7th International Molecular Biology and Biotechnology Congress

25-27 April 2018, Konya, Turkey

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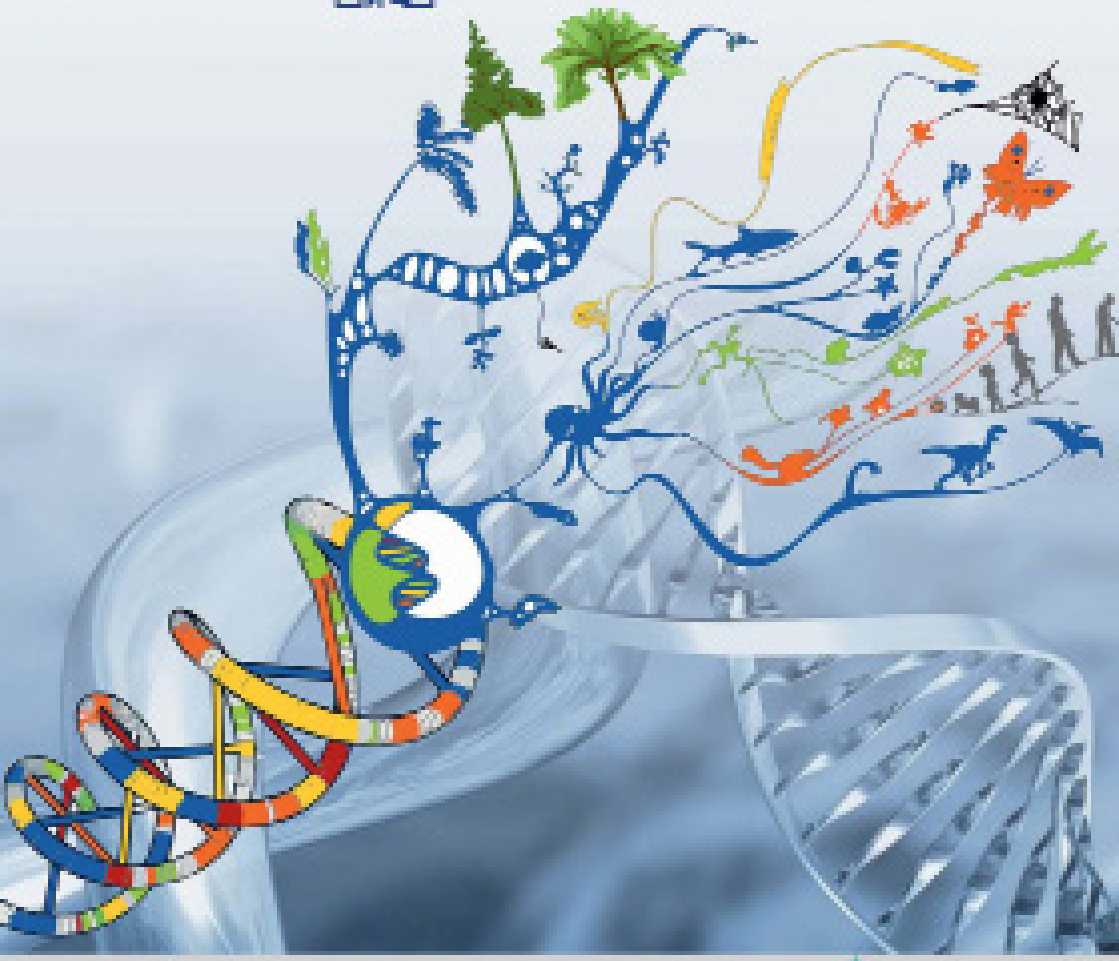

7th INTERNATIONAL MOLECULAR BIOLOGY and BIOTECHNOLOGY