought stress was investigated on grain yield of 10 wheat cultivars (6 bread wheat and 4 durum wheat) grown in a Zn-deficient calcareous soil over 2 years under rainfed condition in Drylands Agricultural Research Institute (DARI) where drought causes substantial yield reduction. Plants were treated with (+Zn: 10 kg Zn/h, as ZnSO4.7H2O) and without Zn (-Zn) under rainfed condition in a randomized complete Block Design (RCBD) with 3 replications. Zinc fertilization could enhance biomass, harvest index and grain weight of genotypes that resulted in 7% grain yield increase on average in both years. Increases in grain yield varied among genotypes from 1% to 16%, and durum genotypes showed higher rising. Accordingly, there was variability in Zn efficiency of genotypes (ratio of grain yield under Zn deficiency compared to that under Zn fertilization). Durum wheat genotypes generally showed low levels of Zn efficiency and grain yield under Zn deficiency. Overall, there was a poor relation between Zn-efficiency values and grain yield increase. The results presented here demonstrate the existence of variation in Zn efficiency among bread and durum wheat genotypes, and suggest the possibility of breeding for higher grain yield and improving Zn efficiency in Zn-deficient calcareous soils under rainfed conditions. Moreover, Zn application would increase bread and especially durum wheat productivities in drylands.

Keywords: Genotypic variation, Zinc application, Drought tolerance, Bread and durum wheat
Diversity of plant species of Khan Gormaz protected area of Zagros mountain range in Tuyserkan

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Khan Gormaz protected area is located in the pre-mountain range of Zagros in the west of Hamedan province. The area is cold and semi-arid. The method used in this research is the conventional method of regional floristic studies. Firstly, the geological studies of this area were identified. The scroll method was used to collect the plants. Among the collected plant species, the Asteraceae family (composite) is the largest taxon in the region in the genus and The Lamiaceae family is ranked second. The predominant type of Astragalus genus is characterized by cold and semiarid conditions in the region. Regarding the region’s richness in terms of plant species, it is recommended that knowledge-based companies be set up in this field in order to arrange for the preservation of these valuable reserves and the optimal exploitation of the resources and to preserve and enhance genetic resources (for example: use of plant seeds, evaluation of the quality of essential oils of medicinal plants of the region, application of Plants in Pharmaceutical Sciences, extraction of effective therapeutic extracts and...). It can also serve as a model of Ecotourism for the optimal use of the Zagros region and preservation of its genetic resources, as well as for the Zagros economy.

Keywords: Plant species, Khan Gormaz protected area, Zagros, Tuyserkan

Effects of the halophyte *Salsola crassa* on remediation of industrial environment

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Due to the increasing number of pollutants, the use of plants for the purpose of environmental refinement has been considered for some time. In this research we used the halophyte *Salsola crassa* M.B. with the aim of reducing industrial pollutants with an emphasis on heavy metals (Zn, Cd, Cu, Ni and Pb), and to determine the aggregation organs. For this purpose, a total of 60 control and treatment plants were identified and enclosed at the beginning of germination in the area of the industrial city of Shiraz. They were irrigated twice a week for three months by industrial wastewater (treatment) and tap water (control), then collected. In order to calculate the amount of heavy metals in the plant tissues, the shoots and roots were separated and prepared for analysis by ICP (by the method of solubility in hydrochloric acid). The results showed that the concentration of Cu in the treated to the control root was 2.07, Ni was 1.68, Pb was 1.06 and Zn was 1.14; while their concentrations in the treated to the control shoots were as follow: Cu (1.61), Ni (1.78), Pb (1.84) and Zn (1.63). The Cd concentration in both the treated and control samples had no significant changes. It can be concluded that the shoots and roots of *Salsola crassa* have been successful in absorbing heavy metals Cu, Ni, Pb and Zn. The roots have been successful in absorbing Cu and Ni, and the shoots absorbed Ni, Pb and Zn effectively. Therefore, this plant could be a good candidate for industrial remediation in the environment.

Keywords: Phytoremediation, *S. crassa*, Heavy metals, ICP
Structural changes of the halophyte Suaeda maritima as a result of contamination with industrial wastewater

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Unfortunately, in line with the industrialization of the modern world, living organisms are highly exposed to pollutants that may be hazardous to them and cause irreparable complications. In the present research we study the effects of industrial raw sewage which contained some heavy metals such as Cd (0.06ppm), Cu (0.16ppm), Zn (0.20ppm), Pb (0.02ppm) and Ni (0.02ppm), on the halophyte plant, Suaeda maritima (L.) Dumort. To achieve this purpose, after dividing the plants into the control and treatment groups, irrigated them with tap water and raw sewage every other day for three months, dug out and separated into shoot and root parts. They were fixed in FAA, and after a month, Preparation process in the method of embedding in paraffin were done according to conventional cell and histology methods. After serial sectioning with rotatory microtome, the sections were died by H&E and methylene blue-Carmen alum staining. Observation and photography were done by light microscope. The results confirm the resistance of the plant under high stress conditions. Despite of the viability and plant growth, treatment plants had less growth and pale color. The epidermal cells of the stem in treatment samples were smaller than the control. The cortex parenchyma cells in both stem and root were deformed, became larger and less in number. The thickness of the cuticle on the leaf was reduced in the treatment. While the xylems were arranged regularly and uniformly in size in the control root, they became irregular and non-uniform in size so that very big and very small xylem elements were outspired outside the stele. As a result, we can definitely state that the pollution of the industrial wastewater can make changes in the structure of all vegetative organs of the plant.

Keywords: Suaeda maritima, Anatomy, Industrial Raw Sewage, Plant

Effects of fire products on soil seed bank germination of Conringia orientalis and Stipa haussknechtii in semi-steppe rangelands

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Fire products (heat, smoke, and ash) stimulate germination in a lot of plant species. However, little information is available on the effects of fire products on germination of seeds of plant species in the seed bank of the semi-steppe rangelands. This study aimed to investigate the fire products on density seeds of soil seed bank in Semi-Steppe rangelands. A stratified random sampling was used to collect soil samples of Karsank region in Chaharmahal and Bakhtiar province. A stratified random sampling was used to collect soil samples of the studied region and treatments with fire products were applied to them. Four treatments including heat shock, one treatment ash and control treatments (without any treatment) were tested. Overall, two species of Conringia orientalis and Stipa haussknechtii had the highest germination densities in the seed bank. The germination percentage of Conringia orientalis increased following 80 °C heat treatment with average germinating seeds of 304 seeds per m$^2$ in comparison with the control. Also, in Stipa haussknechtii, 80 °C heat treatment increased germination density, but this increase was not significant. Treatment of 80 °C heat treatment has the greatest effect on the germination density of these two species in the soil seed bank of the region. This is due to the fact that these two species, such as most plant species in the semi-steppe regions of the country have physiological dormancy. Accordingly, suitable heat treatments can be used to increase the germination density of Stipa haussknechtii and to improve and restore the rangelands.

Keywords: Fire products, heat, density, Semi-Steppe, restoration
Lead uptake and phytoremediation in Marrubium cuneatum plant under different concentrations of sulfur (S)

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Phytoextraction is one of the phytoremediation strategies in which plants take up pollutants from the environment and accumulate them in the plant shoot. Marrubium cuneatum (Lamiaceae) can be used in phytoextraction due to the relatively high biomass production and lead (Pb) accumulation in its shoots. In this research, the effect of increased sulfur (S) concentration on Pb absorption potential of Marrubium cuneatum has been investigated. Plants were cultured in hydroponic media under three S concentrations: 12, 24 and 48 mg/l. Pb (as Pb (NO3)2) was added to the medium in 200 mg/l concentration. After two weeks, the plants were harvested and the roots and shoots were separated. Root and shoot length, fresh and dry weight along with Pb content in root and shoot dry matters were measured. The results indicated that only shoot length, root fresh weight and shoot Pb content, under different concentrations of S, showed a significant difference and other parameters did not show any significant differences. The highest changes were observed in shoot Pb content, with an average Pb concentration of 49 mg/kg, in 12 mg/l S treatment, reaching 1050 mg/kg, in 24 mg/l S treatment. According to the results, it seems that increasing the amount of plant-available S to 24 mg/l can increase the shoot Pb concentration up to 1000 mg/kg but more S increase does not affect Pb absorption significantly.

Keywords: Phytoremediation, Phytoextraction, Marrubium cuneatum, Sulfur, Lead

The conservation status of Allium ubipetrense in Iran

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In terms of climatic and ecological diversity, Iran has a relatively rich and unique flora, and the endemic condition of some plant species has caused the global importance of Iran and its dependent flora. Endemic species are considered as the most important plant species in the country. Iran with more than 126 species of Allium L. is one of the important centers of its diversity. In this research, the conservation status of an endemic species of onion in Iran is determined according to the criteria of the International Union for Conservation of Nature (IUCN). Based on field studies and a survey of 59 accessions from different areas of the distribution of this species in the years 96-92, using three criteria for extent of occurrence (EOO), occupation level (AOO) and number of population and calculations performed by Geocat software, A. ubipetrense R.M. Fritsch despite the presence of relatively numerous habitats, is considered to be rare and vulnerable or endangered plants due to its limited occupation and small populations, and should be studied in terms of conservation. In most of these habitats, individuals of this species are very limited and, in many cases, are seen singly. Given the total number of factors including individual numbers, fertility rates, highly dispersed accessions, limited restoration due to limited seed production and degradation of habitats, which are generally due to various reasons, including warming and excessive weathering, this species is endangered.

Keywords: Conservation status, Endemic species, Allium, Iran
Phytoremediation of cyanide contaminated soil from Muteh gold mine by using *Sorghum bicolor*

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Soil contamination with industrial pollutants has giving rise to concern for humans and other living organisms. Although cyanide (CN) is highly toxic and dangerous substance, it is economically attractive for extracting gold from ore bodies and it has been of great interest to researchers in the last decades. Phytoremediation is a cost-effective plant-based approach of remediation that takes advantage of the ability of plants to concentrate elements and compounds from the environment and to metabolize various molecules in their tissues. Phytoremediation can be used to clean up cyanide in the environment and soil. In the present study, the effects of different concentrations of cyanide on biochemical responses (changes in protein and proline levels in the plant) and tolerance in *Sorghum bicolor* are analysed. In this research, wastewater was prepared and concentrated after mixing with normal soil. After completing the vegetative period, cyanide, protein and proline changes were measured and Performance data was analysed with spss software and comparison multiple range test of means with Duncan test. The analysis of cyanide reduction data and its accumulation in the plant showed that the Phytoremediation is a suitable method for clean-up cyanide contaminated soils, and the absorption of cyanide by the plant causes biochemical changes (increased in protein and proline level) and reduces the amount of cyanide in the contaminated soil. S. bicolor could grow in 12.69 grams per kilogram of cyanide which resulted in approximately 50% cyanide from the wastewater, so increased the protein and proline level. S. bicolor provided the highest phytoremediation efficiency. Thus, this study recommends using *Sorghum bicolor* for phytoremediation of cyanide in gold mine areas.

**Keywords:** Cyanide, *Sorghum bicolor*, Phytoremediation, Protein, Proline

Air pollution tolerance index (APTI) of tree species *Ailanthus altissima* (Mill.) swingle at two seasons and in two areas of Tehran city, Iran

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The circulation of phytotoxic air pollutants from vehicular traffic, industrial factories and geographic location are the main causes of air pollution in Tehran. One of the ways to deal with air pollutants is tree planting in the public roads, parks and green spaces. The Resistance of plants to air pollutants could be evaluated by the value of APTI. The plant with high APTI value is Resistance to air pollution. The present study was aimed to evaluate the tolerance of *Ailanthus altissima* (Mill.)Swingle to air pollution in two residential districts of Tehran during spring and autumn 2017. The leaf sampling of the selected trees was conducted in the Punak (area of 5), and Azadi (area of 9). Then leave samples transferred to the laboratory for biochemical analysis. Four biochemical parameters including ascorbic acid content, leaf extract pH, relative water content (RWC) and total chlorophyll content were assessed and APTI value was calculated. The results of this study showed that the *Ailanthus altissima* (Mill.)Swingle in two seasons spring and autumn in area 9 with higher APTI value (11.65) have significant increase compared to area 5 (with APTI value 9.25). So, *Ailanthus* recommended being planted in the polluted areas of Tehran.

**Keywords:** Air pollution tolerance index (APTI), *Ailanthus altissima*, Tehran city
The changes in photosynthesis pigments and proline contents of safflower in response to endophyte fungus under different levels of lead in soil

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Lead (Pb), as a heavy metal element, is one of the toxic metals in ecosystems that derived mainly from anthropogenic sources and in plants negatively affects their physiology, biochemistry and cellular ultrastructure. Symbiosis of beneficial microorganisms with plant roots can effectively enhance host tolerance to the metal toxicity under heavy metal stress. In this study, the influence of root endophyte fungus Piriformospora indica on some physiological parameters of Carthamus tinctorius L. (safflower) cv. Sina in soil contaminated with four levels Pb (0, 400, 800 and 1200 mg Pb/kg soil) were investigated. By increasing Pb concentration in the soil, chlorophyll a, chlorophyll b and carotenoids contents were significantly reduced, whereas proline amounts of leaf was increased. Inoculation of safflower plants with P. indica yielded in greater chlorophyll a, chlorophyll b, carotenoids and proline contents, in compare to non-inoculated plants. The results from this work showed that endophyte fungus P. indica has an important role in the alleviation of Pb toxicity via increased contents of photosynthetic pigments and proline in the leaves of safflower cv. Sina.

Keywords: Lead, Heavy metal stress; Safflower, Photosynthesis pigments

Identification of the best fruit collection height for the restoration of wild pistachio in Golestan province

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In the Northeastern of Golestan province (Maraveh Tappeh city), the wild pistachio species (P. atlantica) due to unconventional usage is on the slope of extinction and destruction, so the restoration of this species is truly essential. The fruit was collected separately from 11 trees in two range of altitudes (below and above 011 meters above sea level), quantitative traits (healthy and empty) and qualitative traits (length, width, and depth) of seeds were measured and recorded. Quantitative Data Analysis shows a significant difference between the two altitude ranges, so that trees located below 011 meters, produce more healthy fruit and less empty ones. In the two altitudes, the fruit didn’t show any significant differences in terms of length, width, depth, also in weight (with fleshy and wooden shell), significant differences could be absorbed. Finally, the weight of fleshy and wooden shell and the fruit kernel were compared and results showed that the main cause of these significant differences was the fruit kernel. The findings of this study showed that for the restoration of wild pistachio, the fruit should be collected from trees at an altitude of 011 meter above sea level upwards so that the chance of production and establishment of seedling increases.

Keywords: Maraveh Tappeh, Wild pistachio, Plant systematic, Northeastern Golestan, Biodiversity
A survey of Osteopontin and Osteocalcin genes expression in osteoblasts derived from Wharton’s jelly mesenchymal stem cells of human umbilical cord by licorice (Glycyrrhiza glabra L.) root extract

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Osteoporosis or silent disease is one of the most prevalent diseases in elderly menopause women in current century. Estrogen plays an important role in osteogenesis and prevention of bone Fractures. Hormone replacement therapy can increase the risk of breast and ovary cancers and cardiovascular disease. Thus, there is an increasing demand for replacement with plant phytoestrogens. The aim of this study is focused on determining the effects of aqueous extract of licorice root on osteogenesis progress in Wharton’s jelly mesenchymal stem cells (WJMSCs). WJMSCs were isolated from human umbilical cord matrix and treated with different amount of licorice root extract. Extract cytotoxity was measured using MTT assay. The Alizarin red staining and RT-PCR were performed to evaluate the differentiation progress. MTT Assay results showed that a number of concentration of licorice extract such as 600 μg/ml (cell viability values were around 85%) and two lower doses (500 and 400 μg/ml) are non-toxic for osteogenic differentiation. The maximum morphological changes and extracellular matrix staining take place in 600 μg/ml doses of plant extract. RT-PCR result demonstrates in 14 days the groups that treated with licorice extract express osteocalcin and osteopontin genes. Based on the results obtained in this study, it can be concluded that licorice root has positive effects on bone proliferation and differentiation in human mesenchymal stem cells.

Keywords: Extract, Licorice, Wharton’s jelly, Stem cell, Osteogenesis

A survey of alpha-crystalline a gene expression in eye lens fiber cells derived from Wharton’s Jelly mesenchymal stem cells in 3D culture by vitreous fluid

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Wharton’s Jelly Mesenchymal Stem Cells (WJMSCs) are a potential renewable source of cells in replacement therapies of many diseases. Different biomaterials have been used as a scaffold to mimic the stem cell niche, which is important for promoting cellular interactions, cell proliferation, and differentiation. Encapsulation involves entrapment of living cells within the semipermeable membrane for the exchange of nutrients, oxygen and stimuli, whereas antibodies and host immune cells are kept out. The aim of this research was to study the potency of Alginate biopolymer as a microenvironment for the differentiation of human Wharton’s Jelly-derived mesenchymal stem cells (hWJMSCs) to eye lens fiber cells. For this purpose, hWJMSCs were isolated from the human umbilical cord and they were encapsulated and cultured in medium contains different ratios (1:3, 1:1 and 3:1) of vitreous fluid. After 14 days, RT-PCR was performed to investigating the expression of alpha-crystalline A gene. The encapsulation procedure did not alter the morphology of the encapsulated cells. Post-differentiation analysis confirmed the expression of Alpha-crystalline A as eye lens fiber specific marker. Alginate has potential to be used as a three-dimensional (3D) scaffold for culturing and differentiation of WJMSCs to eye lens fiber cells.

Keywords: Alginate, Vitreous, Wharton’s jelly mesenchymal stem cells, Alpha-crystalline
Idarubicin was eight 5-8 times more potent than daunorubicin in acute myeloid leukemia (AML) treatment and have a similar mechanism of action with it. Wilms tumor 1 (WT1) has long been overexpressed in AML. Lately, the formation of G-quadruplexes in oncogenic promoters like WT1 has been widely investigated since stabilization these structures leads to transcriptional inhibition of the oncogene. The aim of this study was to understand the effect of Idarubicin on formation of G-quadruplex motifs in WT1 promoter oligonucleotides. Differential pulse voltammetry (DPV), circular dichroism (CD), polyacrylamide gel electrophoresis, electrophoretic mobility shifts assay (EMSA) and polymerase chain reaction (PCR) were performed in order to study the nature of interactions between idarubicin and WT1 promoter region. Electrochemical characterizations of WT1 promoter motif showed that with increased drug concentration, secondary structures formation in WT1 synthetic oligomer. CD measurements revealed parallel G-quadruplex structure with a major positive signal at 260 nm and a minor negative signal at 240 nm. EMSA assay showed that the high-mobility bands were identified as compact intramolecular G-quadruplex structures. PCR stop assay were performed to further demonstrate that inhibition of PCR induced due to G-quadruplex stabilization in the oligomer by idarubicin. Data clearly revealed that idarubicin had potential to stabilize G-quadruplexes in WT21 oligomer. Accordingly, the WT1 promoter can form stable intramolecular parallel G-quadruplexes. Consequently, we hypothesized ligand-mediated stabilization of G-quadruplexes within the WT1 promoter could have silencing effect on WT1 expression. Keywords: Idarubicin, Promoter G-quadruplex structure, Targeted leukemia therapy, WT1
Immunophenotypical characterization of zebrafish heart-derived mesenchymal stem cells

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Mesenchymal stem cells (MSCs) have been considered as an appropriate source for cellular therapy, tissue engineering, and regenerative medicine. The effectiveness of MSCs in the treatment of wide ranges of disease including hematopoietic recovery, inflammatory diseases, bone regeneration, infarcted myocardium, joint diseases etc. has been well documented; for this reason, isolation and characterization of MSCs from different species for extensive preclinical studies are required. Also, to clarify different aspects of MSCs biology and develop MSCs applications in human diseases treatment, other studies on animal models are required. However, the similarity between zebrafish and human genome has been further examined; zebrafish is considered to be the best for practical purposes in regenerative medicine. The aim of this study was to investigate the immunophenotypical characterization of heart-derived-MSCs for the first time for using zebrafish as a suitable animal model for regenerative medicine in cell therapy. In the present study, MSCs were isolated enzymatically from heart tissue of Zebrafish (Danio rerio) as previously reported in other papers by Fathi et al. (2017). The flow cytometry, as well as RT-PCR, were used to analyze the expression of a panel of immunological cell surface markers CD44, CD90, CD31, and CD34. In the immunophenotyping analysis, the heart-derived-MSCs was found to be immunopositive for mesenchymal stem cell markers (CD44 and CD90), and immunonegative for hematopoietic stem cell markers (CD31 and CD34). These results are, in combination with the RT-PCR results, were also positive and negative for the gene expression of CD44 and CD90 and CD31 and CD34, respectively. These findings are in line with the results of other MSCs; these reactions are generally characteristic of MSCs. Our results show that heart tissue-derived MSCs exhibited expression of a typical set of classic MSC surface markers and lack of hematopoietic marker, which will contribute to a more extensive characterization of zebrafish MSCs prior to their use in cell therapy.

Keywords: Zebrafish (Danio rerio), Immunophenotypical characterization, Immunological cell surface marker

Rat bone marrow-derived mesenchymal stem cells can potentially decrease the hTERT gene expression in chronic myeloid leukemia cell line

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Chronic myeloid leukemia (CML) is a hematopoietic stem cell-derived and progenitor-driven myeloproliferative disorder that may rise from a clinically manageable chronic phase (CP) to an incurable blastic phase. As is the case in all cancers, telomerase activity and hTERT gene expression play an important role in the progression of CML. A significant increase in hTERT gene expression has been reported in each of the three phases of CML, and this enhancement is accentuated during the progression of the disease. The multi-lineage nature of CML suggests that stem cells are important targets of this disease. Therefore, the aim of this study is to investigate the effect of BMSCs on the hTERT gene expression of the co-cultured K562 cell line. In this study, the bone marrow of the adult Rattus norvegicus was collected, mononuclear cells were separated by Ficoll Hypaque and bone marrow-derived mesenchymal stem cells (BMSCs) were isolated. In the following, the flow cytometry method, as well as RT-PCR, were performed to investigate the MSCs-surface markers. On the other word, K562 as chronic myeloid leukemia cell line were cultured in RPMI/1640. After reaching a confluency of cells, BMSCs co-cultured with K562 cell line for 7 days (1:10). At the end of co-culture time, K562 cell line was collected, total RNA was isolated, and cDNA was synthesized and subjected to Real-time PCR for investigating the hTERT gene expression. It was found that BMSCs had high levels of expression of CD44 (94.5%) and CD90 (87.1%) and hematopoietic cell lineage-specific antigens, such as CD31 (0.07%), and CD56 (0.9%) were not expressed in these cells. In addition, quantitative real-time PCR showed that BMSCs cause to significantly decrease the gene expression of hTERT. Taken together, the data showed that bone marrow-derived mesenchymal stem cells cause to decrease the hTERT gene expression of K562 cell line as a therapeutic approach.

Keywords: hTERT gene expression, Bone marrow-derived mesenchymal stem cell, Real-time PCR
Comparative expression of Notch receptors in hESC- retinal progenitor cells and neural tube

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Notch signaling pathway is activated by direct interactions with ligand-expressing cells. During neurogenesis, Notch activity is required to maintain the progenitor state of cells. In fact, Notch signaling regulates proliferation and differentiation balance by symmetric and asymmetric cell division. Taking into consideration that the RPCs progeny is very important during retinal development and there are no much experimental evidence for the expression level of Notch receptors in hRPCs. In this study, the Notch receptors expression was analyzed in RPCs derived from hESCs during in vitro maintenance. So in the current study, RPCs were derived from hESC and the expression of Notch receptors was analyzed during in vitro maintenance. The hESC line (RH6, passage 39-50) and Stem cells of apical papilla (SCAP) (passage 6-10) were cultured as previously described. In order to differentiate hESCs to RPCs, we used SCAPs as an induction factor in a co-culture system. Following one month, neural tube-like structures (NT) appeared. They were mechanically isolated and passaged on matrigel coated plates and were monitored until passage 3. To assess Notch receptors expression, total RNA was isolated from two stages NT and RPCs (P3). The quantitative PCR was performed using specific primers. We investigated whether the hESC-RPC exhibited expression of Notch 1 and Notch 2, NTs and RPCs expressed Notch 1 greatly more than Notch 2. Although, the expression of Notch 1 and Notch 2 was reduced from NTs to RPCs. In this study, the expression of Notch 1 and Notch 2 by RPCs and NTs differentiated from hESCs was evaluated and they both had higher expression of NTs. It is worth mentioning that these results might be dependent on cell niche and maintenance culture conditions which may affect the progeny level of RPCs.

Keywords: Notch receptor, Human embryonic stem cell, Retinal progenitor cell, Neural tube-like structure

Cloning of human high-temperature requirement A2 (hu-HtrA2) gene in a prokaryotic host

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HtrA2/omi is a mitochondrial serine protease which is induced by the stress and encoded by the nucleus. This protease protects the cell survival by maintaining the mitochondrial homeostasis. The HtrA2/omi has a role in cell death caused by apoptosis, necrosis, and anoikis. Disordered HtrA2/omi appears in neurological diseases, cancer and arthritis. For this reason, studying of the structure and function of this enzyme can be a way of preventing and treating diseases and disorders caused by this enzyme. In this study, the HtrA2/omi gene was cloned in pET21a (+) (expression vector). For cloning into pET21a (+) one should clone a target gene between the NdeI (forward primer) and the XhoI (reverse primer) sites in pET21a (+). After PCR, the PCR product was digested by NdeI and XhoI restriction enzymes and then ligated to the vector. Finally, E.coli TOP 10 strain cells were transformed by the ligation product and incubated at 37°C for 16h. After observation of colonies, some colonies were randomly selected and prepared for colony PCR. The results of colony PCR have confirmed the cloning of HtrA2/omi in pET21a (+). Also, the recombinant vectors were extracted for sequencing. The expression and purification of the recombinant protein is underway.

Keywords: PCR, Cloning, HtrA2/omi, pET21a (+)
The nutritional role of *Spirulina platensis* in the proliferation of stem cells

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The *Spirulina platensis* microalgae are a type of cyanobacteria. Since *S. platensis* contains 51-71% of proteins, most of the necessary amino acids for the human body and about 7% of different kinds of lipids and vitamins, it has a vast some of the use in agriculture and aviculture. The effect of some probiotic bacteria, which are similar to *Spirulina platensis* in nutritional elements, on the proliferation of endothelial blood cell, fibroblasts, inflammatory cells have been proven in past studies. Mesenchymal stem cells have a large application in bone and cartilage graft and repair. However, the ability of proliferation of these cells is less; they go to undesirable differentiation after some consecutive passages. Therefore, we used *Spirulina* as an element to supply the growth factor enhancing mesenchymal stem cell’s ability to proliferate. First, the stromal cells from Rat’s bone marrow were extracted. In the second passage, they were proven as the mesenchymal cell by the use of adipogenesis and osteogenesis differentiation medium and staining. After *S. platensis*’s culture in standard situations, we purified its extract. Then the cell treatments were done with 7 logarithmic concentrations (0, 0.1, 0.3, 0.9, 3.0, 9.0, 30 µl/ml) from *S. platensis*’s extract. MTT assay was done to determine the number of living cells, in the next step. Results were analyzed by SPSS. Results obtained from the culture in defined media showed that they differentiated to osteocytes and adipocytes lineage in differentiated media. This evidence proves the existence of mesenchymal stem cells. Statistical analyses showed a significant increase in MSCs proliferation. The maximum level of increased proliferation was in 0.9 µl/ml concentration. Since *Spirulina* secrets nutritional elements, its extract can be used as growth factors for enhancing the proliferation of mesenchymal stem cells. In addition, optimum concentration for cell treatment with *Spirulina*’s extract was found. Therefore purified material from *Spirulina*’s protein can be used for better results in treatment with mesenchymal stem cells.

**Keywords:** *Spirulina platensis*, Rat bone marrow mesenchymal stem cells, Bone marrow stromal cells, Cell proliferation

Cytotoxic effects of iron oxide nanoparticles on Hep G2 cells

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Iron oxide nanoparticles (Fe₂O₃-NPs) are being used in an increasing number of fields such as molecular imaging in the context of Magnetic Resonance Imaging, ultrasound, optical imaging, and X-ray imaging. This widespread application of different sized of Fe₂O₃-NP in the biomedical field raises concerns over their increased exposing to tissues and organs of the human and animals. This study investigated the molecular and cytotoxic effects of Fe₂O₃-NP (50 nm) on Hep G2 cells. Nanoparticles are containing high surface area to volume ratio that converted them to reactive molecules; therefore we evaluated oxidative stress biomarkers such as reactive oxygen species (ROS), lactate dehydrogenase leakage (LDH) and cell death by standard biochemical methods. Assessment of ROS content in a different concentration of Fe₂O₃-NP approved increased ROS level (up to 3.2 folds) concentration-dependent manner. ROS overproduction is accompanied by LDH leakage from the cells (4 folds than control) possibly caused by membrane damages in the presence of toxicant nanoparticle. Mentioned molecular effects increase cell death by more than 80% in a dose-dependent manner that evaluated by MTT assay. Possibly due to the small size of nanoparticle which allows fast and easy entry into the cells and increased ROS overproduction that attack to the membrane and intracellular organelles, the viability of cells significantly decreased in the presence of nanoparticle.

**Keywords:** Nanoparticles, Reactive oxygen species, Lactate dehydrogenase leakage, Membrane injury, Apoptosis
Ankylosing spondylitis (AS) is the prototypic member of a group of inflammatory rheumatic diseases named spondyloarthritides (SpA). The disease is characterized by the inflammatory type of low back pain and progressive spinal stiffness. Macrophages play an important role in the AS auto-inflammatory responses and fibrocartilage destruction. Adenosine triphosphate (ATP) and other nucleotides, released from cells, regulate several immune and inflammatory responses in macrophages by interaction with P2X and P2Y purinergic receptors. In this study, we investigated the association of the clinical manifestation of AS patients with purinergic receptors mRNA expressions. Monocytes were isolated from 23 active AS patients and differentiated to macrophages by a macrophage-colony stimulating factor (M-CSF) for 7 days. The expression level of P2X (P2RX1,2,7) and P2Y (P2RY1,2,4,6,11,12,13,14) receptors mRNA was assessed by SYBR Green real-time PCR. We investigated the association of the clinical characteristics of AS patients including Bath Ankylosing Spondylitis Metrology Index (BASMI), Bath Ankylosing Spondylitis Functional Index (BASFI) and Bath Ankylosing Spondylitis Disease Activity Index (BASDI) with receptors mRNA expressions. The results showed an inverse correlation between P2RX4 and P2RX7 mRNA expression and BASDI score in AS patients (P<0.05). It was also investigated that P2RY2 mRNA expression is positively correlated with patients BASMI score (P<0.01). Our results suggested that AS patient’s clinical manifestation is associated by purinergic receptors expression in macrophages. Therefor modulating the purinergic signaling receptors expression could be used as a therapeutic approach for AS treatment.

**Keywords:** Purinergic receptor, Macrophage, Ankylosing spondylitis

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The protective effect of astaxanthin on sperm DNA fragmentation in cadmium-induced damage in mice

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Cadmium, a common environmental heavy metal, can damage lung, liver, kidney, and testis. Cadmium toxicity can decrease sperm quality and function. The antioxidants can improve sperm parameters and decrease effective factors in male infertility. In this study, we investigated the effect of astaxanthin on sperm DNA fragmentation and sperm viability in presence of cadmium in mice. 30 adults male mice were divided into 5 groups, control group didn’t receive treatment, sham group intraperitoneally received olive oil and salt as a solvent for astaxanthin and cadmium respectively, Cadmium group received cadmium at a dose of 1 mg/kg, a group received astaxanthin at a dose of 10 mg / kg of animal weight and cadmium at a dose of 1 mg / kg, a group received astaxanthin at a dose of 10 mg / kg. Mice received cadmium, astaxanthin, and solvents intraperitoneally for 14 days. Sperm DNA fragmentation (SDF) and sperm viability were evaluated by the sperm DNA fragmentation kit and eosin-nigrosin staining respectively. The results didn’t indicate a significant change in the sperm viability between control and sham groups. Sperm viability significantly decreased by cadmium compared to the sham group. Astaxanthin in presence of cadmium significantly increased sperm viability in comparison with cadmium group, also astaxanthin significantly increased sperm viability compared to sham group. On the other hand, Cadmium significantly increased Sperm DNA fragmentation comparison with sham group, besides astaxanthin significantly decreased sperm DNA fragmentation induced by cadmium compared to cadmium group. Also, astaxanthin significantly decreased sperm DNA fragmentation in comparison with the sham group. Our finding indicated astaxanthin can protect sperm DNA against cadmium-induced DNA fragmentation and also astaxanthin can increase sperm viability in presence of cadmium.

**Keywords:** Astaxanthin, Cadmium, DNA fragmentation, Sperm viability

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The correlation of BATH indices scores and purinergic receptors expression in macrophages from patients with ankylosing spondylitis

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Cadmium, a common environmental heavy metal, can damage lung, liver, kidney, and testis. Cadmium toxicity can decrease sperm quality and function. The antioxidants can improve sperm parameters and decrease effective factors in male infertility. In this study, we investigated the effect of astaxanthin on sperm DNA fragmentation and sperm viability in presence of cadmium in mice. 30 adults male mice were divided into 5 groups, control group didn’t receive treatment, sham group intraperitoneally received olive oil and salt as a solvent for astaxanthin and cadmium respectively, Cadmium group received cadmium at a dose of 1 mg/kg, a group received astaxanthin at a dose of 10 mg / kg of animal weight and cadmium at a dose of 1 mg / kg, a group received astaxanthin at a dose of 10 mg / kg. Mice received cadmium, astaxanthin, and solvents intraperitoneally for 14 days. Sperm DNA fragmentation (SDF) and sperm viability were evaluated by the sperm DNA fragmentation kit and eosin-nigrosin staining respectively. The results didn’t indicate a significant change in the sperm viability between control and sham groups. Sperm viability significantly decreased by cadmium compared to the sham group. Astaxanthin in presence of cadmium significantly increased sperm viability in comparison with cadmium group, also astaxanthin significantly increased sperm viability compared to sham group. On the other hand, Cadmium significantly increased Sperm DNA fragmentation comparison with sham group, besides astaxanthin significantly decreased sperm DNA fragmentation induced by cadmium compared to cadmium group. Also, astaxanthin significantly decreased sperm DNA fragmentation in comparison with the sham group. Our finding indicated astaxanthin can protect sperm DNA against cadmium-induced DNA fragmentation and also astaxanthin can increase sperm viability in presence of cadmium.

**Keywords:** Astaxanthin, Cadmium, DNA fragmentation, Sperm viability
Effect of olive-fruit Roghani cultivar extract on the aggregation and cytotoxicity of alpha-synuclein in Parkinson's disease

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The neurodegenerative diseases are dependent on the formation of amyloidal fibrils of proteins/peptides without any factual treatment. The α-synuclein (α-Syn) is an abundant protein in the brain which its toxic fibrillar aggregates have a key role in diseases called synucleinopathies like Parkinson’s disease. Olives could be candidates to inhibit the fibrillization. The populations with Mediterranean diet regime that contains a high level of olive products are more health and longevity.

In this study, the extracts of olive fruits of some Iranian olive cultivars including Zard, Mary, Arghavan, and Roghani were investigated on the fibrillization of α-Syn. Among the assessed extracts, Roghani extract showed significant effects on the rate of formation and the levels of end product fibrils. Structural characterization using circular dichroism indicated that in the presence of the extract, the protein strongly resisted the formation of beta sheets. TEM results confirmed that unlike control with extended fibrils after 24 h incubation, the treated protein did not proceed to fibrillization. Treating OLN-93 with the aggregated α-Syn indicated that the viability of cells considerably increased in the presence of the extract. Given that other extracts did not show these obvious effects whether on α-Syn fibrillization and cytotoxicity and moreover since Roghani is an Iranian olive cultivar with high consumption, more investigation is needed to elucidate its high therapeutic potential including using different fruits which are harvested in a different period or using other parts of the tree. These studies may provide valuable information for therapeutic and nutrition field.

Keywords: α-synuclein, Olive fruits extract, Parkinson’s disease, Roghani cultivar

Investigation of RKIP tumor suppressor gene expression in triple-negative breast cancer cells under microgravity conditions

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Microgravity is considered to be the main source of space flight shock that affects cells and tissues and has an adverse effect on the health of astronauts. Changes in gravity as experienced by astronauts during space travel lead to changes in the physiology and even the morphology of the cells in the culture medium. In some studies, it has been observed that changes in gravity reduce the growth of some cancer cells and alter the expression of some genes and cellular functions. RKIP protein is a modulator of kinase activity and a cell balancing agent that also acts as a metastatic inhibitor in a variety of solid tumors, including breast cancer. RKIP expression in advanced tumors is reduced and its increase reduces the invasive potential of cancer cells without affecting their growth. In this study, we have cultivated triple-negative MDA-MB-231 breast cancer cell line on a clinostat device in order to the investigation of the RKIP gene expression changes under simulated microgravity conditions. Our results indicate that weightlessness changes the expression levels of RKIP gene. In general, our findings would have created a new light on the effects of microgravity on cell biology and human health.

Keywords: microgravity, RKIP protein, MDA-MB-231 cell line
Investigation of TIM3 and PDCD1 genes role in the asthma treatment by the Zataria multiflora Boiss and carvacrol

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Numerous medicinal and traditional uses for Zataria multiflora and its constituent, carvacrol, have been described such as antispasmodic, anti-inflammatory, and antioxidant effects. In this study, the effects of the hydroalcoholic extract of Z. multiflora and carvacrol by the PDCD1 and TIM3 pathways in moderate asthmatic disease were evaluated. PDCD1 is a cell surface membrane protein of the immunoglobulin superfamily and TIM3 is a Th1-specific cell surface protein that regulates macrophage activation. 48 asthmatic patients were selected according to GINA guidelines and divided into four groups consisted of the placebo before the treatment (group 1), placebo after the treatment (group 2), the Z. multiflora extract before treatment (5 and 10 mg/kg/day, Z.5 and Z.10 respectively) in the group 3, and the Z. multiflora extract after treatment (5 and 10 mg/kg/day, Z.5 and Z.10 respectively) in the group 4, (n =12 for each group). Patients in all groups received medication three times a day for two months. Gene expression analysis was performed by the probe-based quantitative PCR technique on the total RNA of the peripheral blood mononuclear cells. Using the carvacrol treatment without Z. multiflora extract caused 29% increase in TIM3 expression. On the other hand, this treatment reduces the expression of PDCD1 to 71% of the control level. According to these results, it seems the TIM3 gene as an immune response inhibitor interfering with the asthma symptoms treatment by the carvacrol. Using the Z. multiflora extract along with the carvacrol has no significant effect on the TIM3 expression. On the contrary, the Z. multiflora extract increases the expression of the PDCD1 gene by 46%. This supports the immune response inhibitory role of the TIM3 in the asthma symptoms treatment by the carvacrol. The Z. multiflora extract, alone, has no effect on the TIM3 expression but it decreases the PDCD1 expression to the half. The results of this study showed that the Z. multiflora extract and carvacrol during two months treatment lead to TIM3 and PDCD1 expression changes which effects on the immune regulatory pathways.

Keywords: Asthma, PDCD1, TIM3, Respiratory Inflammation, Zataria multiflora, Carvacrol

Investigation of the preventive effect of Rivastigmine and aqueous extract of Olibanum in Alzheimer’s disease

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Alzheimer’s disease (AD) is a multifactorial disorder that its progression is associated with many genetic and environmental factors. Many studies showed that the FMR1 gene is important in the AD because FMR1 protein is participated in regulating of APP production. One of the most common plants to improve memory performance is the Boswellia resinate. It seems that the resin of Boswellia considerably can affect brain development and correct formation of neuritis (axon and dendrites). So, we investigated the preventive effects of aqueous extract of Olibanum and Rivastigmine on the expression of the FMR1 gene in rats that treated with AICl3. 28 adult male rats randomly divided into 4 groups (n=7). Group 1 was treated with distilled water (1 ml), group 2 with AICl3 (0.3 mg/kg), group 3 with aqueous extract of Olibanum (200 mg/kg), Group 4 was treated with Rivastigmine (0.3 mg/kg), all the Group treated with our own substance for 14 days Then treated with AICl3 (20 mg/kg) for 56 days (Except control Group). Animals were tested with Morris-Water-Maze and their hippocampus was isolated and used for FMR1 gene expression analysis by Real-time PCR. Total RNA was extracted by using RNX-Plus solution according to the manufacturer’s protocol. Then, cDNA was synthesized by PrimeScript™ RT reagent Kit (TaKaRa). GAPDH was used as an internal control gene. Morris-Water-Maze test showed that AICl3 impairs memory performance in rats and Real-time PCR data analysis indicated that while AICl3 led to significant reduction in FMR1 gene expression (0.018), Rivastigmine increased its expression (0.0675) but an aqueous extract of Olibanum no significant effect (0.021).

Keywords: Rivastigmine, Olibanum, FMR1, Alzheimer’s disease
Application of medicinal herbs of the west of Golestan province in women diseases

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Ethnobotany is a major economic opportunity of plant science. This study is referred to the people who live in Golestan provinces and have valuable experiences in medicinal plants especially Turkman tribes, in the east of Golestan. In a current study first experienced- people were referred. For Application of medicinal plants, second. All information’s were recorded in the forms, third analyzing of data based on curing of women diseases were made. According to this research, totally 50 medicinal plants were recognized. 11 out of 50 species were considered as medicinal plants for curing, *Urtica dioica* L. (Urticaceae), *Anthemis nobilis* Boiss. (Asteraceae), *Tragopogon persicus* Boiss (Asteraceae), *Peganum harmala* L. (Zygophyllaceae), *Mentha longifolia* (Hud) (Labiatae). Application of medicinal plants was presented for example kidney patients, dryness, constipation, infection, infertility, urinary tract infection.  
**Keywords:** Ethnobotany, Infertility, *Anthemis nobilis*

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Effects of *Cucumis melo* L. seed on seasonal allergy treatment

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*Cucumis melo* (Cucurbitaceae) family widely were cultured in Khorasan Province, Iran. Melo seeds and periphery layer (MSPL) of that is full of protein and amino acids, fatty acids, omega-3s, vitamins B and E, folic acid, caffeic acid and salts such as magnesium, iron, zinc, copper, and potassium, however, this part of Melo discard as a waste product. Based those and as a source of antioxidants and triterpenoids, MSPL with warm and dry (Yellow bile) humor are effective in treating kidney diseases (i.e. kidney stones), immune responses, skin allergies (i.e. eczema), and in scavenging free radicals. Consequently, it is expected MSPL be also effective in treating and reducing the symptoms of seasonal allergies. To examine that, Cucumis melons were prepared (from Torbat Heydarieh region), MSPL were dried and powdered. 40 volunteers (20 male, 20 females) divided into 4 groups including 10 persons for each control or treatment group. Each person used 2.6g powder every 42 hours for 30 days from 16 March to 14 April (times that allergy symptoms are arising). Pea flour was used as a control. Volunteers filled the questionnaire in 2 steps (before and after using powder). The results were analyzed by the Wilcoxon signed-rank test in SPSS. Our results showed MSPL has a considerable impact on reducing symptoms of seasonal allergies that occur on the eye, nose, throat, skin, and ears, respectively. Interestingly, MSPL has no effect on seasonal sensitivity-related coughing. These findings could be applicable to seasonal allergies treatment.  
**Keywords:** *Cucumis Melo*, Melon seed, Seasonal allergy, Herbal treatment
Evaluation of Thiazole-derivatives cytotoxicity effects on breast cancer line (MCF-7) and human non-small-cell lung cancer (A549)

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Cancer is a group of diseases characterized by the uncontrolled cell growth and division, the second leading cause of death worldwide and among more than 100 types of cancer, breast cancer is leading cancer diagnosed in women and lung cancer is one of the most common cancers in men. Due to the progression of cancer, drug resistance, and side effects of current medications, researchers are looking for new synthetic drugs with strong effects on cancer treatments and fewer side effects on cancer patients. In this study, a series of synthetic compound Thiazole derivatives with anticancer activity (8 samples that named C1-C8) were tested. In this regards, the cell viability effect of 8 samples of Thiazole derivatives in different concentrations of 100µm to 1000µm with a three-time repeat count on breast cancer line MCF-7 and Human non-small-cell lung cancer A549 cancer cell line have been investigated. Etoposide as a positive control and DMSO as negative control have been used to determine the accuracy of the work. The effect of compounds treatment on the viability of MCF-7 and A549 cells were measured by 3-(4,5-dimethylthiazol-2-y)-2,5-diphenyl tetrazolium bromide (MTT). MTT assay was used for the assay to measure the toxicity of the drugs. Our results showed that in MCF-7 cells, among the 8 available samples, IC50 was found in sample C7 at a concentration of 300µm and in A549 cells, IC50 was found in samples C1 at a concentration of 400µm, our results could provide more insights and shed further light on the in cancer therapy approach.

Keywords: Cytotoxicity, MTT assay, Synthetic anticancer drugs

The effectiveness of Carla's herbal remedy in the prevention and treatment of breast cancer

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Breast cancer is one of the most important causes of mortality among women that threatens the health of many women with different ages worldwide. Common treatments of this disease such as chemotherapy have produced side effects by using chemical drugs, which is why scientists always seek alternative methods to treat or slow down the growth of this disease. The use of herbal medicines has been commonplace since ancient times, and there is now a new attitude towards the use of these natural substances based on the history of traditional medicine. In this research, the effect of Carla's fruit extract from Cucurbitaceae family on the survival of MCF-7 breast cancer cells and the expression of Caspase 3 gene expression by real-time gene expression (qRT-PCR) were investigated. For this purpose, the cells were treated with different concentrations of Carla fruit extract (25, 50, 75, 100, 150, 200, 12.5 ppm) for 24, 48 and 72 hours and kept under sterile conditions for different intervals. The results of the study demonstrated that with increasing concentration and time, the vitality of the cells had a significant decrease compared to the control samples. At 100 ppm concentration, 50% of the cells (IC50 were loosed. Also, the results of gene expression analysis indicated that caspase 3 expression in 24 hours significantly differs from the control group, but there was no significant difference by other time intervals (48 and 72 hours) in comparison with 24 hours of treatment. According to the data obtained, it can be concluded that Carla can be used as a useful herbal medicine for cancer treatment.

Keywords: Breast cancer, Carla, Gene expression, IC50
Designing the metal catalyzed oxidation system for comparison of injury effects of aluminium, copper, iron and lead on blood proteins

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Industrial metals induce various toxicities such as cytotoxicity, genotoxicity neurotoxicity, hepatotoxicity and hematotoxicity. This study focused on the oxidative effects of aluminium (Al), copper (Cu), iron (Fe) and lead (Pb) ions on blood proteins. Blood samples were collected from healthy volunteers. Age, weight and sex were determined as demographic parameters. This system was designed for simulating the in vivo effects of different doses of metals in controlled laboratory conditions. Blood samples were incubated in isotonic phosphate buffer pH 7.4 and different doses of Al, Cu, Fe and Pb ions at 37 °C with shaking under aerobic condition. Carbonyl content of proteins (PCO) were calculated as oxidative marker. For this purpose, dinitrophenyl hydrazine was used for reaction with carbonyl groups and absorbance of solutions at 280 nm for protein determination. Concentrations of oxy-Hb, metHb and hemichrome were calculated according to Hb absorbance at 560, 577 and 630 nm. The data was analyzed using Excel and SPSS software. Average and standard deviation were determined as statistical parameters, t-student for comparison of groups and correlation test and regression analysis for studying the effects of different doses of metals on biochemical parameters. Volunteers included 45 healthy men between the ages of 19 and 21, weighing between 70 and 90 kilograms.

Results of this study showed a significant increase in carbonyl groups in plasma proteins from 5.8 to 10.5, 13.3, 14.2 and 8.6, after exposure to Al, Cu, Fe and Pb, respectively (p<0.05). Also a decrease in oxyHb and increase in metHb and hemichrome concentrations were observed after incubation with different doses of Al, Cu, Fe and Pb. Our results demonstrated the conformational and structural modifications of hemoglobin and plasma proteins during exposure to industrial metals. Also we presented a reproducible and reliable algorithm of experiments to evaluate the oxidative effects of toxic metals on plasma proteins.

Keywords: Aluminium, Copper, Iron, Lead, Metal catalyzed oxidation systems, Blood proteins

Exosomes derived from ovarian epithelial carcinoma cells regulate angiogenesis by modification of jak/stat3 pathway and SOCS5 expression

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Because ovarian cancer is a most fatal cancer among woman. Exosomes are intercellular carriers that are responsible for cellular communication in the tumor microenvironment they contain different materials such as miRNAs that can change the host cell's settings. jak/stat3 signaling pathway known as an important regulator of angiogenesis is inhibited by SOCS5. The purpose of this study is to determine whether ovarian-derived exosome can reduce the expression of socs5 in endothelial cells. And this decrease will increase the jak/stat3 signaling pathway and enhance the rate of angiogenesis. The exosomes were extracted from the SKOV3 cells medium by using ExoQuick kits. SEM and DLS were used to investigate in terms of size and morphology of exosomes. HUVECs were treated with exosomes (100 μg/ml) or vehicle control (PBS) for 24 and 48 hours were investigated. The PKH test was used to confirm the exosome absorption by the cell. The expression of the genes (jak1, stat3, SOCS5) was measured by qRT-PCR. SEM of purified exosome was displayed spherical and membrane-encapsulated particles with diameters ranging from 50 to 200 nm. a single bell-shaped size distribution with a peak at ~90 nm resulted from DLS. Also, exosome absorption was confirmed by Fluorescent microscope and PKH assay. The results showed that exosome derived from ovarian cancer can accelerate the formation of blood vessels, reducing the expression of SOCS5 inhibitor can accelerate the expression of the jack / stat3 pathways genes and rate of angiogenesis.

Keywords: Exosomes, Jak/stat3, SOCS5, Angiogenesis
Study of the differentiation of rat omentum stem cells to nerve cells using brain tissue extract of rat

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Stem cells have two typical feature self-renewal and potential of producing different cell lineages. Omentum stem cells could be induced to differentiate into neural cells under certain condition. The main goal of this present study is to neural differentiation of OSCs induced by using Rat brain extract. Omentum stem cells were isolated from peritoneum and brain extract from neonatal rat brain, then the cells were cocultured in DMEM and brain extract. After 3 passage OSCs were morphologically observed and processed for RT-PCR. The number of cells neural morphology increased at 1-2 weeks dramatically. The cells were labeled by neural markers including Map2 which assessed by immunocytochemistry. Our finding demonstrated that OSCs efficiently differentiate into the neural cell when cultured in brain extract.

Keywords: Omentum, Mesenchymal stem cells, Rat neonate brain extract, Neural cell differentiation

Gene co-expression network analysis of transcriptomics data of gastric cancer subtypes to explore subtype-specific significant biological processes

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Gastric cancer (GC) is one of the most prevalent cancers worldwide that has heterogeneous nature from pathological and molecular standpoints. So this is important to identify involved mechanisms in incidence and progression of GC. High throughput techniques allow studying molecular profiles of this disease. Understanding the gene expression regulation mechanism can be important to declare complex phenotypes and mainly the causes of cancers' occurrence. The key step in propelling GC therapy toward personalized medicine is a classification of tumors in different subtypes based on different characteristics. One of these classifications has been done by Asian Cancer Research Group who used gene expression data to make 4 molecular subtypes (MSI, MSS/EMT, MSS/Tp53+, MSS/Tp53−). Due to the huge number of variables in transcriptomics data analysis, one of the best solutions is using network-based models. Gene networks can be used to studying gene regulatory mechanisms. In gene co-expression networks, expression correlation of genes is used to construct a gene network. We have applied the weighted gene co-expression network to create co-expressed gene modules for GC subtypes. In the next step, functional annotation analysis using DAVID web-server was used to identify involved biological pathways for the explored modules. The important modules that involved in biological pathways including 8, 10, 10, 9 co-expressed modules were identified and presented for MSI, EMT, Tp53+, Tp53− subtypes respectively. The extracted important pathways for each module are presenting as new and useful strategies for targeted treatment of GC.

Keywords: Gastric cancer, Transcriptomics data, Weighted gene co-expression network analysis, Personalized medicine
A review on secondary metabolites and pharmacological effects of *Berberis* sp.

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*Berberis* sp. is a medicinal plant, native to northeastern Iran and used in traditional medicine for many disorders. Several studies have been conducted to detect its herbal components and therapeutic characteristics. This review article has introduced herbal components and assessed its valuable therapeutic properties. The information in this review article has been gathered from journals accessible in databases such as SID, MagIran, and Civilica. The search terms were *Berberis* and antioxidant activity. Several drug properties, such as antimicrobial, antifungal and anti-parasitic properties, are related to phenolic compounds of barberry. The most antioxidant activity of *Berberis* sp. extracts is due to alkaloids with an isoquinoline core such as Berberine, Oxycocanthine oxide, Berbamine, and Palmatine. *Berberis* sp. has therapeutic effects such as low blood cholesterol, high blood pressure, anti-inflammatory effects and antioxidant properties. Traditional applications of *Berberis* sp. in the treatment of many diseases and its valuable medicinal and herbal components could provide a context for scientists to develop plant-derived medications such as antibiotics, high cholesterol-lowering drugs, blood pressure treating drugs & joint pain medication and the key to conducting clinical trials.

**Keywords:** *Berberis* sp., Compounds, Pharmacological effect, Traditional medicine

Influence of zinc element on chemical composition of essential oil in *Satureja bakhtiariaca* at different phenological stages

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The genus *Satureja* belongs to the Lamiaceae family. *Satureja bakhtiariaca* essential oil contains high amounts of phenolic compounds and has high antioxidant capacity and also includes monoterpenes and sesquiterpenes. In order to evaluate the effect of zinc element, a greenhouse experiment was conducted in a completely randomized block design with three treatments and three replications. Treatments consisted of leaf spraying with zinc chelate (0, 200, 400 and 600 ppm). The main components of the aerial parts essential oil of the control plants in blooming belonged to Carvacrol (61.8%), β-Cymene (24.93%) and α-Terpinen (7.24%). The main components of the plant treated with zinc element at flowering stage in the concentration of 200 ppm, including Carvacrol (63%), p-Cymene (24.23%) and γ-Terpinen (7.34%): in 400 ppm containing β-Carvacrol (67.69%), Cymene (18.67%) & α-Terpene (3.44%) and in 600 ppm containing Carvacrol (56.88%), β-Cymen (28.42%) & α-Terpenein (5.23%). Compared to the control, in vegetative stage the levels of Carvacrol (57.9%), p-Cymene (29.62%) and γ-Terpinen (8.84%) significantly increased in plants treated with 200 ppm zinc concentration; the levels of Carvacrol (58.17%), β-Cymene (57.28%) and α-Terpinene (7.8%) significantly increased in plants treated with 400 ppm zinc concentration. The content of Carvacrol (55.04%), β-Cymen (28.42%) and α-Terpinene (10.03%) significantly increased in plants treated with 600 ppm zinc concentration. Compared to the control, an increase in content of carvacrol at flowering stage with all zinc treatments was observed. Content of p-Cymene and α-Terpinene has been increased at vegetative stage in plants treated with application of all zinc treatments; whereas, the content of carvacrol has been decreased.

**Keywords:** *Satureja bakhtiariaca*, Carvacrol, β-Cymene, Zn
A preliminary study on medicinal plants of Abhar County in the southeast of Zanjan province of Iran

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Medicinal Plants are great genetic reserves that the human has used them for a long time and compared to chemical drugs have fewer complications. Iran is a country with rich flora in the world. The diversity of climate, culture and rituals and fees in different regions of the country is considerable and the history of traditional and local use of wild plants for the treatment of diseases in this country is long. Abhar county located at the southeast of Zanjan province with plains, mountainous areas and altitudes and good water resources has enjoyed high plant diversity. In this research, after preliminary surveys and dialogue with local and experienced people, medicinal plant species have been collected during growth seasons in 2015-2016. Then, plant samples have been transferred to botany lab in the University of Zanjan and have been determined with valid taxonomic references. The results of this study showed that 71 medicinal plant species belonging to 29 plant families are present in this region. Most important plant medicinal families in this survey are Asteraceae, Lamiaceae, Rosaceae, Boraginaceae, Apiaceae, Brassicaceae, Fabaceae, Papaveraceae, Malvaceae and Plantaginaceae and most well-known plant genera are Salvia L., Stachys L., Ziziphus L., Teucrium L., Cichorium L., Achillea L., Plantago L., Malva L., Papaver L., Cotoneaster Medik., Descurainia *Webb & Berthel., Lepidium L., Glycyrrhiza L. and Echium L.. The most usages and medicinal-traditional properties of studied plants are Analgesic, antispasmodic, digestive and anxiety disorder, anti-flatulence, anti-nausea, antimicrobial, anti-diarrhea, wound healing, fever and appetizer and are consumed in a sore throat, inflammation, infection, kidney problems, blood purification, blood pressure and nerve strengthening.

Keywords: Abhar, Medicinal plants, Traditional prescribes

Effect of foliar application of putrescine on free proline, soluble and insoluble carbohydrates in spring safflower (Carthamus tinctorius L.) under water deficit

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Water deficit has negative effect on plant growth and development. Polyamines are growth regulator in plants and increased their tolerance to drought. In this research the effect of putrescine foliar application (40 and 60 µM) on leaf proline, shoot and root soluble and insoluble sugars contents in safflower plants under different levels of water supply (100% and 40% field capacity) was studied as factorial arrangement based on complete randomized block design with three replications. Interaction of water deficit and putrescine for proline content and soluble sugars was significant. Proline content (leaf) and soluble sugars (shoot, root) significantly decreased with 40 and 60 µM putrescine under water deficit. Putrescine as osmolyte and reactive oxygen species scavenger reduced production and accumulation of compatible osmolytes. In non-stressed plants, effect of putrescine on proline and soluble sugars was different related to concentration. Insoluble sugars increased with both concentration of putrescine in shoot and decreased in root. Chlorophyll content maintenance in stressed plants increases drought tolerance. Reducing proline in stressed plants treated by putrescine can be related to enhancing chlorophyll content. Since proline and chlorophyll were synthesized from the same precursor (glutamate). Also increasing antioxidant enzymes activities in these plants can improve plant resistance. Decreasing soluble sugars with putrescine might be attributed to better water status of polyamine treated plants. Putrescine via increasing chlorophyll, relative water contents and antioxidant enzymes activities can enhance safflower resistance to drought without increasing osmolytes biosynthesis.

Keywords: Drought stress, Putrescine, Safflower, Osmolytes
Evaluation of taxol content of hazelnut leaf in the north-west of Iran

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Taxol is one of the most effective anti-cancer drug available in the market. The increasing demand of this compound due to its use in treating a wide range of cancers, as well as its high cost have been triggered efforts to find alternative resources to get this drug. Hazel (Corylus avellana), which is already cultivated for nutritional aspects, is now attracting attention for its phytochemical content. Notably, the discovery of taxol and related taxanes in this plant species prompted extensive interest to explore the production of these compounds in C. avellana. In this research, leaf samples from different populations collected from the Fandoglo forest (Ardabil province), Meshginshahr (Ardabil province) and Tarom gardens (Zanjan province). In these three regions, 18 genotypes were evaluated. Extraction of Taxol was done using the Bestoso et al. (2006) method and Taxol content was measured by HPLC. The results indicated that there are significant differences between genotypes in terms of taxol content. So, the amount of taxol was varied from 3.82 ppm in the genotype 9 (Meshginshahr) to 0.33 ppm in genotype 14 (Ardabil). In addition, comparing the genotypes collected from three different regions showed that the mean taxol content of Meshginshahr, Ardabil and Zanjan provinces were 2.17, 1.36 and 1.16, respectively.

**Keywords:** Corylus avellana, Genetic variability, Taxol

Effect of two species of mycorrhizal fungi on growth characteristics and essential oils of peppermint (Mentha piperita L.) under salt stress

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In order to investigate the effect of two species of mycorrhizal fungi to reducing the salinity damage on peppermint (Mentha piperita L.), a pot experiment was carried out in research greenhouse of the Payame Noor University, Lorestan in 2017. The experiment was factorial based on a completely randomized design with three replications. The first factor included the planting bed (Mix two parts of the sand and a part of the soil) at 4 levels (sterile and non-sterile planting bed, and Sterile planting bed inoculated with Glomus mossaeae and Glomus intraradices), and the second factor was salinity at three levels (0, 40 and 80 mM NaCl). After plant establishment, salinity treatments were applied at intervals four days for two months. Essential oil extraction was carried out using dried leaves in shade with hydrodistillation by the Clevenger, and essential oil content (%) and essential oil yield (ml per plant) were measured and calculated. The results of this study showed that with increasing salinity, plant growth characteristics such as plant height, stem diameter, the number of lateral shoots and leaves, leaf area, essential oil yield, and fresh and dry weight of leaf, stem, and root decreased, but essential oil content increased. Inoculation with both species of mycorrhizal fungi increased growth, shoot yield, essential oil content and yield in salinity conditions, so that, the essential oil yield in plants treated with G. mossaeae and G. intraradices in 80 mM NaCl were 35.6 and 51.7 percent higher compared with control, respectively. Based on the results and considering the compatibility of mycorrhiza with the environment, the use of these fungi is recommended to reduce the destructive effects of salinity in peppermint.

**Keywords:** Peppermint, Salinity, Sodium Chloride, Growth Characteristics, Essential Oil
Study of flavonoid compounds in *Scutellaria multicaulis*, *S. nepetifolia*, *S. patonii* and *S. condensata* from Zagros, Iran

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The genus *Scutellaria* L. belonging to Lamiaceae family and Scutellarioideae sub-family has 425 species throughout the world and 22 species in Iran from which 10 species are endemic. *Scutellaria* species are considered as medicinal plants containing significant chemical compounds. The aim of this research is to determine the flavonoid constituents in species such as *S. multicaulis* Boiss., *S. nepetifolia* Benth., *S. patonii* Janzad & Safikhani and *S. condensata* Rech. f. in the south-west, center, and west of Zagros. Consequently, the flavonoid solution of air-dried leaves from each *Scutellaria* species was extracted using crude 85% MeOH. The purified flavonoid was analyzed using Liquid chromatography-mass spectrophotometry (LC-MS/MS) on a triple quadrupole mass spectrometer. It is noted that the MS2 detection of this process was performed at a negative and positive mode ESI. The results of this study showed a total of 63 flavonoid compounds. Some of the most important flavonoid compounds in each species contain baicalein and taxifolin in *S. nepetifolia*, norwogonin and liquiritin in *S. patonii*, skullcapflavone I and chrysin in *S. condensata*, and acacetin and 6-hydroxyluteolin-7-O-glucuronide in *S. multicaulis*. Regarding the existence of morphological similarity among *S. nepetifolia*, *S. patonii*, and *S. multicaulis*, three species were definitely separated. Based on the results of this research, LC-MS/MS is a highly appropriate technique to detect different chemical compounds and valuable in the chemotaxonomic aims of this genus.

**Keywords**: LC-MS/MS, Secondary metabolite, Skullcaps, Chemotaxonomy, Irano-Turanian region

Ethnobotanical study most used medicinal plant species of Naft-chak region (Guilan)

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Ethnobotany is a scientific study on the relationship between humans and the plant world, focusing on the traditional knowledge of indigenous and local people in each area. Naft chak mountainous region is located on the northern slope of Alborz Mountains, in the south of Rahimabad (Guilan Province), between 1500-3012 m.a.s.l. The purpose of this study is to collect and identify plants of important medicinal, which the extensively by local residents, are used in the treatment of disease. The region of Naft chak was discovered using a geographical map and then the plant species were collected from various localities. In the final stage, the collected plants were identified scientifically after deposition in the University of Guilan herbarium. A total of 56 medicinal species belonging to 31 families and 50 genera are identified in the area. The most abundant families of the region are Lamiaceae with 9 taxa (43%), Lamiaceae with 8 taxa (38%) and Asteraceae with 4 taxa (19%), respectively. Nowadays, despite the scientific advances and facility of access to chemical drugs, continuously the effective role of the medicinal plant can be seen clearly in the aboriginal life of these areas.

**Keywords**: Ethnobotany, Medicinal Plant, Naft-chak

**Keywords**: LC-MS/MS, Secondary metabolite, Skullcaps, Chemotaxonomy, Irano-Turanian region
Introducing the medicinal plant species of some forest area in Masuleh, Guilan province

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Medicinal plants have been considered among the Iranian people from ancient times and some of these plants were also considered sacred through healing. Iran has unique and rich potential in the field of medicinal plants and traditional medicine. The development of traditional medicine and the use of medicinal plants to maintain health and treatment of diseases require comprehensive and complete knowledge of the characteristics and properties of medicinal plants. This study has been carried out in order to collect and identify of medicinal plants in some part of Masuleh forest areas of Guilan province. The historical city of Masuleh is located in the western part of the Alborz mountain range and has the average annual precipitation of 973.3 mm and the mean annual temperature of 12.3°C. For this investigation, using the geographical map, the studied area was identified and then the collected medicinal plants were identified. A total of 83 medicinal plant species belonging to 48 families and 69 genera were recorded. In addition to the botanical name, the vernacular name and parts used of plant species were also questioned through interviews with local people.

Keywords: Traditional application, Medicinal plants, Masuleh

Ethnobotanical study plant species according to local information in Kah-kuh forests of Siahkal (Guilan Province)

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The use of plants as medicine is part of the culture of indigenous people that has been formed over the centuries in rural areas. Medicinal plants constitute a significant percentage of Iranian plant species, and the flora of Iran has a high ability in this regard. The purpose of this study is to identify medicinal plants in the area and use the knowledge and experience of indigenous people to introduce the properties of medicinal plants. In this study, identification and introduction of plants in the area were carried out using field surveys, popular information, and library studies. The plant species were identified after collection and for each plant species the scientific name, local name, biological form, extremity used, and common therapeutic. A total of 53 medicinal species belonging to 33 families, 43 genera were recorded in the survey. The families with a high number of medicinal species are including Rosaceae (10 species), Lamiaceae (5 species), Asteraceae (3 species). These families are rich in beans and one of the most important families of flowering plants is in terms of medicinal properties.

Keywords: Traditional medicine, Medicinal plants, Kah-Kuh
Effects of vermicompost and its extract on emergence and growth parameters in Valerian (Valeriana officinalis)

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In this experiment, the first was performed to investigate the effect of vermicompost extract (25, 50, 75 and 100%) on the germination index of valerian seed at Petridis. In the second experiment, the effect of solid vermicompost with 5 levels (0, 20, 40, 60 and 80% V) on the emergence index and seedling growth, a greenhouse study was conducted in Faculty of Agricultural Sciences, University of Guilan. Results first experiment showed maximum germination capacity and emergence index also minimum mean germination time T50 obtained 50% vermicompost extract. Treatments 25 and 50% increased the length to shoot and treatment 100% decreased. The second experiment 20% treatment solid vermicompost increased emergence index and fresh weight vigor and decreased mean emergence time. Also, treatment 20 and 40% solid vermicompost increased chlorophyll a, b and total chlorophyll, carotenoid and 90 old day's plant than control. High concentration solid vermicompost was decreased these parameters. The results of this study can be concluded the effect of vermicompost on seed germination and growth seedling depended on concentration.

Keywords: Chlorophyll, Concentration, Emergence index, Valerian

Effects of exogenous sodium nitroprusside and hydrogen peroxide foliar application on essential oil yield and content of Basil (Ocimum basilicum L.) under salt stress

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Salinity is one of the most important environmental factors controlling plant growth and development. And recognizing their biochemical and physiological changes to salinity is necessary in order to reduce its negative effects. Therefore, in order to study the effect of the external application of sodium nitroprusside (SNP) and Hydrogen Peroxide in order to reduce the negative effects of salinity induced oxidative stress, the medicinal plant was grown hydroponically. This experiment was based on a factorial arrangement based on randomized complete block design with two levels of salinity (0 and 50 mM Sodium Chloride) and three levels of sodium nitroprusside (0, 100 and 200 μM) and three levels of solubility Hydrogen Peroxide spraying (0, 2.5 and 5 mM) was performed with three replications. After essential oil extraction, the essential oil yield was calculated according to its content in the plant; The extracted essential oils were also measured by detection of secondary metabolites by the GC-MAS device. The highest amount of essential oil was obtained at a salinity level of 50 mM with external application of 100 μM SNP and 2.5 mM hydrogen peroxide. Also, according to the results, methyl chavicol, linalool, and cadinol showed higher amounts than other metabolites in the treatments. The amount of melkavicle as the most important basil substance was affected by the external use of SNP and its highest rate was observed in the external application of SNP (200 Mm). As a final result, we can say that exogenous application of SNP and H2O2 While positively affecting the accumulation of active ingredient of basil reduces the oxidative stress caused by salinity by increasing the accumulation of active ingredients.

Keywords: Sodium nitroprusside, Hydrogen peroxide, Basil (Ocimum basilicum L.), Salt Stress, Secondary Metabolite
Production and accumulation of secondary metabolites in plants are influenced by genetic and environmental conditions such as height. *Stachys byzantina* is one of the most valuable medicinal species belonging to the Lamiaceae family. Traditionally, the plants in this family are widely used in the treatment of diseases. In this study, to investigate the effect of altitude on the amount of phenolic compounds of this leaf and flower plant of *S. byzantina* were collected from three habitats in Mazandaran, the first habitat (Sari with a height of 376 m), the second habitat (Kiyasar with a height of 1612 m) and the third habitat (Filband with a height of 2357 m), in June 2017. The methanol extract was prepared to measure anthocyanin, total phenol, flavonoid and flavonol and the content of these compounds was evaluated using spectrophotometric methods. A one-way analysis of variance (ANOVA) was performed and the treatment means were compared using Tukey’s HSD all-pairwise comparisons at the *p* < 0.01 level as a post-hoc test. The results indicated that the leaf had higher phenolic compounds than the flower and the total phenol content and anthocyanin increased with altitude decrease. Also, the highest amount of flavonoids and flavonol has been observed in Kiyasar habitat. In general, it seems that altitude does not have the same effects on the production of phenolic compounds in the *Stachys byzantina* plant and possibly other climatic conditions affect the changes in phenolic compounds.

**Keywords:** Iranian, Ecophysiology, Lamiaceae, Essential

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Salinity is an important environmental factor limiting plant growth and development. Salicylic acid is a phenolic compound and plant growth regulator that mitigates the adverse effects of salinity stress. This study was conducted to evaluate the effects of salicylic acid on the growth and physiological responses of the *Melissa officinalis* L. grown under salt stress. First, sodium chloride at 0, 25 and 75 mM concentrations were added as irrigation to the soil of 40 days plants and then, different concentrations of salicylic acid (0, 150 and 300 ppm) were sprayed on the plants twice during the growth period. After 45 days, the interaction of salicylic acid and salinity on growth, photosynthetic pigments, osmotic adjustments and volatile compounds were studied. The results of the analysis of variance showed that fresh and dry weights, chlorophyll and carotenoids contents of seedlings decreased significantly under salt stress while proline contents increased significantly. Application of salicylic acid decreased effects of stress and at 300 ppm concentration, with 75 mM of NaCl increased proline as a tolerance index. Citronella with 52.17 percent of the total volatile compounds had the highest concentration in the aerial parts of the plant. As salinity and salicylic acid levels increased some of the volatile compounds such as Geranial, Neral, karahanaenone, and Geraniol were decreased, whereas Caryophyllene and Cis-pinocarrol increased slightly. Therefore, salicylic acid helps plants to tolerate salt stress with elevated proline and change content and type of volatile compounds.

**Keywords:** *Melissa officinalis*, Plant hormone, Sodium chloride, Gas chromatography-mass spectrometry, Citronella
Production of phenyl ethanol glycosides in *Scrophularia striata* in response to cadmium

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Scrophularia striata is a native plant in Iran that has valuable compounds including phenylethanoid glycosides such as echinacoside and acteoside. These compounds have strong antioxidant properties and are produced by the plant to deal with various environmental stresses such as heavy metals. Cadmium is the heavy metals that enter the soil due to industrial contaminants. Plants display different physiological responses to this contamination. In order to study the physiological responses, these plants were placed in a Hoagland medium containing cadmium at concentrations of 0, 50, 200, 400 micro molar and collected at 12, 24, 48, 72 hours. HPLC was used to measure the phenylethanoid glycosides composition in both root and shoot organs in test and control groups. The results showed that both compounds in both organs at concentrations of 200 micro molar at 48 times increased significantly compared to control groups. Also, it was found that there is a direct relation between the amount of metal in the food solution and the amount of metal absorption and accumulation in the plant. With increasing cadmium concentration in the medium, dry weight of the shoot and root decreased significantly compared to the control, but there was no significant difference at 12, 24 and 48 hours. In general, it can be concluded that the resistance of the plant to cadmium contamination is 200 micro molar for 48 hours during the defensive reactions against this stress occur.

**Keywords:** Acteoside, Echinacoside, Phenylethanoid glycosides, *Scrophularia striata*

Medicinal plants of Golestan provinces of Iran

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The ethnobotany is a branch of botany which referred to the knowledge of people. In this current research, experiences of Turkeman people who live in the east of Golestan were considered. In order to obtain all information first making a questionaries', fill out all the forms, interview with skilled people and production of all medicinal plants were made. Totally, 50 medicinal plants were recorded such as *the longifolia* (Hud) (Labiatae), *Peganum harmala* L. (Zygophyllaceae), *Falcaris* sp. (Apiaceae), *Glycyrrhiza glabra* L. (Fabaceae), *Euphorbia bungei* Bioss. (Euphorbiaceae), *Teucrium polium* L. (Labiaceae), *Malva neglecta* wallr. (Malvaceae), *Satureja mutica* Fish. (Labiatae), *Plantago lanceolate* L. (Plantaginaceae), *Chenopodium album* L. (Chenopodiaceae), *Calendula persica* C.A.Mey. (Asteraceae), Medicinal plants in this area are needed to be attention, because of some hazel factors, these plants are threatened by grazing, overused, and other harmful factors. It would be recommended. This area protected from the previous harmful factors.

**Keyword:** Ethnobotany, East Golestan, *Plantago lanceolate* L. (Plantaginaceae)
Effect of Boron and Zinc elements on some vegetative and reproductive characteristics of *Rosa damascena*

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*Rosa damascena* is a plant its growth and flowering levels are severely dependent on nutrients, and adequate nutrition should be provided to achieve proper function. This research was conducted in one of the gardens around Meymand in Fars Province in a randomized complete block design with four replications. In this study, different concentrations of the element Boron (0, 1 and 2 per thousand) and Zinc (0, 1 and 2 per thousand) were sprayed in two stages, 10 days apart, one month before flowering. Different vegetative and reproductive characteristics were evaluated. Based on the results, the highest number of stamen was obtained by one-thousandth of Boron, while the lowest number of stamen was observed in the control. Although the lowest number of petals was observed in the Boron spray, however, the Zinc treatment significantly increased the number of petals. Also, the use of nutrients increased the plant length as well as the highest and lowest plant lengths was observed in the boron spray and control. The soluble solids content of petals was also significantly increased under Zinc spray application. Although the treatments did not show a significant effect on the phenol content of petals the application of micronutrients had a significant effect on the quality of *Rosa damascena.*

**Keywords:** *Rosa damascena*, Vegetative growth, Flower quality, Microelements

Evaluation of quantitative and qualitative traits of coriander (*Coriandrum sativum* L.) using different drying methods

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The present study was performed to investigate the effect of different drying methods such drying in shade, sun and oven on some of the qualitative and quantitative characteristics of coriander based on completely randomized design, regarding to the importance of aromatic vegetables such as coriander and also the high effect of drying methods in quality and quantity of the plants. This experiment was performed by five treatments including drying in the shade, sun, oven (45 °C, 55 °C and 65 °C) with three replications. Some traits such as drying time, total chlorophyll, essential oil content, antioxidant activity, a phenol compound, microbial pollution level, and color were measured. Results indicated that the maximum total chlorophyll, phenol compound, and antioxidant activity were obtained in the drying method in 45 °C in oven, shade and 65 °C in an oven, respectively. The best color quality was observed at 45 °C in an oven. The results showed that drying in the oven could reduce microbial contamination, however, it reduced essential oil content. The fastest method for drying was using the oven at 65 °C.

**Keywords:** Coriander, Drying, Microbial contamination, Oven
Evaluation of trigonelline content in some Trigonella species

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Trigonelline (TG) which is derived from the methylation of the nitrogen atom from nicotinic acid, is thoroughly known for hypoglycemic, hypolipidemic properties. This study was taken to analyze genetic diversity in respect to evaluation and fluctuation of trigonelline content between five species of genus Trigonella L., comprised of T. uncata, T. monanta, T. arcuata, T. persica, and T. coerulescens. Hence the seeds of mentioned species were collected from different cultivation areas of Iran. By HPLC method, mobile phase composition was SLS (pH adjusted to 3 with HCl): acetonitrile (50.0:50.0 v/v) and the detection wavelength was at λ=265 nm. The result showed significant variation in alkaloid TG in gradient among different mentioned species. Majority of them were rich in TG and its recorded value was higher than standard pharmaceutical levels. Seeds of T. coerulescens, local selected clone from Azarbayjan-e Gharbi-Makoo with the amount of 92.78 µg/ml, presented a higher level in comparison with other species, and the lowest one (18.22µg/ml) belonged to T. monanta, which was cultured in Kermanshah- Javanrood. In addition, the highest amount of trigonelline obtained between T. uncata ecotypes belonged to Tu1 that was cultivated in Hormozgan-Hajiabad. Generally, TG content can be influenced by genotypic characteristic rather than environmental factors, however totally the resultant interaction between genotype and ecological factors impresses the content of alkaloid as a secondary metabolite. Our research confirmed that trigonelline was distinctive among the species and it can be considered as a taxonomic character for discrimination the taxonomic relationships between the different taxa at the inter- and intra-specific levels.

Keywords: T. coerulescens, T. persica, Trigonelline, HPLC

Effect of TiO2 spraying on growth parameters of Dracocephalum moldavica L.

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Nanoparticles are particles of which at least one dimension is between 1 and 100 nm. Titanium dioxide nanoparticles can stimulate cell division, increase cell size, increase light absorption and photosynthesis and so increase plant growth. In order to study the effects of spraying Titanium dioxide particles on growth features of Dracocephalum moldavica L., an experiment was carried out based on randomized complete block design with three replications under laboratory conditions. Treatments consisted of control (non-spraying), spraying concentrations of 50 and 100 ppm NPs at the stage of six adult leaves. According to the results, root length in concentrations of 50 and 100 ppm NPs reduced significantly. In 50 ppm concentration, the fresh and dry weight of root increased significantly. Shoot length and fresh and dry weight in both concentrations of 50 and 100 ppm increased significantly. This study showed that spraying 50 ppm concentration of Titanium dioxide nanoparticles improve growth of Dracocephalum moldavica L. plant.

Keywords: Dracocephalum moldavica L, Nanoparticle, Titanium dioxide, Growth features
Effect of Arbuscular Mycorrhizal fungi symbiosis on growth parameters of *Dracocephalum moldavica* L. medicinal plant under salt stress

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Salinity is one of the most important abiotic stresses which affects plant growth and yield. Mycorrhizae is one of the most useful symbioses which has positive impacts on the growth and yield of plants. In order to study the effect of Arbuscular mycorrhizal fungi, Glomus mosseae on growth features of *Dracocephalum moldavica* L. under salt stress, an experiment was carried out based on randomized complete block design with three replications under laboratory conditions. Treatments consist of Mycorrhizal fungi at two levels: non-mycorrhizal (NM) as control and Glomus mosseae, and four levels of salinity (0, 50, 100, 150mM) at the stage of six adult leaves. Results showed that salt stress decreased the root length and root dry and wet weight. Mycorrhizal plants had higher root length and root dry and fresh weight in comparison with NM ones under salinity stress conditions. Mycorrhizal inoculation increase shoot dry and fresh weight in comparison with NM ones under salinity stress conditions. Total protein of shoot and root in all salinity levels in Mycorrhizal plants in comparison with NM plants increased significantly. Mycorrhizal plants showed higher chlorophyll content than non-Mycorrhizal plants. This study showed that Mycorrhizal inoculation improves growth of *Dracocephalum moldavica* L under salinity stress.

**Keywords:** *Dracocephalum moldavica*, Symbiosis, Arbuscular Mycorrhizal, Salt stress

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Evaluation of the anticancer effect of hydroalcoholic extract of rhizome and aerial part of *Acorus calamus* L. on MCF7 cell line of breast cancer

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Breast cancer is a severely heterogeneous disease that owing to interfacing effect of hereditary and environmental risks factors and leads to a progressive accumulation of genetic and epigenetic changes in the breast cancer cells. The aim of this study is to investigate the anti-cancer effect of hydroalcoholic extract of rhizome and aerial part of the *Acorus calamus* L on the MCF7 cell line of breast cancer. *Acorus calamus* plant was picked out from its natural habitat in Mazandaran province, by shade dried and saved the form. Subsequently, the hydroalcoholic extract was obtained from dried Rhizome and leaves powder has extracted in Soxhlet apparatus. The MCF7 and 4SKIN cells, as the normal fibroblast cells, were treated with 8 different concentrations (0-2mg/mL) of *Acorus calamus* extract after 24, 48, and 72 hours of incubation, cell viability was investigated through MTT Assay. Statistical analysis was done by SPSS 16 software. According to our results, the hydroalcoholic extract of the rhizome and the leaves of *Acorus calamus* could inhibit the growth of MCF7 cancer cells in a concentration and time-dependent manner. The viability of breast cancer cells was significantly decreased according to the increase of the extract concentration and incubation time. This study shows that the hydroalcoholic extract of the rhizome and the leaf of *Acorus calamus* have significant inhibitory effects on breast cancer cells. However, more studies are needed to find out the effective components of the *Acorus calamus* extract in order to be applied as a new and effective medicine for the breast cancer prevention and treatment.

**Keywords:** *Acorus calamus*, Cytotoxicity, MCF7, Hydroalcoholic
Qualitative and quantitative assessment of the essential oils of Yarrow (Achillea wilhelmsii C. Koch.) wild masses in the west-Azerbaijan province of Iran

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Yarrow (Achillea wilhelmsii C. Koch.) is a medicinal plant that has many medicinal properties that grow in Iran. In this study after identification of the plant habitats in West-Azerbaijan province, samples of the plants were collected at the flowering stage from three regions Mahabad-Borhan, Miandoab-Qaryaghi, and Simineh-City entrance. After drying the plants, the essential oils were isolated by Clevenger's apparatus and the components were analyzed by a gas chromatography/mass spectrometric (GC/MS). The results showed that the percentage and type of essential oil components were different among the masses. Totally 24 essential oil components were identified that 15 components were present in the Mahabad-Borhan population, 14 components were in the Miandoab-Qaryaghi population, 9 components were in the Simineh-City entrance population. The most important components in the Miandoab-Qaryaghi included Camphor (42.45%), Gamma-terpinene (10.35%), Piperitone (7.08%), Alpha-thujone (7.04%), Camphene (6.62%), 1,8-cineole (6.21%) and trans-Carveol (5.82%). The most important components in the Mahabad-Borhan included Camphor (27.97%), Gamma-elemene (18.57%), Piperitone (16.83%) and 1,8-cineole (8.72%). Also, the most important components in the Simineh-City entrance included Camphor (50.01%), 1,8-cineole (14.56%), Alpha-thujone (8.89%), Transe pipertiol (6.29%), Beta-thujone (6.04%) and Borneol (5.73%). According to the results, the components of Camphor, Alpha-thujone, 1,8-cineole, Piperitone, and Camphene are the predominant essential oils components in all three masses. The highest percentage of the essential oil composition in all three populations is related to Camphor. A population of the Simineh-City entrance with a high content of Camphor, the population Mahabad-Borhan with a high content of Gamma-elemene and Piperitone and the Miandoab-Qaryaghi with a high content of Camphor and Gamma-terpinene, can be used for the pharmaceutical industry and breeding programs of this plant.

Keywords: West-Azerbaijan, Yarrow, Essential Oil, GC/MS, Camphor

Identification of the essential oils of Tanacetum pinnatum Boiss. growing in Kordestan province of Iran

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Tanacetum pinnatum Boiss. is a medicinal plant belongs to the family Compositae or Asteraceae. The genus Tanacetum contains, totaling over 200 species and distributed over West Asia and Europe. 26 species are in Iran that 12 species are native. In traditional medicine, this herb is used as anti-allergic and anti-inflammatory drugs and for the treatment of some digestive problems. In this study, a sampling of the plant was done in the flowering stage from Kordestan-Chenareh (Veleh) region. The essential oil of the plant was isolated by Clevenger's apparatus and the components were analyzed by a gas chromatography/mass spectrometric (GC/MS). The results showed that the 14 components that represented 95.48% of the total essential oil composition were identified. The most predominant were beta-Phellandrene (20.03%), Chrysanthenyl acetate (17.78%), 1, 8-cineole (13.31%), Borneol (10.08%), Bornyl acetate (8.33%), and Camphene (6.40%). Due to the components of the essential oils, this plant can be used for the pharmaceutical industry and the breeding programs.

Keywords: Medicinal Plant, Kordestan, Compositae, GC/MS, beta-Phellandrene
Decolorization of recombinant laccase from *Bacillus subtilis*

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Bacterial decolorization is associated with involvement of various enzymes such as lignin peroxidase, laccase, azoreductase and biotransformation enzymes. Especially, bacterial laccase plays an important role in the decolorization process. Hereby, the broad substrate range makes laccases candidates for many industrial and biotechnological applications, such as bioremediation, decolorization of synthetic dyes, and biosensors. In this research, the recombinant laccase was trialed for its ability in industrial dye decolorization. The laccase-producing bacterial strain was isolated from Petrol-contaminated soils in Istanbul. In this research, the laccase gene was cloned from *Bacillus subtilis* and efficiently expressed in *Escherichia coli* BL21DE3 in a biologically active form. A dye decolorization experiment was conducted using six dyes, Turquoise blue Hf6 (λmax=613nm), Remazol Black R.B (λmax=595nm), Remazol Brilliant Red RR (λmax=590nm), Coomassie brilliant blue (595nm), Crystal violet (570nm), Methyl red (520nm). The reaction mixture contained 0.1 M citrate-phosphate buffer (pH 6.0), dye (4 mM), purified laccase (100 U), and the CuSO₄ (0.1 mM). The reaction incubated at 50 °C under mild shaking conditions. The control samples were run in parallel without the addition of laccase. After 6 hours of incubation at 50 °C in the test tubes, significant differences were observed in the control tubes at pH 7. Acknowledgment: The authors thank Pamukkale University, Scientific Research Project Funding (PAÜBAP) for their financial support [Project number: 2016FEBE043].

Keywords: *Bacillus subtilis*, Decolorization, Laccase, *E.coli*.

Isolation and identification of bioethanol producer *Saccharomyces cerevisiae* from Iran's alcohol producer manufactures

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Reducing fossil fuels, increasing fuel prices and carbon dioxide emissions, and concern about climate change are encouraging factors for biofuels production. Microbial biofuels are liquid and gaseous fuels which produced by microorganisms. Bioethanol is a suitable alternative biofuel for petroleum-based fuels. For the industrial and high-scale production of bioethanol, the first step is the isolation of suitable yeast strain with high tolerance to ethanol and high-level ethanol production. Hence, the aim of this study was Isolation of industrial strain *Saccharomyces cerevisiae* with high tolerance to ethanol from Iran's alcohol manufactures. Sampling from several Iran's alcohol manufactures was carried out for the isolation of *Saccharomyces cerevisiae* with the highest ethanol production and tolerance. Selected yeast strain was examined by morphological, biochemical tests such as sugars fermentation, absorption of nitrogen compounds and carbon compounds, and also molecular test. The selected yeast strain produced 8% ethanol and tolerated 12% ethanol. The morphological, biochemical and molecular tests confirmed that the yeast strain is *Saccharomyces cerevisiae*, and this strain was named *Saccharomyces cerevisiae* Sahand 101. The isolated industrial yeast strain with the suitable characteristics in terms of ethanol production and tolerance to ethanol can be used for further studies and increase ethanol production by optimization methods at the level of flask and bioreactor.

Keywords: Isolation, Identification, Biofuel, Yeast, Bioethanol
Optimization of ethanol production by industrial strain of *Saccharomyces cerevisiae*

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Ethanol production can be increased by changing in dependent and independent parameters of medium. Therefore, the process of optimizing the medium is based on the interactions between the compounds of the medium and other factors involved in cell metabolism, which is carried out by an effective experiment design tool. In fractional factorial methodology such as Taguchi, with reducing the number of experiments, fraction of the complete factorial experiment is only performed. In this study, the growth rate of yeast and the amount of ethanol production were investigated in aerobic and anaerobic conditions. Selected *Saccharomyces cerevisiae* strain produced 8% of ethanol in molasses with brix18 at 29°C. Then, four factors of the medium, including molasses, urea, ammonium sulfate and pH, were selected in order to optimization by Taguchi method. After optimization, production of ethanol reached to 10% and 11% in flask and bioreactor culture, respectively. The results showed that nitrogen source has a significant effect on the production of ethanol by yeast under anaerobic conditions.

**Keywords:** Bioethanol, *Saccharomyces cerevisiae*, Optimization, Taguchi method

Isolation and characterization of *Bacillus* bacterium producing protease enzyme from the Sardabroud River, Mazandaran Province

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Enzymes are important bacterial-produced compounds that have many industrial uses. So that many applications used in various industries, including the textile industry, pharmaceuticals, leather, detergents, among *Bacillus* bacteria is one of the most important microorganisms that produce the protease enzyme. In this research separated samples have taken from water and sediment of Sard-abrod river of Mazandaran province. Then separated samples have been identified by biochemical tests. In order to investigate the ability of bacteria to produce protease enzyme, strains cultured in the skimmed agar culture medium, after producing a transparent hollow around colonies. Different strains were compared in terms of the production of protease enzymes. Two isolates were selected from the all pure isolates. The optimum production of protease enzyme was measured by isolating at different times and at different temperatures. The highest protease activity for both selected isolates was determined by 24 and 48 hour incubation times and 37°C, respectively. The results of biochemical tests indicate that the isolated bacteria has related to the *Bacillus* genus, which has the ability to produce protease and hydrolyze casein. Optimum pH for the enzyme was determined by measuring its activity and its stability at various pH values at 37°C.

**Keywords:** *Bacillus* bacteria, Isolation, Protease enzymes, Mazandaran province
Isotherm and kinetic of removing Cr (VI) with *lelliottia amnigena* isolated from Babolroud river

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Water is one of the main sources for human life. These days while population increase need of water increase too and industrial activities causes pollutant entry to the water sources that one of these pollutant is heavy metal specially Cr(VI). There are different methods for removing heavy metal that one of the cheapest and harmless is biosorption. In this study the ability of *lelliottia amnigena* that isolated from Babolroud river, Babolsar, Mazandaran had been measurement and used to remove Cr (VI) different parameters such as different concentration of Cr, check the active and passive (live and dead) condition of bacteria, desorption agent, determining the type of isotherm, the different method of bacterial dying, kinetic, pH parameter, concentration of bacterial biomass, age of bacteria, temperature, shaker. SEM_EDX and FT-IR analyses for show the extra surface before and after remove metal of bacteria had been evaluated too. In addition, binary experiment with Cr and Cd, Cr and Pb, Cd and Pb and ternary experiment with Cr, Cd and Pb that the bacteria was not resistant to Cd and Pb had been done. For determine the name of this bacteria 16s RNA sequence assay had been done. Finally, the result of this research showed that *lelliottia amnigena* have high potential for biosorption of these metals singly and have high proper efficiency for binary and ternary of all these metals.

**Keywords:** *Lelliottia amnigena*, Biosorption Cr (VI), FT-IR, SEM

Use of *Bacillus* sp. isolated from the Babolroud River in absorbing the Congo red dye

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The pollution of water, as the most considerable mineral in human lives, is meeting its climax as a problematic issue. Among various methods of purification of water, the biological methods appear to bear more safety and to cost cheaper than other methods for their practicably use of plants, fungi, yeast, and bacteria to absorb azo dyes from water. Obviously, the purification of water is necessary for the continuation of distinctive aspects of life; for instance, farming, and drinking. This analysis delves into investigating *Bacillus* sp. that was isolated from Babolroud river, Babolsar, Mazandaran, Iran. Congruently, a thorough observation occurs on the ability of this *Bacillus* sp. in different parameters; the dye concentration (50-1000 mg/l), the examination of the active and the passive form of bacterial conditions (live and dead), the adsorption and the impacts of adsorption agent, kinetics (0-180 min), and the determination of isotherm type. Linearly, during this research usage of (SEM) and (FT-IR) were analyzed to determine the shape of the bacteria and the absorption graph before and after dye absorption. Thereupon, the features like the temperature (10-50 °C), the concentration of bacterial biomass, the age of bacteria, pH (1-13), the shaker, the different methods of demolishing bacterium were evaluated. Additionally, the binary and ternary of dyes absorption process was carried out and applied on the Red Congo, the Malachite Green, and the Methylene Blue; as a result, it was observed that the bacteria is able to absorb these dyes from the experimental environment.

**Keywords:** Absorption, Bacillus, Congo red, FT-IR, SEM
Isolation of antimicrobial compounds producing actinobacteria from the shore soil of Lake Urmia

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In recent years, the emergence of multi-drug resistant bacteria has caused many problems for patients, clinical settings, and human societies. Therefore finding a new source of effective antimicrobial agents is inevitable. At the present, the marine microorganisms, especially actinobacteria, have been considered as potential sources of antimicrobials compounds. Also, extreme ecosystems are untapped sources of microorganisms with the ability to produce new biological compounds. At the present study, the shore soil of Lake Urmia (which is a hypersaline lake) was investigated to find out antimicrobial compounds producing bacteria. In order to demonstrate the production of antimicrobial compounds, isolated bacteria were exposed to pathogenic bacteria and the inhibition zone was investigated. The results showed one isolate had antimicrobial activity against pathogenic bacteria. According to morphological characteristics and sequence of the 16s rRNA gene, this isolate was identified as Streptomyces. The results of this study revealed bacteria from shore soil of Lake Urmia could be potential sources of antimicrobials and even other new biological compounds. Further studies are needed to purify and identify the chemical properties of the produced antimicrobial compounds.

Keywords: Antimicrobial compounds, Actinobacteria, Lake Urmia

Intra- and Extra-cellular Biosynthesis of Silver Nanoparticles by Yarrowia lipolytica

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Nanotechnology is an interdisciplinary science that involves the production, manipulation, and use of materials with nano-scale. Nanobiotechnology has emerged as an important branch of nanotechnology. The combination of biological principles with physical and chemical processes generates specific nano-sized structures. Nanobiotechnology applies the capabilities of biological systems in generating a variety of nano-sized structures. Plants, algae, bacteria, and fungi are examples of such systems. Nanobiotechnology has played an important role in providing eco-friendly alternative routes for synthesizing nanoparticles. Yarrowia lipolytica is a non-pathogenic yeast and generally regarded as safe with various biotechnological applications. The green synthesis of silver nanoparticles by Y. lipolytica DSM 3286 was investigated in this study. Microscopic observations, scanning electron microscopy (SEM) and spectroscopy analysis revealed the presence of silver nanoparticles. The nanoparticles exhibited a specific peak at 410 nm and the presence of silver elements was confirmed by energy dispersive X-ray (EDX). Scanning electron microscopy (SEM) and transmission electron microscopy (TEM) micrographs showed both intra- and extra-cellular synthesis of silver nanoparticles.

Keywords: Yarrowia lipolytica, Synthesis, Intracellular, Extracellular, Silver nanoparticles
Biological synthesis of L-dopa from L-tyrosine by native yeast isolated from the Caspian Sea

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L-DOPA (L-3, 4-dihydroxyphenylalanine) is the most effective treatment for Parkinson's disease. In the present research, isolation and characterization of L-tyrosine-degrading yeast strains and evaluating their biocatalyst ability for the biological conversion of L-tyrosine into L-dopa was investigated. L-tyrosine-degrading yeast strain Mar19 with the highest ability to converting L-tyrosine into L-dopa was phenotypically and molecularly characterized and its ITS1–5.8 S–ITS2 rDNA sequence was submitted as Debaryomyces sp. strain Mar19 (GeneBank accession no. KX268664). The effect of various parameters on bioconversion L-tyrosine into L-dopa was investigated under resting cell strategy. Based on our results, the maximal L-dopa concentration (0.56 g/l with a molar yield of 40%) was achieved after a 48 hours reaction. spectrophotometer observations and mass spectroscopy as well as Fourier transform infrared spectroscopy (FTIR) analyses revealed the presence of L-dopa in the bioconversion reaction. This is the first report on the biological synthesis of L-dopa from L-tyrosine using the genus Debaryomyces isolated from the Caspian Sea.

Keywords: L-tyrosine, L-dopa, Biological conversion, Debaryomyces sp. Strain Mar19

A study on the characterization of extracellular amylases producing from a Bacillus cereus isolate

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Starch is a polymer of glucose linked to another one through the glycosidic bond. Starch hydrolysis is carried out by an enzyme in a moderate and specific condition. Amylases have potential application in a number of industrial processes such as food, fermentation and detergent industries. For commercial applications, amylase is mainly derived from the genus Bacillus. The aim of this study was to determine the characterization of extracellular amylases producing Bacillus cereus. Soil samples were collected from agricultural and gardening area, in Borujen city. Bacterial isolates were screened for amylolytic properties by starch hydrolysis test on a starch agar plate. The isolate with a maximum zone of hydrolysis was identified by various staining and biochemical tests and molecular experiments. Times courses of cell growth, amylase production, was studied by Bacillus strain. Effect of pH on enzyme production was studied by adjusting the incubation at pH 6,7, 8, 9 and 10 and Similarly, effect of aeration speed and glucose concentration was studied by adjusting aeration speed 75, 100, 150, 200 rpm and glucose concentration0/5, 1, 2 (w/v) in the production medium. Enzyme production was studied in various media including NB, molasses and milk media. A bacterial strain identified as Bacillus cereus was isolated from the Borujen soil. Maximum enzyme production was achieved 7/04 U/ml after 72h cultivation in amylase production medium containing 1%starch as an inducer. In this study maximum enzyme production was 7/412 U/ml in pH=8 and aeration speed 200 rpm after 72h. Bacillus cereus showed the highest enzyme activity in beet molasses the amount 4/89 U/ml. Due to the characterization, this isolate and amylase enzyme production by Bacillus cereus can be introduced as a candidate to produce the favorite amount of the enzyme.

Keywords: Amylase enzyme, Bacillus cereus, Culture medium
Screening and identification of halophilic and halotolerant bacteria producing extracellular lipase from West Mazandaran

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Hydrolytic enzymes of halophilic bacteria such as lipase are of particular importance in the industry due to their high stability over industrious hard conditions, such as high temperatures, various pH changes, and salinity levels. In order to screen and purify the halophiles, a selective MH medium was prepared. By analyzing the optimum salt growth, 40% of the purified strains of moderate halophiles and 60% were halotolerant. The optimum growth temperature of all isolates was determined at 25-45°C. After analyzing the morphology and salinity of isolates, biochemical tests, as well as the ability to produce 7 hydrolysis enzymes, were investigated. Approximately 40% of the isolates were able to produce extracellular lipase enzymes, the major producers of which were gram-positive bacteria. The lipase enzyme activity of one of the isolates was measured by titration. In this method, olive oil was used as a substrate. The activity of the lipase enzyme measured by the strain was 1.37 U/ml. The results show that some isolates have the ability to produce several enzymes, and this will depend on the medium culture in which surrounding the bacterial colonies.

Keywords: Lipase, Extracellular Hydrolytic Enzymes, Halophilic bacteria, Halotolerant bacteria, West Mazandaran

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Isolation and characterization of halophilic and halotolerant extracellular nuclease producing bacteria from the West of Mazandaran

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Halophilic microorganisms are considered as an important source of extracellular hydrolytic enzymes with high potential for industrial, pharmaceutical and biotechnological purposes. A sampling of saline springs in the west of Mazandaran province. To isolate the microorganisms from the selected culture medium, relative halophilic bacteria were used. Several re-cultivations were carried out to purify the bacteria. After the morphological identification of isolates, several biochemical diagnostic tests, such as TSI, OF, SIM, Catalase activity, and bacterial Oxidation were evaluated. Then, concentrations of 2.5 and 5% of salt were determined as the optimum concentration of saline growth. Screening of halophilic bacteria producing DNase enzyme was carried out on a medium containing the enzyme substrate. In addition to the compounds used in the DNase medium, some Toluidine Blue was added as a reagent. According to the salt content of each isolate, a certain percentage of salt was added to each enzymatic culture medium to provide optimum conditions for the growth of the isolates. Samples were incubated for 3 to 7 days for production of extracellular enzyme deoxyribonuclease at 37°C. The results showed that 5 isolates were able to change the culture medium around the colony from green to yellow, and thus, the nuclease activity of these isolates was evaluated positively.

Keywords: Nuclease, Deoxyribonuclease, Extracellular hydrolytic enzyme, Halophilic bacteria
Optimization of culture media for extracellular expression of *Trichoderma reesei* endoglucanase II in *Escherichia coli* using response surface methodology (RSM)

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Cellulose is considered to be the most abundant carbohydrate polymer on the earth. Bioconversion of cellulotic biomass has recently garnered a lot of attention for sustainable production of eco-friendly biofuels and chemicals. Enzymatic total hydrolysis of cellulotic biomass could break it down into its constituent monomeric sugars which could be further fermented into commercial products such as alternative fuels. Endoglucanase (EC 3.2.1.4) capable of hydrolyzing the internal β-1,4-glycosidic bonds of cellulose and disrupting its crystalline structure which exposes the individual cellulose polysaccharide chains. Enzyme production costs remain to be a limiting factor in the industrial synthesis of bioethanol. A great deal of research has been geared to decrease costs of enzyme production (down to 20–30 cents per gallon of ethanol), yet less-sustainable corn-derived fuel remains to be a cheaper alternative (3–4 cents per gallon). An appealing strategy for further reducing of enzyme production costs involves the optimization of the culture medium composition and the expression conditions for recombinant production. In this study, the expression of synthetic *Trichoderma reesei* endoglucanase II gene (EGII) in *E. coli* was greatly improved by adjusting the expression condition. In this regard, a recombinant gene was designed and codon optimized for periplasmic expression this protein. Then, gene subcloning employed to insert the synthesized gene into the pET-26b expression vector. Thereafter, the response surface methodology method was employed to design 20 experiments to find out the optimum points for isopropyl β-D-1-thiogalactopyranoside concentration, post-induction period and cell density of induction (OD600). The expression fluctuations were assessed using standard activity assay. Our results indicated that the synthetic EGII gene was successfully codon optimized and subcloned into the expression vector. The enzyme activity results revealed that the optimum levels of the selected parameters are 0.331 mM for isopropyl β-D-1-thiogalactopyranoside concentration, 10.98 H for the post-induction period and 3.41 for cell density (OD600). These optimized conditions led to a 1.7-fold increase in the EGII activity which is highly promising for large-scale EGII overexpression for industrial applications.

**Keywords:** Endoglucanase, *Trichoderma reesei*, Recombinant protein, RSM

Screening of marine bacteria as a catalyst in the biological synthesis of tellurium nanoparticles

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Nanoparticles are tiny materials having size ranges from 1 to 100 nm. Over recent decades, Tellurium applications continued to emerge in different fields including metallurgy, glass industry, electronics, and applied chemical industries. Tellurium nanostructures are used in labeling, imaging, and targeted drug delivery systems and are tested for antibacterial or antifungal properties. In this paper, the green synthesis of Te nanoparticles (TeNPs) by the marine bacterial strain is described. Twenty tellurite-resistant bacterial strains screened according to selective enrichment technique. Based on the results, only strain MTe05 was able to synthesis and accumulates of TeNPs. The strain of MTe05 was selected as superior strain and identified as *Alcaligenes* sp. MTe05 based on the phenotypic and phylogentic analysis. The prepared nanoparticles were examined using Optical emission and absorption spectroscopy analyses, scanning electron micrograph analysis and Fourier-transform infrared spectroscopy (FTIR). TeNPs display an optical absorption bond at 400 nm and the presence of elemental tellurium was confirmed by Energy dispersive x-ray (EDX) analysis. This strain synthesized nanoparticles under a variety of conditions. Scanning electron microscopy (SEM) and Transmission electron microscopy (TEM) micrographs showed both extra and intracellular synthesis of Te nanoparticles.

**Keywords:** *Alcaligenes* sp. strain MTe05, Green synthesis, Tellurium Nanoparticles, Intra/Extra-cellular
Application of marine bacteria as biocatalyst in the synthesis of selenium nanoparticle

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Biogenic formation of various nanostructured materials, including selenium nanoparticles (SeNPs), by live microorganisms, which is widespread in nature, has been increasingly attracting the attention of researchers over the last decade. Selenium as a practical material is an important element in semiconductors with specific physical characteristics such as the anisotropy of thermococonductivity, high photoconductivity, and thermoelectric activities. Because of the biological and industrial properties, SeNPs have wide applications in the fields of medicine, microelectronic, agriculture and animal husbandry. The aim of the current study was to use the potential aquatic bacteria as biocatalysts to bioreduction of sodium selenite to selenium nanoparticles (SeNPs). Fifty selenite-resistant bacterial strains screened according to selective enrichment technique. Based on the results, only strain MeSe15 was able to synthesis and accumulates of SeNPs. The strain of MeSe15 was selected as superior strain and identified as Acinetobacter sp. MeSe15 based on the phenotypic and phylogenetic analysis. The production of selenium nanoparticles in the bioconversion mixture was studied using UV-vis spectroscopy. Se nanoparticles display an optical absorption bond at 430 nm and the presence of elemental selenium was confirmed by Energy dispersive X-ray (EDX) analysis. This strain synthesized nanoparticles under a variety of conditions. Scanning electron microscopy (SEM) and Transmission electron microscopy (TEM) micrographs showed both extra and intracellular synthesis of Se nanoparticles. Keywords: Nanoselenium, Acinetobacter sp. MeSe15, Spectroscopy, Intra/Extracellular synthesis

Isolation and identification of marine yeasts with the potential biological synthesis of selenium nanoparticle

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Nanotechnology is the term used to describe the creation and exploitation of materials with structural features in between those of atoms and bulk materials, 34 with at least one dimension in the nanometer range. Metal nanoparticles have various functions that are not observed in bulk phase and have been studied extensively because of their exclusive catalytic, optical, electronic, magnetic and antimicrobial wound healing and anti-inflammatory properties. The microbial synthesis of nanoparticles had been recently recognized as a promising source of nanomaterials. Due to the mass production of NPs as the easiness of controlling yeasts in laboratory circumstances, the synthesis of numerous enzymes and rapid growth with the use of simple nutrients, the yeast strains possess more benefits other organisms. Selenite is the most toxic form of Se; hence, biogeochemical cycles involving reduction of selenite (Se+4) to elemental selenium (Se0) are of paramount importance. Selenium nanoparticles (SeNPs) are gaining importance in electronics and optics due to their enhanced semiconducting, photoconducting, photoelectric, and catalytic properties. In this paper, the production of this nanoparticle was made by the most efficient marine yeast isolated from waters of different regions. Optical images, scanning electron microscopy (SEM) and spectroscopy analysis revealed the presence of selenium crystals. Nanoparticles displayed a characteristic peak at 323 nm and their presence was confirmed by EDS (Energy dispersive X-ray spectroscopy) studies. This yeast synthesized nanoparticles under a variety of conditions. SEM and transmission electron microscopy (TEM) showed the extracellular and intracellular synthesis of nanoparticles. Keywords: Marine yeast, Selenium Nanoparticles, Spectroscopy, Intra/Extracellular Synthesis
Exopolysaccharides are a natural component that produces by various kinds of bacteria and they have many applications in the food industry. A consolidating and stabilizer role for color and taste of foods and in the pharmaceutical industry. They decrease blood cholesterol and triglyceride and their anti-cancer effects have been emphasized. In this research exopolysaccharide production by Bacillus Sp.IR12 has been done by a liquid culture medium that contains magnesium sulfate, ferrous sulfate, chloride ammonium and D-potassium phosphate, glucose and salt. We study designed test by Taguchi 9.test under 3 variable of temperature 28, 30, 32, time by 48, 72, 96 degrees and salt density 1%, 2%, and 3% and at the end results had been analysis by Quality-4. Results show that by time factor intervention maximum production rate was exopolysaccharide at 32 that was equal to 66/4, but for time factor maximum of exopolysaccharide production was 72 hours equal to 85/4% and for salt density factor we observed that maximum exopolysaccharide production was at a salt density of 3% and equal to 35/5%. General results show that one of these factor effects on optimization of exopolysaccharide production depends on other factors and this bacterium has a good potential for exopolysaccharide production and we can say that this bacterium is the proper choice for industrial usage.

**Keywords:** Exopolysaccharides, Taguchi method, Optimization, Bacillus
Immmobilization of isolated bacteria from east of Kurdistan province in calcium alginate for biosorption of nickel

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Looking at the role of microorganisms in various industrial and biotechnological processes and considering the advantages of using immobilized microorganisms the importance of immobilization is determined. Preventing the free migration of microorganisms during the processes, easy recycling, and reusability, increasing the efficiency of microorganisms in the processes, and enhancing the stability of cells are some of the advantages of cell immobilization. The purpose of this research is to study the nickel absorption on stabilized bacteria in calcium alginate. In this study, bacterial cells were immobilized on calcium alginate. Among immobilization methods, Entrapment and drop methods were applied. The effects of pH, contact time, and concentration of metal ions on nickel absorption were investigated and the desired contact time and optimum pH were obtained. Considering that the active groups have a major role in the absorption of toxic metals, organisms containing these groups can be used. The results of this study showed that the optimum pH was 6 the time of equilibrium was about 150 minutes the maximum absorption rate of the metal by the stabilized bacterium was about 0.45 mM g⁻¹. The bacterium studied was from bacterial Micrococcus. 

Keywords: Immobilization, Calcium alginate, Nickel, Biosorption, Kurdistan

Study of the biodegradability of Azo dye by bacteria and Aspergillus niger

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Azo dyes are one of the most important kinds of dyes used in the textile industries. One of the most determined characteristics of them is having one or more Azo groups (N = N), which act as a bridge between two organic parts. It should be noted that at least one of these groups is aromatic. They are synthetic colorants pollutants released into the wastewater during textile processing in industries and pollute the environment. One of the best ways for solving this environmental problem is wastewater purification by the microorganisms. In this study, the degradation of Azo Orange 7 by using Aspergillus niger and different bacteria have been investigated. Removing the dye was determined by a spectrophotometer. The results show that by using Aspergillus niger and gram-positive bacterium Corynebacterium, 95 and 83 percentage of the dye are removed, respectively. Increasing the weight of the fungus mycelium and the growth of bacteria are enhanced by increasing of dye's concentrations. These microorganisms probably have biodegradability of an Azo dye as a source of the essential nutrient.

Keywords: Biodegradation, Azo dye, Bacteria, Aspergillus niger
Tannase production by *Trichoderma* species in tannin containing medium as a sole carbon source

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The aim of this study was an evaluation of tannase production by *Trichoderma viride* (Tv), *Trichoderma harzianum* (Th), *Trichoderma longibrachiatum* (Tl), and *Trichoderma reesei* (Tr) in the medium containing tannin as a sole carbon source. *Trichoderma* species were cultured in *Trichoderma* fermentation medium (TFM) which contained 1% Tanic acid solution in 0.2 M acetate buffer (pH=5.5) for 5d. Tannase activity determined by addition of 0.5 mL of culture supernatant (crude enzyme) to 2.0 ml of 0.35% (w/v) tannic acid solution in 0.05 M citrate buffer (pH 5.5) in a test tube at 40 ºC. The absorbance was measured at 310 nm. The protein content in the TFM supernatant after 48 h fermentation was estimated by the dye binding method of Bradford. The level of protein produced in the medium by *Trichoderma viride* was lower (P<0.05) than the other species. There were no differences in enzyme production and enzyme specific activity between *Trichoderma* species. The specific activity of tannase for Tv, Th, Tl, and Tr were 43.00, 48.50, 66.50 and 58.00 U/ml, respectively.

**Keywords:** *Trichoderma*, Tannase, Tannin, Enzyme activity

Optimization of beta-carotene production by yeast strains from a waste leather factory

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Yeasts have a special value for human in biotechnology because of the production of pigments. *Rhodotorula* species produces high amounts of beta-carotene. The aim of this study was to maximize the production of beta-carotene at least prices from native yeast species. The four isolated studied were isolated on specific environments during three stages of sampling. Subsequently, using biochemical test and The PCR method two isolates Aa1 and Aa4 were identified. The production of beta-carotene was determined by isolating identified by standard strain in different conditions of salt, nitrogen source, carbon source, aeration, temperature, pH. Optical pigment absorption at470 nm was read by the spectrophotometer. Among the four isolates, only the isolate Aa1 produces a carton-free pigment. The genetic identification of the two isolates Aa1 and Aa4 was similar to those of *Rhodotorula mucilaginosa* and *Debaryomyces Hansenii*, respectively. The results showed that, the maximum production of carotene was obtained after optimization of 75.6 μg / ml for *Rhodotorula mucilaginosa* and 32.7 μg / ml for *Rhodotorula glutinis* standard strain. The isolation of native species and the optimization of their functional activities in the laboratory is not only useful in the production of high-quality industrial products, but also the use of native species is very economical.

**Keywords:** Carotinoide, Beta-carotene, Antioxidant, *Rhodotorula*
Cloning and expression of ocriplasmin in *Pichia pastoris* expression system

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Ocriplasmin, a 27 kDa serine protease, represents a new treatment option for numerous vitreoretinopathies involving symptomatic vitreomacular adhesion (VMA) and vitreomacular traction. When given as an intravitreal injection, ocriplasmin degrades fibronectin and laminin, results in posterior vitreous detachment (PVD). The methylotrophic yeast *Pichia. Pastoris* is a highly efficient expression system that is used in molecular biology for production of recombinant proteins. Being a eukaryote, *P. pastoris* is capable of doing many of the post-translational modifications performed by higher eukaryotic cells including proteolytic processing, proper folding, disulfide bond formation and glycosylation. The aim of this study was to evaluate the expression of recombinant Ocriplasmin in *P. pastoris* expression system. Ocriplasmin gene was cloned into a high copy number vector, pPinkHe and transferred first to *E. coli Top10*. PichiaPink™ vector containing the ocriplasmin was linearized by cutting at a unique site to promote integration into the *P. pastoris* genome and transformed into *P. pastoris* competent cells. The transformed cells were selected on MD agar selection plates after incubation at 28°C for 6 days. A *P. pastoris* clone containing Ocriplasmin was cultured in BMGY and the expression was carried out using BMMY medium and analyzed by SDS-PAGE. Screening of transformed cells on MD agar results in several colonies that were positive for the target gene. Expression of recombinant Ocriplasmin in *P. pastoris* results in 27 kDa protein that was in expected size. The result of this study indicated that *P. pastoris* is an efficient expression system for production of recombinant Ocriplasmin.

**Keywords:** Ocriplasmin, *Pichia pastoris*, Cloning, Expression

Isolation of chrysogen parasitic bacteria from soils in Shiraz petrochemical complex and determination of chrysan remnants by them

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Environmental pollution is one of the most important problems facing the world today. These contaminations occur as a result of the absorption and accumulation of chemicals in the food chain and the damage to and the damage to the plants and living organisms in that climate. Chryseone is a four-ring aromatic compound and one of the environmental threats. In this research, the decaying bacteria of chryogen were isolated from the soils of Shiraz petrochemical region, and then the decomposition of chryosogen was studied under laboratory and kinetic conditions. At first, the specimens were incubated for 7 days at a temperature of 32°C after chilling in a basic mineral medium containing chryogen. After separation and differential chemical testing, the isolate superior strains. Then, the strains were evaluated for growth kinematics and chromatographic analyzes, which were analyzed by chromatography using gas chromatography. At the end of the molecular tests, 16s rRNAs were sequenced to identify the superior strains. Society and Sample: Shiraz petrochemical company and samples of contaminated soils in this region. Sampling method: A random sample was taken from several surface grafting points. Out of 23 bacterial species, two isolated bacteria, *Pseudomonas putida*, and Micrococcus luteus were grown in the shortest possible time, and *Pseudomonas putida* chromatography analysis showed that in the course of 10 days, the bacterium decomposed 85% of 0.1% chrysen slowly. In this experiment, it was determined that two bacteria *Pseudomonas putida* and Micrococcus luteus are two major degenerators of Chryson, and these two bacteria can be tested for the rest of the multi-ring articular hydrocarbons.

**Keywords:** Chrysanth, Bioforms, *Pseudomonas putida*, Micrococcus, Parasitic bacteria
Biodegradation of dibutyl phthalate using *Deinococcus* spp. isolated from Lout desert of Iran

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Phthalates are fatty liquids, which are colorless and odorless, with very low solubility in water; they are used as a plasticizer in most polymer products. These compounds have gained attention due to their high levels of toxicity, carcinogenicity and the emergence of multiple diseases in humans and living organisms. In this study, dibutyl phthalate (DBP) biodegradation ability was investigated using *Deinococcus* spp. isolated from Lout desert of Iran. The strains LD4 and LD5 were inoculated into TSB medium and incubated at 30 °C for 24 hours. Then 5 ml of growth medium was transferred to 100 ml of fresh minimal salt medium with 1 ml of sterile 7% DBP as a sole carbon source. The flasks were incubated on an orbital shaker at 30 °C and 150 rpm for 24-48 hours. Biodegradation of DBP was investigated by two assessment methods including the optical density of growth culture using spectrophotometer (wavelength at 600 nm) and high-performance liquid chromatography (HPLC). The results demonstrated that both *Deinococcus* strains, LD4 and LD5, started degradation after 6 hours then reached to a maximum after 24 hours of incubation. The results of growth assessment in the culture medium containing DBP showed that optical density of growth medium was increased by both LD4 and LD5 from 0.211 to 0.751 and 0.226 to 0.763, respectively. The results of this research revealed that the native *Deinococcus* strains isolated from Lout desert are suitable for the degradation of phthalates in industrial wastewater and contaminated sites.

**Keywords:** Dibutyl phthalate, Biodegradation, *Deinococcus*

Investigation of the possibility of stimulating the growth of single-cell yeasts using the combination of nanoparticles, L-carnitine and HUFA

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Several studies have shown that nanoparticles can increase cell differentiation or enhance cell growth at specific concentrations. While nanoparticles inhibit cell growth in higher or even lower concentrations. Due to the importance of the application of single cell yeasts, the problem in the cultivation of some species and strains of yeast, this study was able to stimulate the growth of yeast cells using these materials. Therefore, Saccharomyces service yeast was grown under the influence of these materials in single and combination treatments. The yeast count was performed after 48 hours incubation period. The yeasts counting results showed that the highest concentrations compared to control revealed 82% increase in TiO2 treatment, 72% in HUFA with lecithin treatment and 40% in treatment L-carnitine. While the combined treatment with L-carnitine and nanoparticles increase by only 24%. Several studies have shown that nanoparticles can enhance cell differentiation or proliferation. In comparing the amount of biomass produced for different treatments, as well as the highest biomass was obtained in the same treatment compared to control. The study showed that the use of L-carnitine could differ the metabolism of fatty acids in yeast which alter culture conditions. The results of this study showed that providing to use of Nanoparticles and L-carnitine could increase of the yeast biomass production up to two folds, which in term of the cost of materials used and their values are highly functional.

**Keywords:** Nanoparticles, TiO2, Lecitin, L-carnitine, HUFA
Isolation and screening of capable halotolerant fungi in hydrocarbon contaminations removal from aqueous environments

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Bioremediation is an eco-friendly approach for oil contamination removal discarded into the environment. Fungi with their high extracellular enzymes production capability are efficient microorganisms in pollutant removal from environments. Therefore, this study was aimed at isolation of capable fungi in oil removal from some contaminated soils of Iran. Following serial dilution preparation of three collected soil samples, the samples were cultured on potato dextrose agar plates. Then, the isolates were evaluated for their growth in the Bushnell-Hus medium as the minimal salt medium containing 2% (v-v) crude oil as the sole source of carbon. To identify the selected fungal isolate, morphological studies were performed using an optical microscope. The isolation and screening process resulted in obtaining a fungal isolate with 78% capability in oil removal from the minimal salt medium. Morphological studies on the colonies appeared on potato dextrose agar plates and slide culture images showed that the isolate belongs to the genus, Aspergillus. Considering the significant efficiency of this fungi in oil removal from oil containing minimal salt medium as the sole source of carbon, Aspergillus sp. can be a suitable candidate for further investigations for application in oil-contaminated zones.

Keywords: oil pollutants, Aspergillus sp., Bioremediation, Isolation

Antibacterial activity of silver nanoparticles synthesized by Shewanella sp. ME1 isolated from sediments of the Caspian Sea

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Nanoparticles are been synthesized by different chemical and physical methods. The biological approaches to the synthesis of nanoparticles are better than chemical and physical procedures because this method requires no toxic solvents and no dangerous material for the environment. In this research biosynthesis of silver nanoparticles has been explored by Shewanella ME1 isolated from sediments of the Caspian Sea. After samples collected and identification of isolates based on morphology, physiology, and molecular characteristics, one was chosen and its ability to synthesis silver nanoparticles was evaluated. The isolate was inoculated in LB medium and incubated at 28 ºC for 24 hours. The supernatant of the broth was examined for the ability to produce silver nanoparticles. The isolate supernatant to aqueous AgNO₃ solution led to the appearance of brown color in solution after 24 h of reaction, indicating the formation of silver nanoparticles. The UV-Vis spectrum exhibits an absorption band at around 414 nm suggesting the formation of silver nanoparticles. SEM photographs showed that the silver nanoparticles formed were fairly uniform in size with a spherical shape and an average diameter of 47 nm. Antibacterial properties of nanoparticles were examined against human pathogenic bacteria by Kirby-Bauer method. The formation of a clear zone around the disk depicts antimicrobial activity. The diameter of zones of inhibition was measured (Escherichia coli 23±1 mm, Bacillus subtilis 22/3±0/5 mm, Pseudomonas aeruginosa 20±1 mm, Klebsiella pneumoniae 14±1 mm).

Keywords: Silver nanoparticles, Antibacterial activity, Shewanella
Microbial production of xanthan gum by *Xanthomonas campestris* using acorn starch as carbon source

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Xanthan gum is one of the most important biopolymers produced by *Xanthomonas* species. Xanthan gum enhances the stability of solutions at wide ranges of pH and temperature. Adding this substance to the solution increases stability in a wide range of pH and temperature. Glucose and sucrose are the main substrates for the microbial production of xanthan and the relatively high price of these carbon sources, has prevented widespread consumption of this biopolymer. In this research, acorn starch was utilized as an alternative and inexpensive substrate for xanthan production. The starch content of acorn fruit was 58.9%. Therefore 67.7 g of acorn powder was dissolved in 1 L of culture medium. A fresh culture of *X. campestris* on GYC solid medium was transferred to YPD liquid medium. Then, culture medium containing a specific amount of cell was inoculated to the xanthan production medium. Xanthan titers after 24, 48 and 72 h (average of 3 replicates) were 12.8, 15.7, and 21.34 g/L, respectively. The results of this study, which showed an increase in the production of 28% of xanthan gum compared to the hydrolyzed starch substrate, suggests that oak fruit can be used as an abundant and inexpensive carbon source for the production of xanthan gum microbial. It can be used to optimize production conditions.

**Keywords:** Xanthan gum, *Xanthomonas campestris*, Acorn starch

Screening of biosurfactant-producing microorganisms for application in oil industries

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Biosurfactants are surface-active agents produced by different microorganisms like bacteria, yeasts, and fungi. Biosurfactants have advantages over chemical surfactants such as: biocompatibility, less toxicity, high variety, high efficiency in uncommon temperatures, salinity and pHs, they can be synthesized from cost-effective and renewable sources and they can also be produced through fermentation. One of the main applications of biosurfactants is their usage in microbial enhanced oil recovery (MEOR) processes. The aim of this study is a separation of microorganisms with the ability to produce biosurfactants with the applications in oil processes. Local samples were gathered from different spots in Tabriz refinery like clarifier, oil contaminated soils, oil sludge, and kerosene. Separation of bacteria was carried out in a growth medium of mineral salt (MSM) with crude oil as the only carbon source and heating time of 47°C. Molecular characterization of the chosen bacterium was done via 16S rRNA. Biosurfactant production by separated bacteria was investigated using three semi-quantitative methods of hemolysis, drop collapse and oil spread technique. A quantitative study of production in the chosen isolate was carried out using the method of dry weight measurement. 20 isolates were separated and purified from the samples. The OT9 isolate was chosen as the proper isolate based on the results of tests mentioned above and was characterized as the *Bacillus licheniformis*. The value weight for the produced biosurfactant in OT9 isolate was 2.2mg/mL.

**Keywords:** Biosurfactant, *Bacillus licheniformis*, Microbial Enhanced Oil Recovery
Optimization of effective parameters on biosurfactant production by *Bacillus licheniformis* OT9 using response surface methodology

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Biosurfactants are various amphipathic structures produced by different microorganisms. Amphipathic structures of these materials lead to a reduction in surface tension, interfacial tension, and solubility of immiscible components like water and oil. One of the main applications of biosurfactants is their usage in microbial enhanced oil recovery (MEOR) processes. The aim of this study is the optimization of growth medium and physicochemical parameters effective on biosurfactant production in this isolate. Carbon and nitrogen sources, crude oil, NaCl concentration, temperature and time were investigated as the effective parameters on biosurfactant production by *Bacillus licheniformis* OT9 using experiment design (minitab.v16). The produced biosurfactant amount in each experiment was measured by the surface tension reduction (tensiometry). First, molasses and yeast extract were chosen qualitatively as the carbon and nitrogen sources. Then, using the Plackett–Burman design, 4 factors (nitrogen, temperature, time and crude oil) were determined as effective among the 6 factors mentioned above. After that, optimization of the effective parameters was carried out using Central Composite Design (CCD). The statistical method presented a quadratic equation as a model with the regression value of 94.13%. The optimum values were determined to be: yeast extract 1.12 gr/l, crude oil 1%, time 24 hours and temperature 55°C. These optimum values resulted in the maximum value of produced biosurfactant and surface tension reduction down to 39.5 mN/m.

**Keywords:** *Bacillus licheniformis*, Biosurfactant, Response Surface Methodology, Optimization

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*Variovorax paradoxus* OT16, a potential bacterium for enhanced oil recovery

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Understanding the fact that global demand for energy in the coming years will be at its highest level, need for a change in oil and gas engineering is felt to increase the productivity more than ever. Microbial enhanced oil recovery (MEOR) is a method that has been facilitated by the use of metabolites produced by microorganisms. The purpose of this study is to isolate and investigate the efficiency of potential microorganisms in improving oil recovery from reservoirs. In this research, the oil sludge was collected from Tabriz oil refinery and the effective bacteria were isolated. The superior isolate was identified by sequencing of 16SrRNA and compared with NCBI gene bank. The efficiency of MEOR was investigated at 47°C, heating times of 1 to 14 days with different amounts of bacterial inoculation using the syringe method. Upon completion of the mentioned times, for measuring the amount of oil recovery, 3 mL of 5% brine solution was injected into the syringes and an adequate time was given to the oil to be extracted. *Variovorax paradoxus* OT16 was identified based on 16SrRNA sequencing and its effect on the MEOR process was investigated. For 1 mL injection and heating time of 24 hours, the recovery factor was reached 98% which indicates a high efficiency for this bacterium. According to the tests, this isolate produced gas and acid and these two metabolites affect the physical and chemical properties of porous media in time and as a result improve oil recovery.

**Keywords:** Microbial enhanced oil recovery, *Variovorax paradoxus*, Oil sludge
Increasing oil recovery using *Bacillus halodurans* OT24 isolated from oily clarifier

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Considering the growth of global demand for oil and gas, the use of new technologies for extracting in situ oil of reservoirs with enhanced oil recovery processes (EOR) is necessary. Microbial enhanced oil recovery (MEOR), by the use of microorganisms and their biological products, improve the extraction of residual oil of reservoirs and is more cost effective and less harmful to the environment. The purpose of this study is to investigate efficient microorganisms in MEOR. In this study, the oil contaminated samples were collected from clarifiers in Tabriz petroleum refinery and the effective bacteria were isolated. The syringe model was used for MEOR studies. Experiments with bacterial inoculation amounts of 0.1, 0.2, 0.5 and 1 mL at different heating times were carried out. After giving the system an adequate time, 5% brine solution was used to recover the oil from syringes. In the end, according to the results, the oil recovery factor calculated. Based on molecular and biochemical methods Bacillus Halodurans OT24 was identified and its efficiency in the MEOR process was investigated. The highest calculated recovery factor was 90% in 1 mL of inoculation suspension and 24 hours of heating time. According to the results of the study, the most important factors causing the recovery factor for this bacterium were acid production and a high amount of biomass.

**Keywords:** Microbial enhanced oil recovery, *Bacillus halodurans*, Oily clarifiers

Screening and isolation of baker yeast mutants with freezing and high osmotic pressure resistance and more gas generation ability

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The baker yeast Saccharomyces cerevisiae is known as the major microorganism in the bread-making process and its applications worldwide. Therefore, lots of efforts have been made to develop the properties of its strains, like creating strains resistant to stresses such as; air-drying, freezing, high temperature, high osmotic pressure, and high-sucrose concentrations. The aim of this study was screening and isolation of baker yeast mutants resistant to freezing and high osmotic pressure with high gas generation ability. For this reason, the random mutagenesis by UV radiation process was done in different time cycles in order to find the time point when there are 40% viable cells for each strain. Mutagenesis at that point was repeated and 17 mutants of three baker yeast strains of Y1, Y3 and H were screened and isolated after culturing on YPDA media with high NaCl and Sucrose concentrations. The amount of CO2 gas generated by the resultant mutants was measured by a gas-measuring device and 10 mutants with gas generation ability were isolated. Consequently, two more efficient isolated mutants of each strain were analyzed by freezing tests at -20°C, thus the ones with higher viability in comparison to their wild types were chosen. Eventually, 3 mutants, Y1M1, Y3M1, and HM2, with the highest resistance to freezing and high osmotic pressure stress with high gas generation ability were screened and isolated.

**Keywords:** Baker yeast, Screening, Isolation, Random mutagenesis, Gas generation
Isolation of natural producing Actinobacteria using the iChip technique as a novel screening method and investigation of their antibacterial activity

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Actinobacteria are a large part of soil microbial population which their number is more than one million bacteria per gram soil. Generating a broad range of active compounds as secondary metabolites such as antibiotics has invariably been important. The aim of this research was a screening of natural active compounds produced by actinobacteria using the iChip technique. Various soil specimens were collected from different areas of Mazandaran, Golestan and Fars provinces, Iran. After thermal pretreatment, soil specimens were loaded on iChip and incubated in their own natural environment for 2 weeks. Micro colonies grown in iChip wells were transferred to fresh SCA and SMS media. In order to isolate active actinobacteria, primary screening was performed using cross streak method. The crude extract of active isolates was obtained from each grown medium, then their antibacterial activities were evaluated using disc diffusion method. Out of 87 actinobacteria isolated using the iChip technique, four isolates with most antibacterial activity against some gram-positive and gram-negative pathogens were chosen. The results demonstrated that all isolates had good antibacterial activity against Staphylococcus aureus, Bacillus subtilis, Escherichia coli, as well as Pseudomonas aeruginosa, Enterobacter aerogenes, Shigella sonnei. Moreover, the results of the current study revealed that iChip-mediated screening for natural environments, in finding new microbial species and their domestication from nature, can potentially be sustainable.

Keywords: Actinobacteria, IChip, Natural antibacterial compounds

Introduction of a long chain polyunsaturated fatty acid producing indigenous microalga Dunaliella

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In the present work microalgal cell bank at Agricultural Biotechnology Research Institute of Iran, Northwest and West region was screened in terms of lipid production and fatty acid profile. Due to health benefits of Long-chain polyunsaturated fatty acid (LC-PUFA) such as blood pressure /immune system regulation, neurological/retinal development and cardiovascular protection, the main focus of the study was on LC-PUFA. LC-PUFAs are fatty acids with ≥18, 20 carbons and ≥2 unsaturated bonds including two families of ω6 (n-6) and ω3 (n-3). The main current source for LC-PUFA, fish, has numerous limitations such as its carcinogen, non-carcinogen, antibiotic and mutagen contaminants, undesirable odors, flavors, and tastes as well as their depleting stocks. These provoked the attempts to find safe alternative sources. Few photosynthetic microalgae have emerged as biosynthetic machinery to synthesize LC-PUFAs. Dunaliella species is among limited microalgae approved as human nutrition. Followed by total lipid and fatty acid profile determination, an isolate indigenous of Urmia Lake, Dunaliella sp. ABRIINW-11, with maximum lipid of 40% of the dry weight was selected. Fatty acid profile analysis demonstrated its high level of LC-PUFA as 80% of total lipid. This amount is higher than that of plants, animals as well as oily fish. The n3-PUFA fraction of LC-PUFAs in this isolate was nearly 90%. Human evolved on the long chain polyunsaturated fatty acid producing indigenous isolate Dunaliella sp. Dunaliella species is among limited microalgae approved as human nutrition. Followed by total lipid and fatty acid profile determination, an isolate indigenous of Urmia Lake, Dunaliella sp. ABRIINW-11, with maximum lipid of 40% of the dry weight was selected. Fatty acid profile analysis demonstrated its high level of LC-PUFA as 80% of total lipid. This amount is higher than that of plants, animals as well as oily fish. The n3-PUFA fraction of LC-PUFAs in this isolate was nearly 90%. Human evolved on the n3/n6 ratio of 1/4 to optimum ratio of 1/1. The n3/n6 ratio in the isolate of our interest is high (5/1) which compensates for very limited n3/n6 (~1/15) daily intake in our diet which causes many chronic diseases.

Keywords: Dunaliella, Indigenous isolate, Long-chain polyunsaturated fatty acids, The n3/n6 ratio
The incessant rise of greenhouse gas emission has led to global warming and climate change. Global concern on this phenomenon has suggested the microalgae-based CO₂ sequestration for diminishing greenhouse effects. Microalgae have recently gained enormous attention worldwide, to be the valuable feedstock for renewable energy production, due to their high growth rates, high lipid productivities and the ability to sequester carbon. In this paper, the effect of extremely high CO₂ concentration on two indigenous Dunaliella (CH₂ and SH₃33) microalgae was investigated. We exposed two indigenous isolates of Dunaliella (CH₂, SH₃33) to 0.03%, 10%, 20%, and 30% CO₂ concentrations. Results demonstrated that increasing CO₂ concentrations developed in the sharp increase of the total lipid content in both isolates. Lipid productivity increased significantly with CO₂ rising. In addition, unsaturated fatty acids and thereby the total omega-3 fatty acid content of our studied isolates increased considerably with the rise of CO₂ concentration. As stated by results, in microalgae Dunaliella (CH₂, SH₃33) CO₂ mitigation can be an effective conversion of CO₂ to value-added products.

**Keywords:** Extremely high CO₂ concentration, CO₂ mitigation, Dunaliella (CH₂ and SH₃33)

**Comparative toxicity of graphene oxide nanostructure and reduced graphene oxide-silver nanocomposite to Chlorella vulgaris through microscopic and oxidative stress analysis**

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The specific physical and chemical properties of nanomaterials and enhanced application of them have raised concerns about their probable adverse impacts on the environment. Investigation of toxicity of nanomaterials on algal species can be defined as an important issue for understanding possible risks of these nanomaterials to aquatic environments. In this research, Graphene oxide nanostructure (GO) and reduced graphene oxide-silver nanocomposite (Ag-rGO) were synthesized and their characteristics were determined by XRD, SEM, EDX and Raman techniques. Chlorella vulgaris cells were subjected to GO and Ag-rGO at the same concentrations (1, 10, 20 and 30 mg/L) for a period of 24 hours for evaluating uptake and toxicity impact of these nanomaterials on the H₂O₂ content and contents of phenol and flavonoid. According to the results, Ag-rGO led to a significant reduction in the phenol and flavonoid contents and a significant increase in the H₂O₂ content in comparison with the control. GO had no considerable effect on these parameters. Images of fluorescence microscope revealed that the position of the nucleus has altered in the Ag-rGO treated cells. Nucleus had a lateral position in the control and GO treated samples while it was in the center of the Ag-rGO treated cells.

**Keywords:** Chlorella vulgaris, Nanomaterials, Ag-rGO nanocomposite, Oxidative stress, Fluorescence microscopy
Green synthesis of silver nanoparticles using cell-free extract of *Polysiphonia* algae and their anticancer activity against breast cancer MCF-7 cell lines

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Silver nanoparticles (AgNPs) are one of the best tools for cancer treatment due to their unique physical and chemical properties. Although chemical methods for the synthesis of silver nanoparticles are effective, they are generally expensive and may carry traces of toxic substances, which are hazardous for human and environment. For this reason, in recent years, researchers have tried to develop biological methods for nanoparticles synthesize using living organisms or their products, which are environmentally friendly and harmless for humans. In this study, cell-free extract of *Polysiphonia* algae used as a reducing agent for green synthesis of AgNPs. The colorless AgNO3 extract solution changed to dark brown after 60 minutes that indicating the formation of the AgNPs. The progress of the reaction measured by UV-visible spectroscopy showed the maximum absorption peak at ~420 nm wavelength that confirmed the synthesis of silver nanoparticles. The scanning electron microscope (SEM) and transmission electron microscope (TEM) micrograph showed the most of biosynthesized silver nanoparticles have a predominately-spherical structure with the size range of 5 to 25 nm that is suitable for penetration into cancer cells. Fourier Transform Infrared Spectroscopy (FTIR) analysis of algal extract was used to identify the functional groups responsible for the reduction and stabilization of silver nanoparticles and confirmed the presence of phenolic, aldehyde/ketone, alcoholic, and amine compounds in the extract. To investigate the anticancer effects of silver nanoparticles on MCF-7 cell lines, cell survival and proliferation were measured by MTT and flow cytometry and the results confirmed the anti-proliferation potential of AgNPs against breast cancer cells. In conclusion, the formation of silver nanoparticles with anticancer effect only 60 minutes after the start of the reaction in the presence of a very low concentration of algal extract and at the ambient temperature indicated that *Polysiphonia* is an ideal option for the production of silver nanoparticles with high efficiency and speed.

**Keywords:** Green synthesis, Silver nanoparticle, *Polysiphonia*, Breast cancer, Algal extract

Protective effects of nanosilver particles on Sunflower (*Helianthus annuus* L.) under super gallant herbicide stress

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The innovation of technology in the agricultural industry is the most important factor in creating modern agriculture. Meanwhile, nanotechnology has provided a good basis for the production of food and agriculture. An experiment was conducted in a completely randomized design with 4 levels of Super Gallant herbicide (0, 25, 50 and 75 ppm) in 3 replications to investigate the protective effects of nanosilver (50ppm) on the content of photosynthetic pigments in sunflower (*Helianthus annuus* L.cv.Lakomka) under the herbicide stress. Nanosilver particles were sprayed on the leaves for 2 weeks when the plants were 2-4 leaves. Different concentrations of herbicide were sprayed on the aerial part of the 4-6 leaved plants (on 2 groups: control and nanosilver treated plants). The results showed that leaf area, the fresh and dry weight of roots and shoots and content of chlorophyll a and b decreased with increasing the herbicide level in 2 groups, while contents of carotenoids and anthocyanin increased with increasing stress level in 2 groups. All parameters of nanosilver treated plants were higher than the control, significantly. It seems that the nanosilver spray has increased the biomass of sunflower and it has improved plant tolerance to super gallant stress by increasing the content of carotenoids and anthocyanin compounds.

**Keywords:** Nanosilver particles, Sunflower, Super gallant, Carotenoids, Anthocyanin
Antibacterial effects of saffron formulated extract in chitosan nanoparticles on *E. coli* H7O157

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Today, the use of natural antibiotics is increasing due to microbial resistance to chemical antibio... resulted in the least inhibition. The results of this study showed that both the saffron extract and its formulated extract in chitosan had antibacterial effects against *E. coli* strain H7O157. In addition, the MIC of a formulated extract of saffron was less than the non-formulated extract. **Keywords:** Saffron extract, Chitosan, Antibacterial, *Escherichia coli*

Anti-diabetic effects of ZnO nanoparticles synthesized by *Eryngium billardieri* aqueous extract on diabetic rats

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Nanotechnology is a discipline of applied knowledge. The progress and efficiency of the green synthesis method in the use of natural reducers, fixation of final materials without the use of toxic agents, lack of energy consumption and improvement of the properties of the materials provided. Diabetes is a chronic disorder in carbohydrate metabolism, fat, and protein, and is characterized by increased blood glucose levels in the patient. Cardiovascular failure, renal failure, and decreased neurological activity are the long-term complications of this disease. *Eryngium billardieri* is a perennial herb of the Apiaceae family. This genus contains tannin, saponins, alkaloids, flavonoids, and beta-carotene. The element of micronutrient zinc (Zn) is essential for a variety of biological processes, including glucose metabolism. In this study, ZnO nanoparticles were synthesized by chemical and biological methods using an aqueous extract of *E. billardieri*, and their anti-diabetic effects on diabetic rats were investigated. Structural properties of nanoparticles were investigated using XRD, TGA, UV-vis DRS, FT-IR and SEM analyze. The size of nanoparticles synthesized by chemical methods, 21.5 nm, was determined to decrease the size of the nanoparticles synthesized by the chemical method (25 nm). Also, the presence of plant extract molecules with zinc oxide prepared by these techniques was confirmed. Injection dose was determined for groups receiving nano-materials, 10 mg/kg body weight. The results indicate a significant reduction in fasting blood sugar in the recipient group of bioavailable nanoparticles compared to the control group (p<0.05). **Keywords:** Diabetes, *Eryngium billardieri*, Green Synthesis, Zinc oxide nanoparticles
Synthesis of metal oxide nanocomposites and investigation of the effects on *E. coli*

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Infectious diseases are one of the challenges in the field of medicine. Recently, the use of conventional antibacterial agents has led to microbial resistance to these drugs. This situation is about to reach a dangerous level, which could be the leading cause of mortality in the world in the future. Zinc oxide nanoparticles have anti-cancer, anti-microbial and anti-oxidant activity. This nanoparticle can inhibit the growth of pathogenic bacteria, including *Shigella, Escherichia coli* and *Staphylococcus aureus*. Copper oxide nanoparticles are cheap metal oxides. This metal oxide is a p-type semiconductor with optical, catalytic, antifungal, antiviral and antibacterial activity. In this study, the nanocomposite ZnO / CuO was synthesized using [Zn (NO₃)₂·6H₂O] and [Cu (CH₃COO)₂·H₂O] by microwave technique. To investigate the antibacterial effects of this material on *E. coli*, dilution method was performed at concentrations of 0.8-0.05 mg/ml, ZnO, CuO, and ZnO/CuO nanocomposites in Nutrient broth medium with three replications and investigation survival percentage in 600 nm, as well as MIC and NIC determination were used. Structural properties of nanocomposite were investigated using XRD, FT-IR and SEM analyzes. The nanocomposite size of ZnO/CuO was determined using a Debye-Scherer formula of 25.6 nm. Also, the shape of nanomaterials was spherical. Based on the results of bacterial viability, nanocomposite ZnO/CuO at 0.4 mg/ml and with a survival rate of 12% (88% bacterial degradation) concentration has the highest inhibitory effect on *E. coli*.

**Keywords:** Metal oxide, Anti-bacterial, Nanotechnology, Copper oxide

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Application of titanium dioxide nanoparticles under salt stress on compounds of essential oil (*Dracocephalum moldavica L.*) Under Hydroponic Condition

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NaCl stress is one of the reduction reasons of the medicinal plants and crop yield. This research was aimed to investigate the effect of titanium nanoparticles under salinity stress on quality of essential oil components *Dracocephalum moldavica L.*. The study was conducted in a completely randomized design in greenhouse, controlled environment, with three treatments including 0 (control), 50, 100 mM NaCl and four treatments including (0, 50, 100 and 200 mg/l) TiO₂ Np with three replications per treatment. The essential oil was first extracted and the active components of the essential oil were separated and identified by gas chromatography (GC) and gas chromatography-mass spectrometry (GC-MS) methods. By comparing the results obtained from this experiment in different treatments, contain 5 important essential oil components, the main compounds essential oil was, (Neral, Geraniol, Geranial, Neryl Acetate and Geranial Acetate) That Effects on plants. The highest percentage of Neral was 0.55% That treated with a concentration of 50 mg/l titanium nanoparticles and 100 mM salinity, and the highest percentage of Geraniol was 7/51%, in 100 mg/l titanium dioxid NPS and control salinity condition. The highest percentage of Geranial (E-citral) was 44.5% in 50 mg/l titanium nanoparticles and 0 mM salinity. And the highest percentage of Geranyl acetate was 22.01% and Neryl acetate 0.92, in un-titanium dioxide NPS treatment and 50 Mm salinity compared to the control. in plants treated with titanium dioxide, NPs 100 and 50% salinity were more than other treatments and controls, such as menthol and Linalool (0/71%,1/46%) respectively, therefore, in the present research, titanium dioxide nanoparticles treatment in low concentrations improved the harmful effects of salinity stress and resistance of *Dracocephalum moldavica L* plant and increased yield essential oil by effects on essential oil components.

**Keywords:** Abiotic stress, *Dracocephalum moldavica L.*, Essential oil, GC-MS, Nanoparticle
In this study, ZnO nanoparticles on some growth and developmental indices of the green alga *Chlorella vulgaris* were investigated. Under the influence of nanomaterials, oxidative stress is produced, resulting in the production of reactive oxygen species (ROS). Increasing activity of ascorbate peroxidase (APX) and superoxide dismutase (SOD) and decreasing activity of catalase (CAT) as antioxidant enzymes in treated algae by nanoparticles were observed compared to control. Moreover, the study of growth changes in different concentrations of nanoparticles was carried out and by the increasing concentration of nanoparticles, growth of alga was decreased. By the increasing concentration of nanoparticles, the total phenol and flavonoid content and chlorophyll a, b, total chlorophyll content, as well as carotenoids, decreased in treated samples compared to control. Also, using scanning electron microscope (SEM), the effect of nanoparticles on the shape of algae was observed.

**Keywords:** Nanoparticles, Cellular toxicity, Photosynthetic pigments, Antioxidant enzymes, *Chlorella vulgaris*

PEG/PLA diblock-copolymer nanocomposites: the antineoplastic drug delivery vehicles against human ovarian cancer *in vitro*

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In this study, PEG/PLA nanocomposites were synthesized by chemical precipitation and used as cisplatin (CP) and herbal extract carriers for their controlled release against human ovarian cancer cells. Extraction of watermelon seeds (WS) and Persian olives (PO) was done by the Soxhlet method and the presence of extract's phytochemicals was studied using the colorimetric method. CP, WS, and PO loading onto PEG/PLA nanocomposites was done by oil-in-water emulsion method. The obtained copolymers were evaluated by FTIR, UV/Vis absorption by Nanodrop, electron microscopy. The drug loading efficiency and the drug release profiles (dialysis) were measured in vitro by a Nanodrop at 215-800 nm, which showed a long-term release mechanism. CP release profile by dialysis showed 97.37% after 72h, long-term release-mechanism and stability. MTT analysis results have shown that the prepared nanocomposite system showed a significant effect on apoptotic induction in OVCAR-3 cells. The highest % cell death rate after administration of WS and PO loaded PEG/PLA (~99.5%, p<0.001) and the combination of CP-WS-PEG/PLA and PO-CP-PEG/PLA (~97.5%, p<0.001). Our colorimetric results have shown that our WS and PO extracts contained phenols, flavonoids, triterpenoids, and vitamins C, etc., which caused a considerable amount of cytotoxicity in cancer cells. The CP release from PEG/PLA showed a dose- and concentration-dependent manner.

**Keywords:** Nanocomposite, Pharmaceutical carriers, PEG/PLA, Cisplatin, Herbal extracts, Ovarian cancer
Biochemical and morphological study of graphene oxide on soybean

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Nanomaterials are at least one dimension in size from 1 to 100 nm. The nanoparticles made can be introduced into the environment through atmospheric emission, household sewage, agriculture and accidental release during production, transportation or decontamination. The same concerns have raised concerns about their potentially harmful effects on the health of living organisms and the environment. Graphene oxide carbon nanotubes are one of the most important members of the graphene family. In recent years, the impact of these nanotubes on food products has been studied by researchers. The soybean or oilseed bean, called Glycine max L., is a plant that is often referred to as “miraculous candy”. In this paper, a variety of soybeans called Koars in 5 levels (0, 100, 200, 400, 800, 1600 mg/L) of nanoparticle graphene oxide concentration in a completely randomized design with three replications in the vegetative stage, biochemically and morphologically, was evaluated. The results indicated an increase in growth parameters including seedling length, root length, fresh weight and root and shoot dry weight and leaf area at 400 mg/L concentration. The results of biochemical tests revealed significant changes in the catalase-peroxidase SOD, H2O2. Anatomical results showed the presence of nanoparticles on the surface of the root and skin, indicating the absorption of this nanoparticle. However, the presence of these nanoparticles was not observed in the leaves. According to the results of this research, the positive effects of graphene oxide on all the dimensions of soybean plants can be found.

Keywords: Graphene oxide, Soybean, Nanomaterials

Effect of graphene oxide nanoparticles on Soybean germination

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The presence of nanoparticles in the medium and the possibility of their absorption and displacement by the plants is the motive of this research. Graphene oxide carbon nanotube is one of the most important members of the Graphene family. In recent years, the effect of these nanotubes on food products has been studied by researchers. In this research, seeds of soybean variety Koars were evaluated at 6 levels (0, 100, 200, 400, 800, 1600 mg/L) of Graphene oxide nanoparticles in a completely randomized design with three replications. Microscopic examinations revealed the presence of these nanoparticles in the skin of the seeds and in the seedlings in the epidermis and the outer layers of the root hair. Incremental growth parameters were observed including root length, fresh weight and root dry weight under 400mg/l treatment. These findings can help improve the quality of fertilizers using nanoparticles.

Keywords: Graphene oxide nanoparticles, Soybean, Germination, Seedlings
Effect of biologically synthesized silver nanoparticles on the phenol and flavonoid parameters of *Rosmarinus officinalis* L.

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Today, limited information is available about nanomaterials effects on human health, in spite of its widespread use. Since the particles in nanoscale have new properties, the silver particles exhibit significant physicochemical properties and biologically activity because of small size properties. These distinctive characteristics make silver nanoparticles more practical in medical programs including antibacterial, antifungal, antiviral, anti-inflammatory and destroying cancer cells. Also, the silver nanoparticles have been more considered by researchers due to their high surface to volume ratio. *Rosmarinus officinalis* L. belongs to a lamiaceae family, has antibacterial and anticancer properties. This plant has anti-oxidant properties, because of phenolic diterpenes in their leaves extraction. Polyphenols are one of the largest natural anti-oxidant in plant kingdoms. Usage of polyphenols in plant led to decrease of disease like cancer. *Rosmarinus officinalis* L. plant, because of polyphenols such as carnosic acid has maximum anti-oxidant activity. This experiment was arranged in a factorial based on a completely randomized design with three replications in a research greenhouse of Sana Institute of Higher Education. Purchased seedlings from the agricultural greenhouse of Karaj after planting in soil and assuring from the successful establishment in a pot and the emergence of 4 to 6 leaves of the plant, treated with biologically synthesized silver nanoparticles (prepared using sage ethanolic extraction and silver nitrate) (40 mM) applied on the plant by spray solution. Pods containing sage transported to a greenhouse with 16 hours of light (8000 to 10000 Lux light) and 8 hours of darkness, relative humidity of 60% and an average temperature of 5 ± 25°C. The nutritional requirements of the plant were provided by a Hoagland nutrition solution. Total phenolic and flavonoid compounds were measured after performing the above steps. The results illustrated that the plant samples treated with synthesized-nanoparticles had 78% phenol and 82% flavonoids more than control samples. From the above results, it can be concluded that synthesized nanoparticles by plant extract can act as an elicitor in increasing secondary metabolites and subsequently can be used to produce more of these important compounds with medicinal attributes followed by a new pathway will be identified to control diseases.

**Keywords:** *Rosmarinus officinalis* L., Biologically synthesized silver nanoparticles, Phenol, Flavonoid

The effects of nano perlite on the unicellular *Haematococcus pluvialis* algae

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Algae, as the organisms producing food in aquatic ecosystems, respond to the chemical composition of the aquatic environment, like other aquatic organisms, which in turn could change their biochemistry, physiology and morphology characters. Nano-perlites are one of the common nanoparticles, which are used to remove heavy metals from the water. In current study, we investigated the various effects of nano-perlites on *Haematococcus pluvialis* algae. The results showed that nano-perlite significantly reduced the growth rate of algae in different concentrations. Also, the ratio of chlorophyll *a* to chlorophyll *b* and auxiliary pigments content were increased and the efficiency of photosystem II reaction center was reduced partially. The obtained results proved the toxic effects of this nanoparticle. The amount of astaxanthin was also reduced in treatment with nano-perlites due to a significant reduction in the amount of algae biomass. The results of volatile compounds detection by GC-MS analysis, indicated an increase in phenolic compounds compared to controls at 25, 50 and 100 mg / L treatments of nano-perlite.

**Keywords:** nano-perlites, *Haematococcus pluvialis* algae, Tension, nanoparticles
The new generation of therapeutic systems for cancers focusses on the concept of targeting and slowing the release of drugs to long-term the therapeutic efficiency and reduces the side effects which mainly originate from the high dosage of the drug in the blood. Today, there are many reports on the use of magnetic high surface area nanoparticles with the ability to respond to pH changes of environment. The use of these nanoparticles enables the targeted transfer of the drug to the target tissue. In the present study, we have developed a novel and efficient targeted anticancer drug delivery system based on the use of a magnetic core-shell structure of Fe$_3$O$_4$@CaAl LDH nanospheres as a carrier for L-Dopa in the treatment of Melanoma. The structural properties of Fe$_3$O$_4$@CaAl@L-Dopa were characterized using various techniques like XRD, SEM, TEM, EDX, FT-IR, VSM, TGA, XPS and BET. According to the obtained results, Fe$_3$O$_4$@CaAl@L-Dopa has a uniform core-shell structure with about 120 nm in average size. This hybrid nanocarrier has a high drug loading (52 wt.%) and drug efficiency (71%). Also, in this drug delivery system, the drug release was sensitive to pH changes. The amount of L-Dopa release from Fe$_3$O$_4$@CaAl LDH nanoparticles was much faster in lower pHs (98.5%) which indicate Fe$_3$O$_4$@CaAl@L-Dopa can be used efficiently in the cancerous cells. Therefore, this system can decrease the cytotoxic effects of L-Dopa by selective L-Dopa delivery. Furthermore, the cytotoxic investigation of L-Dopa and Fe$_3$O$_4$@CaAl@L-Dopa using the MTT assay on Mel-Rm cell lines shows the decrease in proliferation ratio of Mel-Rm cell lines as compared with the free L-Dopa and Fe$_3$O$_4$@CaAl LDH. Also, the values of IC50 for Fe$_3$O$_4$@CaAl@L-Dopa (3.8701 µM) was lower than the corresponding values for free L-Dopa (5.48 µM).

**Keywords:** L-Dopa, LDH, Magnetic nanoparticles, pH-responsive drug carriers, Melanoma cancer cells

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**Magnetic drug delivery system with a smart pH-responsive layered shell for controlled transportation of L-dopa**

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**Allivation anatomical and morphological responses okra (Hibiscus escoletus L.) seedlings to Multi wall carbon nanotubes treatment**

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Carbon nanotubes (CNTs) are known to have many unique physical and chemical properties. Because of these features of CNTs have been used in agriculture to increase the crop yield, mainly in the germination process and growth. In this experiment, okra seedlings were treated by 4 multi-walled carbon nanotubes (MWCNTs) (0, 50, 100 and 200 mg/lit). After treatments we measured effects of multi-walled carbon nanotubes (MWCNTs) on morphological and anatomical tissues (stem, root and leaf) characteristics of okra seedling plants. Results showed that growth parameters were increased under MWCNTs treatments. However these parameters were decreased by increase of MWCNTs concentration. In other hand, results of anatomical study showed that thickness of stem and root were affected by changes of cortex zone. This present study, showed that thickness of mesophylos layer and vascular region have been increased by low concentration of MWCNT, but these parameters have been decreased by high concentration. In this study indicated that moderate and high concentrations of MWCNTs treatments caused increase stomata index and decrease stomata length. Finally, it can be concluded that carbon nanotubes have a positive effect on low and medium treatments and in high concentrations have negative effects on okra seeds.

**Keywords:** Anatomy, Carbon Nano tube, Growth indices, Okra
Biosynthesis of gold nanoparticles by Evening primerose (*Oenothera biennis*) seed extract and optimization of synthesis conditions by Taguchi method

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The development of nanotechnology in the fields of medicine, technology and engineering, promote the optimal methods of production of nanomaterials. Today, organic and inorganic nanoparticles are commonly used in drug delivery systems, early diagnosis of diseases, control of agricultural pesticides and fertilizers, genetic engineering and industries. One of the best and most biocompatible methods to production of nanoparticles is through the bottom-to-up production with the help of effective herbal compounds. The Evening primrose medicinal plant, is able to synthesize and stabilize nanoparticles due to its active compounds, tannin, terpene, flavonoids, omega-6 fatty acids, vitamin E and phenolic compounds. The most important therapeutic effects of this plant are anti-inflammatory, anti-cancer, cholesterol-lowering, sedation, anti-asthma and anti-pertussis. In this study, 16 experiments with 4 factors in 4 different levels were designed with the use of the Minitab 17 software and the Taguchi experiment design method, for the purpose of the synthesis of gold nanoparticles in the appropriate size, by EP seed extracts. Then seed extracts prepared by ultrasonic-assisted extraction and GNPs were synthesized by adding chlorauric acid after adjusting the levels of each experiment. The size of synthesized GNPs, were determined by UV-visible spectrophotometry and DLS analyzes in the range of 1-7 nm. Data analysis results indicate that the appropriate levels of investigated factors are, concentration 200:400 μl gold chloride to seed extract, pH=10, temperature 70 C and time 24 hour, and also time and pH are the most effective factors in biosynthesis of suitable and stable gold nanoparticles.

**Keywords:** Gold nanoparticles, Bionanotechnology, *Oenothera biennis*, Taguchi method, Biosynthesis, Biocompatible

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Biosynthesis of gold nanoparticles by Lavender (*Lavandula angustifolia*) seed extract

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The tendency to use and produce nanoparticles is growing daily, due to their industrial and medical properties. Nonetheless, metallic nanoparticles, especially gold nanoparticles, are widely used in the fields of cancer treatment, pharmacy, targeted transfer of biological compounds and cosmetics production. The use of plant tissues has attracted scientists from among the affordable and low-risk environment methods of producing nanoparticles in suitable size and scale. In this study, lavender seed extract was prepared by the heat-soaking method. Then gold nanoparticles were produced by combining the extract with gold nitrate and dilution with deionized water. Also, effective factors such as pH, temperature and time are studied by UV-visible spectrophotometry and the optimal synthesis conditions are pH = 9.5, temperature 35 °C, and time of 30 minutes. By increasing the temperature and time, size and amount of nanoparticle synthesis increased and no nanoparticles were synthesized in the acidic pH. Then the size and structure of produced nanoparticles were investigated by DLS and TEM analysis. The results showed that the gold nanoparticles produced with a crystalline, spherical and homogeneous structure with an average diameter of 50 nm.

**Keywords:** Biosynthesis, Bionanotechnology, *Lavandula angustifolia*, Seed extract, Gold nanoparticles
Effect of titanium dioxide nanoparticle spraying on anatomical properties of *Aptenia cordifolia*

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Nanoparticles are atomic or molecular collections with a minimum dimension of 1 to 100 nanometers, which have different physicochemical properties compared to their normal fabric. Today, the widespread use of these nanoparticles causes in their release into the environment. The plants as major members of the ecosystem are also affected by these nanoparticles. Due to the photocatalytic features of Titanium dioxide, it is one of the most widely used nanoparticles. In this study, the effect of titanium dioxide spraying on *Aptenia cordifolia*, according to completely randomized design has been examined. This examination has been operated with three replications in four concentrations (0, 0.01, 0.03 and 0.05%) of nanoparticles in different stages of the plant development. The results indicated that the number of stomata on the dorsal epidermis of the leaf did not influenced of nanoparticles, but, the number of stomata on the ventral epidermis has been increased in treated plants. The high concentration of titanium dioxide treatment caused some anatomical changes, such as increasing the diameter of the vascular apertures in the root and reducing the diameter of the vascular cylindrical region in the stem of the treated plants compared to the control plant. The diameter of vascular craters also decreased in plants under treatment with 0.05% nanoparticles compared with other plants. It seems that, decreasing diameter of vessels is the probability of increasing wall thickness due to lignin precipitation.

**Keywords:** Titanium dioxide, *Aptenia cordifolia*, Anatomical properties

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Optimization of nanoliposomes encapsulation Avastin using an experimental design

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Ocular drug delivery is one the most challenging fields for pharmacologists. Avastin (Bevacizumab) a synthetic and whole humanized monoclonal antibody which binds all isoforms of vascular endothelial growth factor (VEGF), is used for the treatment of different ocular diseases which are caused by abnormal neovascularization. The short half-life of the drug in vitreous necessitates frequent drug administration so employing controlled-release drug delivery systems such as nanoliposomes has been particularly noticed. The goal in the present experiment was optimizing and evaluating liposomal encapsulation efficiency of Avastin in nanoliposome. Using the dehydration-rehydration Method, Bevacizumab (Avastin) was encapsulated into liposomes (Avs-NLP). Dynamic light scattering analyzing revealed that size of Avs-NLP was 139.9 nm. The effect of some independent variables including DPPC (1, 2-dipalmitoyl-sn-glycero-phosphocholine), cholesterol, and Avastin concentrations and freeze/thawing cycles on entrapment efficiency % (DEE) were evaluated utilizing response surface methodology (RSM). Optimal conditions for the maximum DEE (39.9%) was obtained with 1.6 mg Avastin, 2 mg DPPC, 0.37mg cholesterol and 7 freeze/thawing cycles. The optimization of the formulations for valuable and expensive drugs is essential to have the maximum encapsulation efficiency and the minimum experiments.

**Keywords:** Avastin (Bevacizumab), Nanoliposome, Ocular drug delivery
Effect of humic acid and nano calcium chelate on some growth and biochemical characteristics of *Gerbera jamesonii*

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In order to investigate the effect of humic acid and nano calcium chelate on growth and biochemical characteristics of *Gerbera*, an experiment as a completely randomized design with two factors with three replications in hydroponic conditions and in the pot was performed. The first factor was included: humic acid in 4 concentrations of 0 (control), 500, 1000 and 2000 mg/l as a drench and the second factor: nano calcium chelate at 4 concentrations of 0 (control), 1, 2 and 3 gr/l as a foliar application. Indices such as leaf number, area and length, total leaf chlorophyll and leaf carotenoid were measured. The results of the analysis showed that humic acid at a concentration of 2000 mg/l and a concentration of 3 g/l nano calcium chelate significantly increased the number of leaves, leaf length, and leaf area compared to the control. Also humic acid (500 mg/l) and nano calcium chelate (2 g/l) caused a significant increase in total chlorophyll content and carotenoid in comparison to control.

**Keywords:** Chlorophyll, *Gerbera*, Growth characteristics, Nutrition

Molecular dynamics study on the stability of peptide nanotubes placed in POPC phospholipid bilayer

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The self-assembly and stacking of cyclic peptides form cyclic peptide nanotubes (CPNT). These nanotubes are able to transfer many substances such as drugs through the cell membrane. For this reason, they must maintain a stable structure in the phospholipid bilayer. Up to now the transfer of different molecules such as water, ions, glucose, glutamic acid, 5FU anticancer drug and so on through peptide nanotubes have been investigated theoretically and experimentally. In this study structure and stability of CPNT [(D-Ala-Glu-D-Ala-Gln)₃] placed in POPC (1-Palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine) bilayer is analyzed after 40ns molecular dynamic simulation. This nanotube has been previously synthesized experimentally. This analysis indicates the role of the non-bonded interaction between the side and main chains of cyclic peptide rings and their phospholipid environment, besides the role of non-bonded electrostatic interactions between neighboring cyclic peptide rings in CPNT’s stability. In addition, the number of rings placed in the lipid bilayer along with their arrangement so that similar charges wouldn’t sit on top of each other effect CPNT’s stability. The result of this study can help further experimental investigations in this field.

**Keywords:** Cyclic peptide nanotube, POPC membrane, Molecular dynamic simulation
Growth and reproductive responses of tuberose (*Polianthes tuberosa* L.) to silica and silica nanoparticles nutrition in greenhouse condition

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Tuberose (*Plonianthes tuberosa* L.), a member of Agavaceae family is an ornamental bulbous plant and one of the most important cut flowers in tropical and subtropical areas. The cultivation and growing of this plant take place by the bulb. One of the most effective cases on the growth and the quality of tuberose is appropriate nutrition. This experiment was conducted as a completely randomized design based factorial design with three factors [types of silica in two levels (normal and nanoparticle), nutrition method in two levels (soluble and foliar application), and concentration in three levels (0, 200, 400 mM)] and five replications (each replication consisted of a pot containing two bulbs) in greenhouse condition with soil medium. The results showed that the nano-silica treatment at 200 mM concentration with soluble method had the best effect on some of the traits such as leaf number, the fresh and dry weight of flowering stem, flower stem length, and diameter. Also, the spraying of nano silica at 400 mM concentration had the best effect on the fresh and dry weight of the bulb. Also, the foliar application of 400mM nano silica had the best effect on the fresh and dry weight of the bulb. In addition, the effect of the application of soluble silica at 200 mM concentration on the traits of leaf number, fresh and dry weight of the stem, plant height and a number of florets was lower than control.  

**Keywords:** Bulb, Silica, Cut flower, Bulbous plants, Nanoparticle

Green synthesis of copper nanoparticles by *Datura stramonium*

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Nanotechnology is the application of material science at the molecular level. The use of plants for the synthesis of nanoparticles has been a major concern for reducing costs and adapting to the environment, so much research is being done on the green synthesis of nanoparticles by plants. In this study, the synthesis of green nanoparticles of copper was investigated using *Datura stramonium* ethanolic extract. Ethanolic extracts from leaves were prepared by maceration and Soxhlet methods. The extracts were added to 1 mM copper sulfate solution and the production of copper nanoparticles was investigated using UV-Vis, XRD, FTIR, and TEM. The results of the spectrometry showed a strong peak at 654.9 nm, indicating the production of copper nanoparticles. Analyzes XRD, FTIR, also confirmed the formation of copper nanoparticles. TEM sedimentation of electron microscopy images shows that nanoparticles are mainly spherical with an average size of 42 nm. The results show that copper nanoparticles produced by *Datura stramonium* leave ethanolic extract to have good size, environmentally friendly and without the use of inappropriate chemicals and can use for various studies.

**Keywords:** Nanoparticle, Ethanol extract, Green synthesis, *Datura stramonium*
Lisinopril loaded solid lipid nanoparticles; evaluation of the effect of chitosan coating on their invitro characteristics

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Lisinopril belongs to a family of anti-hypertensive drugs named as angiotensin converting enzyme inhibitors (ACE inhibitors). This drug is used as hypertension-related disorders such as systemic hypertension and glaucoma via multiple extra-vascular administration routes. The bioavailability of lisinopril is poor and restricted to maximum 25% and thus many studies are conducted to increase its bioavailability. Application of nanotechnology is the one of the most strategies which were used to overcome the mentioned problem. In this study, it was tried to overcome the mentioned limitation though application of chitosan-coated solid lipid nanoparticles (SLNs). SLNs were prepared through solvent diffusion technique; here a relatively small volume of lipid phase (stearic acid, glyceryl mono stearate and stabilizer in ethanol) is diffused in a relatively large volume of aqueous phase at stirring condition and higher temperature. Invitro characteristics of the formed nanoparticles were determined via different analysis such as loading calculations, SEM image, DSC, FTIR analysis and also drawing the release profile of the drug. The optimized particle sizes of drug loaded SLNs and chitosan-coated SLNs were 105±6 nm and 142±11 nm respectively. The bigger particle size of last sample was due to chitosan-coating. The zeta potentials of these nanoparticles were calculated as -52mV and +23mV, respectively. The high negative sign of the SLNs was due to the anionic nature of the lipid fraction of its constituents and the positive value of the last sample was related to coating with cationic chains of chitosan polymer. SEM images of these nanoparticles showed nanometer scale of their sizes and also differences in their surface morphology due to branched coating of SLNs surfaces. These findings confirm the formation of such chitosan-coted SLNs. The entrapment efficiency of these two nanocarriers were 18 and 34 %, respectively. The coating of hydrophobic SLN structure with a hydrophilic coat as chitosan could yield a more adjutant situation to encapsulate hydrophilic drugs. The data from DSC and FTIR analysis showed no unwanted interactions between lisinopril and nanocarriers. Although the electrostatic interactions of the negatively charged surface of SLNs and positively charged chitosan chains were proved via changes in their FTIR spectrums. Drawing of release profile of lisinopril from the uncoated and coated SLNs showed that where the 80 % of the drug was depleted in first 3 hours of release time in uncoated SLNs, this interval was elongated to 10 hours in chitosan-coated SLNs. The results of this study confirms the capability of chitosan-coating onto the surface of SLNs in order to control the drug release of a hydrophilic drug; lisinopril at sustain release manner.

Keywords: Lisinopril, Solid lipid nanoparticles (SLN), Chitosan, Burst release, Sustain release

Evaluation of the apoptotic effect of nano-based compound of gemini curcumin on AGS gastric cancer cell line

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It has been shown that curcumin has effective anti-cancer properties. However, the absorption efficacy of curcumin is too low to have significant results in therapy. To overcome this problem, we first improved the bioavailability of curcumin by making a Nano-based compound of Gemini Curcumin and then studied the effects of this compound on cell viability and apoptosis. Human gastric carcinoma AGS cell lines were treated with Gemini Curcumin (GC), and cell viability was evaluated using the MTT assay. Cell cycle analysis was assessed by flow cytometry and also the apoptotic potential of GC was measured using Fluorescence microscope and Annexin-FITC/PI Apoptosis Assay. Expression levels of some genes involved in apoptosis pathways such as BAX and BCL2 were analyzed by qPCR and Western blotting. GC affected the cell viability of AGS cells in a dose- and time-dependent manner with IC50 values of 15 μM at 72 h of treatment. Our results showed that treatment with GC increases BAX/BCL2 ratio and induces significant G2/M cell cycle arrest in AGS cells. Gemini curcumin induces G2/M-phase arrest in human gastric cancer cells, which highlights its potent anti-cancer activity and potential application in gastric cancer therapy.

Keywords: Nano curcumin, AGS cell line, Gastric cancer, BAX/BCL2
Physiological effects of copper oxide nanoparticles on tobacco cell culture

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Copper oxide nanoparticles (CuO NPs) appear to be promising agents for application in the agriculture and food industries, but information concerning the response of plants to interaction with nanomaterial is limited. Accordingly, a cell biology approach was applied to evaluate the physiological responses of tobacco cell cultures to CuO NPs. The cells were found to increase the activity of antioxidant enzymes (catalase, peroxidase, and superoxide dismutase) after 48 hours of exposure to different concentrations of CuO NPs (0, 10, 25, 75, and 100 mg L⁻¹). The production of reactive oxygen species and malondialdehyde in a dose-dependent manner was also observed. Further, a significant reduction in total phenol and flavonoid contents was evident. The overall results suggest that exposure to CuO NPs caused significant physiological and biochemical level changes and oxidative stress in tobacco cells.

Keywords: Copper oxide nanoparticles, Physiological response, Tobacco cells, Toxicity

Photochemical synthesis of silver nanoparticles and evaluation of their toxicity on cell suspension culture of Nicotiana tabacum

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Silver nanoparticles (Ag NPs) are one of the extensively used nanomaterials in the industry because of their special physicochemical properties as well as antibacterial and anti-inflammatory activities. The Ag NPs released from these products could enter into the environments through numerous routes. Therefore, it is necessary to consider the potential environmental impact of Ag NPs, especially on plants. In this present study, Ag NPs were synthesized using photoreduction method and their potential toxicity on cell suspension cultures of Nicotiana tabacum was evaluated. The synthesized Ag NPs were characterized by using X-ray powder diffraction, scanning electron microscopy, and dynamic light scattering techniques. Cell viability was assessed using triphenyl-tetrazolium chloride (TTC) assay. Subsequently, the dry weight of the samples was measured as growth characteristic of cell cultures. The average particle size of Ag NPs was about 40 nm. A significant decline in viability observed when cells were treated with Ag NPs after 2 day, which was approximately 95, 19, 11, 2 and 3 % for 5, 10, 15, 20 and 25 mg L⁻¹ of Ag NPs, respectively. In the case of the lowest and the highest concentrations (5 and 25 mg L⁻¹), the final dry weight was 72 and 44 % compared to the control. Our results suggest that Ag NPs are highly toxic to N. tabacum cells.

Keywords: Nicotiana tabacum, Oxidative stress, Silver nanoparticles, Toxicity
Effects of fulvic acid and iron nano chelate on some morphological and biochemical characteristics of *Gerbera jamesonii* cv. Dune

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In order to investigate the effect of fulvic acid and iron nano chelate on growth and biochemical characteristics of Gerbera, (*Gerbera jamesonii* cv. Dune) an experiment as completely randomized design with two factors fulvic acid with 4 concentrations of 0 (control), 50, 100 and 250 mg/l as drench and iron nano chelate at 4 concentrations of 0 (control), 1, 2 and 4 gr/l as foliar application, with three replications in greenhouse conditions in pot and in hydroponic conditions was performed. The result of this study showed that interaction effects of fulvic acid and iron nano chelate increased chlorophyll index, leaf length, the number of leaves and total dry weight (stem and head of flower) compared to control. Stem fresh weight and stem diameter were not affected by these treatments. **Keywords:** Chlorophyll, Gerbera, Growth characteristics, Nutrition

Using MoS₂ nanoparticles in breast cancer treatment

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It is possible to target the hormone receptors and disrupt the growth of breast cancer cells. Although negative-triple breast cancer lacking three types of estrogen, progesterone and epidermal growth factor receptors, they express both androgenic and vitamin D receptors. Targeting these receptors simultaneously can be an effective therapeutic strategy for this cancer. In this regard, the effect of MoS₂ nanoparticle and its toxicity was evaluated on human breast cancer cell lines (MCF7), (MDA-MB-468) and normal cell (PBD-2-Hb). The toxicity effect of MoS₂ was determined on cancerous and normal cells at concentrations of 5, 10, 20, 35 and 50 (μg/ml) for 24 and 48 hours using the MTT test. The obtained IC₅₀ at 35 (μg/ml) concentration for MCF7 cell line in 24 and 48 hours was 96.9 ± 1.09 and 87.03 ± 1.25, respectively and for the MDA-MB-468 cell line, it was 73.61 ± 1.21 and 64.6 ± 1.48 respectively. It was found that MoS₂ at different concentrations does don’t have a toxic effect on normal cells, but it can be considered as an anticancer agent on breast cancer cells. Therefore, the combination of this synthetic MoS₂ and chemotherapy treatment can be effective in eliminating breast cancer cells. **Keywords:** Breast cancer, Estrogen receptor, Androgen receptors, MoS₂
Toxicity effect of green synthesis of silver nanoparticles on male mice liver
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Concerns have arisen about the health and environmental effects of the growing commercial use of silver nanoparticles. However, the toxic mechanisms and target tissues of silver nanoparticles have not been well defined. In this study, silver nanoparticles were green synthesized using Eucalyptus extract, then we examined the tissue toxicity of mice after oral administration of silver nanoparticles at three concentrations of 20, 50 and 100 ppm. Colloidal nanosilver was mixed with mouse food for 30 days, respectively. The hematoxylin-eosin staining studies on liver tissue sections in the group receiving 100 ppm silver nanoparticles showed severe damage to liver hepatocytes such as swelling and inflammation and disturbing the liver function. The result of the cytotoxicity test (MTT) showed cytotoxicity at concentrations of 20 ppm and above. According to this result, it can be concluded that the application of silver nanoparticles in high concentrations need to further research for its toxicity effects on organs and blood factors.

Keywords: Silver nanoparticle, Toxicity, Liver, Green synthesis, Mice

Effects of Silver Nanoparticles on Tissue Culture of Aloe vera (Aloe barbadensis)
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Aloe (Aloe barbadensis) is one of the most valuable economic and medicinal plants. The natural reproduction of this native Iranian plant is slow, it can not meet the needs of the country's pharmaceutical industry. In this study, the effects of silver nanoparticles on plantlet propagation in vitro of Aloe vera (Aloe barbadensis) were investigated. Aloe vera meristem was cultured on MS medium (Murashig, Skoog, 1962) containing Benzyol amino-purine (3 mg/l) and Indol Acetic Acid (2 mg/l) hormones, and morphological traits (stem length, Number of stems, root length and number of roots). In vitro plantlets were investigated in treatment of silver nanoparticles with five levels including zero (0, (control), 5, 10, 15, 20 mg/l) during 2 subcultures. Statistical analysis of the data was performed using one-way ANOVA at the probability level of 0.05 with SPSS software (Ver.22). The average stem length (8 cm) and proliferation rate (5 plantlets) regenerated from the Aloe vera meristem. The regenerated plantlets in the treatment of silver nanoparticles had the highest mean shoot length in the treatment of 5 mg/l with a probability level of 0.05. Reduction of number of stems in treatments of silver nanoparticles decreased compared to control (0 mg/l) and the highest. The length and number of roots in the treatment of 15 mg/l were more than 0.05 in the control treatment. Different concentrations of silver nano particles caused the morphological changes of Aloe vera plantlets in vitro.

Keywords: Nanoparticles, Aloe vera plantlets
Improved cytotoxic effect of liposomal cinnamaldehyde on MDA-MB-231 breast cancer cell line

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Presently, the emergence of the herbal liposomal formulation as a kind of drug delivery system in cancer treatment is developed. Cinnamaldehyde (CA) as a natural phytochemical show potentially antiproliferative effect in cancer cells. Cinnamaldehyde encapsulated in a liposome to study its anticancer properties compared with free cinnamaldehyde on MDA-MB-231 breast cancer cells. In this way, liposomal cinnamaldehyde was prepared by thin film hydration technique. The formulation is characterized by particle size, zeta potential, scanning electron microscopy (SEM) and Fourier transforms infrared spectroscopy (FTIR). MTT, flow cytometric assay and DAPI staining was applied to recognize the apoptotic rate and molecular mechanism, respectively. The liposomal cinnamaldehyde indicated average size of 59.64 nm with encapsulation efficiencies of > 80%. The IC₅₀ values for free cinnamaldehyde and liposomal cinnamaldehyde were estimated 43.61 ppm and 29.52 ppm, respectively. So our results demonstrated that encapsulation of CA increases the cytotoxic effect of CA. Remarkably, encapsulated CA leads to a decrease in the proliferative percentage. This research proposes that encapsulation of cinnamaldehyde significantly increases the anticancer effect in comparison with free CA.

Keywords: Cinnamaldehyde, Drug-delivery, Apoptosis, Liposome

Immobilization of urease enzyme on carbon nanotube and iron nanoparticles with comparison of their physiochemical features

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Urease enzyme is widely used in various industries and medicine. Increase the physicochemical properties of this enzyme can make it easier to use in various fields. Different nanomaterials are now used to immobilize enzymes. Carbon nanotubes and iron nanoparticles are among the most commonly used nanomaterials to immobilize enzymes that give them unique properties. In this study, the urease enzyme was extracted from red beans and then immobilized physically on carbon nanotubes and iron nanoparticles by covalent bonds. The temperature and magnetic properties of the enzyme after the immobilization were investigated, indicating an increase in thermal resistance after immobilization on carbon nanotubes. Also, enzyme activity was reduced by immobilization on each of nanomaterials. Finally, the fixation of the enzyme on iron nanoparticles can lead to multiple uses of the dialysis device. Also, at higher temperatures, the enzyme inactivity decreases.

Keywords: Enzyme, Immobilization, Urease, Carbon nanotube, Iron nanoparticles

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Keywords: Enzyme, Immobilization, Urease, Carbon nanotube, Iron nanoparticles
Anti-proliferative effects of the nano-oxali-palladium complex in comparison with oxali-palladium against human colon cancer cell line of HCT116

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Platinum (II) complexes are widely used in the treatment of several human malignant diseases, such as colon cancer. Previous reports have represented several disadvantages of cis-platin particularly related to general toxicity which leads to undesirable side effects and to drug resistance phenomena limit its clinical application. Recently, Pd(II) analogues were considered as anti-tumor drugs due to its structural similarities between Pd and Pt, as well as low toxicity. So, in the present study, we have investigated the anti-cancer activity and death induce ability of a newly synthesized nano-oxali-palladium using ginger extract and free oxali palladium by (MTT) assay after 24 and 48 hours incubation times. The 50% cytotoxic concentrations (Cc50) of nano-oxali-palladium were determined 32.5 and 18 µM after 24 and 48 hours incubation times, respectively. Also, 13 and 18 mg/ml of ginger extract respectively. On the other hand, this value was evaluated 600 and 433 µM for free oxali-palladium after 24 and 48 h incubation times. Also, 13 and 18 mg/ml of ginger extract after 24 and 48 hours incubation times, were induced 50% death in HCT116 colon cancer cell line. Above results show that the newly synthesized nano-oxali-palladium using ginger extract has a more cytotoxic effect on colon cancer cell line after 24 and 48 h incubation times. Also, the anticancer activity of ginger extracts used in nano-oxali-palladium increases the cytotoxicity of this compound.

Keywords: Green chemistry, Ginger extract, Nano-oxali-palladium, Colon cancer

Multi-walled Carbon Nanotubes Functionalized Effect on Expression of TXS and DBTNBT Genes Involved in Paclitaxel Biosynthetic Pathway in Taxus baccata

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The frequency of secondary metabolites is often less than 1% of total carbon in plants, therefore, biotechnologists have focused on cell culture, tissue and plant organ as an alternative way for production of secondary metabolites. Nowadays, using elicitors is one of the effective approaches to increase productivity of secondary metabolites in plants cell cultures. Abiotic elicitors contain metal ions, inorganic compounds, nanoparticles, etc. Nanoparticles are able to carry metabolites and transfer oligonucleotide molecules in plant cells. In this study, the effect of multi-walled carbon nanotube functionalized with carboxyl was evaluated on gene expression alteration of two genes involved in paclitaxel biosynthesis pathway in Taxus baccata cell suspension. Cell suspension was treated by multi-walled nanotube functionalized on 12th day of cells growth at Nano carbon concentrations of 100 and 250 mg l-1. The levels of gene expression were measured by qRT-PCR after 4 and 72 h in compare to control. Our results clearly showed that the expression level of TXS gene in upstream of paclitaxel biosynthetic pathway was increased significantly at 100 mg l-1about 2.1-fold 4 h after treatment. The expression level of this gene was also increased about 2.4 and 7.2-fold 72 h after the treatment at 100 and 250 mg l-1 concentrations, respectively. On the other hand the expression level of DBTNBT gene in downstream pathway was found to have increased about 6.9 and 7.8-fold 72 h after the treatment at 100 and 250 mg l-1 concentrations, respectively. From these results, we could conclude that the intracellular paclitaxel production have increased with metabolite passing through the pores in cell wall that created by multi-walled carbon nanotube.

Keywords: Taxus baccata, Paclitaxel, Multi-walled Carbon Nanotube Functionalized, TXS, DBTNBT
The impact of silicon and nano silicon on improving resistance of Isatis cappadocica to arsenite

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Arsenite, as a higher toxicity and its low concentrations causing stress and reducing of plant growth. The application of silicon in plants under stress can increase resistance. Some plant species, with an accumulation of high concentrations of arsenic, have the ability to refine contaminated areas. Previous studies have proven the hyperaccumulation of Isatis cappadocica, and hence the production of sufficient biomass is one of the important factors in the success of plants for purification, in this research, we tried to use silicone and nano silicon to produce more resistant seedlings against arsenite. For this purpose, the effects of silicon and nano silicon (25 mg/L) under arsenite treatment with concentrations of 0, 25 and 50 μM were investigated under hydroponic conditions. After harvest, growth parameters including the length and biomass of root and shoot were measured. The results showed that 50 μM arsenite treatment reduced the biomass of the plant, and the biomass of root and shoot decreased by 55% and 56%, respectively as compared to control. While the application of silicon and nano silicone improved the biomass and reduced the toxic effect of arsenite. Resulting of nano silicon application showed that the biomass of root and shoot increased by 50% and 20%, respectively as compared to 50 μM arsenite treatment. This positive effect was more pronounced in nano silicone treatment and resulted in a significant increase in the biomass of the root and the shoot. This increase in growth parameters, indicating an improvement in plant resistance to arsenite, is being treated with nano silicon.

Keywords: Arsenite, Silicon, Nanosilicon, Isatis cappadocica

Evaluation of essential oil and volatile compounds of basil (Ocimum basilicum) exposed to multi-wall carbon nanotubes under salinity stress in hydroponic culture

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Mass propagation of medicinal plants in hydroponic condition, especially in salt soils have a special importance. The experiment was conducted as a factorial and in a completely randomized block design. Hydroponically grown O. basilicum L. plants were exposed to 4 levels (0, 25, 50 and 100 mg/l) of Multi-walled carbon nanotubes treatment and 3 levels (0, 50 and 100 mM NaCl) of salt stress. Plants daily irrigated with quarter-strength Hoagland solution and Harvested inflorescences and leaves of each sample were air dried at room temperature. The hydro-distilled oils were analyzed by GC/MS. Fifty components were identified in the inflorescence and leaf essential oils of the basil plants. Salinity and MWCNTs treatment in low to moderate concentration increased the essential oil percentage. Results demonstrated the major pathway of the essential oil was Phenylpropanoids pathway that main constituents was estragole and in shikimic acid pathway the main constituents linalool was identified that were (60/48 % and 10/18%), respectively. α-cadinol, Caryophyllene, and Germacrene D were the other the most constituents that (4/26%, 4/92 and 6/56%) respectively. In total, considering the previous reports, it seems that the essential oil composition of hydroponically grown O. basilicum L. had volatile constituents probably with potential applicability in the pharmaceutical and food industries.

Keywords: Ocimum basilicum, Carbon nanotubes, Essential oil, Salinity
Effect of the zinc oxide nanoparticle foliar application on some growth characteristic and defense mechanism of *Dracocephalum moldavica* L. to salinity stress in hydroponic culture

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In this study, seeds of *Dracocephalum moldavica* were exposed to different concentrations (0, 25, 50 and 75 mg L\(^{-1}\)) of zinc oxide nanoparticle under different levels of salinity stress (0, 25, 50 and 100 mM NaCl). The experiment was conducted as a factorial and in a completely randomized block design. In each pot 5 plants hydroponically grown in containing growth chamber of cucupit and perlite with 2:1 ratio. They were daily irrigated with quarter-strength Hoagland solution. Germinated seeds were subsequently allowed to grow in the culture media to test the further response of the seedlings in terms of growth characteristics to the employed treatments. Seeds subjected to salinity showed a reduction in morphological properties in both levels of salt stress and in 100 mM NaCl level was the most. Growth characteristic such as the number of leave, lengths of shoots, fresh weight, number of nodes and rooting percentage of plants. However, the inclusion of ZnO-nanoparticle at the 25 mg L\(^{-1}\) concentration significantly alleviated the salinity stress-induced reduction in growth attributes and had the significant (p< 0.01) with 50 and 75 mg L\(^{-1}\) concentration of treatment. This happened due to increased water uptake, up-regulation of mechanisms involved in salt tolerance, and reduction in oxidative injury indices including Gpx, PPO, CAT, and APX. The improved plant performance under salinity stress was a consequence of changes in various antioxidant and also biosynthesis of proteins, phenolics, and specific metabolites such as proline. Results demonstrate that treatment by lowest concentration of ZnO-nanoparticle can induce tolerance in seedlings against low to High levels of salinity through enhancing water uptake and activating plant defense system.

**Keywords:** ZnO-nanoparticle, Medicinal plants, Hydroponic culture, Salt stress

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Biocompatibility and bioactivity study of nanofibrous electrospun chitosan containing copper doped nanoparticle bioglass

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Regeneration and repair of damaged tissue and organs in bioengineering have been named "tissue engineering". The tissue engineering depends on the biologic factors, the cells, and the biocompatible three-dimensional scaffolds that mimic the extracellular matrix. Fabricating artificial skins, skin substitutes and wound healing are one of the most important areas in tissue engineering. In this study, the first, copper-doped bioglass nanoparticles were prepared using the sol-gel method. Then, the chitosan nanofibers containing the copper-doped bioglass nanoparticles were fabricated with the electrospinning method. Morphology and physicochemical properties of the nano-fibers assessed by X-ray diffraction, infrared spectroscopy, and electron microscopy techniques. Antibacterial properties of the derived composite scaffold were investigated using the Kirby-Bauer test with gram-negative bacteria (*Escherichia coli*) and gram-positive (*Staphylococcus aureus*). Biocompatibility of composite scaffold was measured using the MTT method. The results of this study shown, the prepared scaffold has an appropriate fibers diameter with biocompatible and antibacterial properties, and hence potentially may be used as a wound dressing in skin tissue engineering.

**Keywords:** Tissue Engineering, Chitosan nanofibers, Copper, Wound-dressing, Biocompatibility, Antibacterial activity
Design and fabrication a local surface plasmon resonance based on nanobioprobe in order to *Shigella flexneri* bacteria detection

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*Shigella flexneri* bacteria is an important human pathogen which causes an acute bloody diarrhea known as shigellosis or bacillary dysentery. So, rapid detection of this pathogen in order to reduce the health risks of this pathogen is importance. Recently, biosensors due to a variety of reasons have replaced with many expensive diagnosis methods. In fact, gold nanoparticles due to high potential for conjugation with various biomacromolecules provide the suitable way in order to design the various nanobiosensors. Herein, an LSPR nanobioprobe has been developed based on gold nanoparticles-polyclonal antibody conjugated system in order to detect *Shigella flexneri* bacteria. In this study for the synthesis of nanobioprobe, specific polyclonal anti-*Shigella* protein has been conjugated at the surface of gold nanoparticles and this process has been confirmed by dynamic light scattering (DLS) measurement and observation of LSPR bond shift in UV/Vis spectrophotometer. After that, the sensitivity of the nanobioprobe was studied via monitoring the LSPR band alterations in the presence of different concentrations of *Shigella* Liquid medium (from 0-1000 CFU/ml). According to the results, the sensitivity of the nanobioprobe is about 100 CFU/ml whereas in comparison to the limit detection of ELISA tests is much more. So, LSPR based nanobioprobe has the probability of being applied for the rapid diagnosis of the wide variety of pathogenic agents.

**Keywords:** Local surface plasmon resonance (LSPR), Nanobioprobe, Gold nanoparticles, *Shigella flexneri*

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Preparation and evaluation of Nano - micronutrients of zinc and iron slow release and its effect on germination and physiological indices of wheat bread (*Triticum aestivum*)

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In recent years, research on the application of Nano science in various fields, including in the agricultural sector, has been considered. Considering the importance of evaluation and performance of nanoparticles in plant nutrition, in this study, the effect of iron and zinc nanoparticles on germination and seedling growth of native and modified wheat cultivars under laboratory conditions has been investigated. A factorial experiment was conducted in a completely randomized design with 5 replications at the Faculty of Life Sciences of Kharazmi University of Tehran. The investigated factors included different concentrations of iron nanoparticles (0.01 and 0.02 g/l), Nano-fertilizer (0.01 and 0.02 g/l) on two wheat cultivars including Bait (susceptible to Zinc and iron deficiency) and N-91-17 (modified with high iron and zinc absorption capacity). By applying iron and zinc treatment in experimental units, the basic characteristics of wheat growth including root length, stem, germination percentage, germination rate and seedling weight were evident. The results of germination test showed that Vigor index based on seedling length, seedling weight, stem fresh weights, root fresh weight and seedling length in iron Nano code and zinc compared to control in treatment of 0.20 g/l at probability level one The percentage is meaningful. In general, the results of the experiments showed that iron and zinc nanoparticles increase germination strength as well as more uniform seeding of wheat.

**Keywords:** Zinc Nanoparticles, Fertilizer Nanoparticles, Germination of Wheat
The interaction effects of ZnO nanoparticle application and salinity stress in two populations of *Trigonella* plants

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Fenugreek (*Trigonella foenum-graecum* L.) is a member of the family Fabaceae. It is an important crop cultivated in semi-arid regions of the world. Salinity is one of the most important abiotic factors limiting plant growth and yield. The purpose of this study was to investigate the effect of ZnO nanoparticle and salt stress on vegetative growth of two populations of trigonella plant. In 4 leafy stages, the interactive effects of ZnO nanoparticle solution (0, 1000 and 3000 mg/L) and salinity stress (0, 75, 150 and 225 mM NaCl) were studied. The effects of ZnO nanoparticle and salinity stress were investigated on some biochemical parameters such as lipid peroxidation, proline, protein and nonredundant carbohydrates content. Data were analyzed with Minitab. Specifically, increased salt concentration caused an increase in proline and reductant sugars contents in two populations, while the same factor caused an increase in MDA in mashhadian population. Interaction effects of 3000 mg/l ZnO and 225 mM NaCl caused the increase of proline and reductant sugars in Ardestanian population and the reduction of two mentioned parameters in Mashadian population. 225mM NaCl enhanced Car content and decreased Chlb content in Ardestanian population. ZnO nanoparticles improved these adverse effects. 3000 mg/l ZnO nanoparticle ameliorated reduction in protein caused by high salinity. We suggest that The effect of ZnO nanoparticles on reducing the effects of salinity stress depends on the concentration of nanoparticles used, as well as the level of resistance of Fenugreek populations under salinity stress.

**Keywords:** *Trigonella*, Salinity stress, ZnO nanoparticle
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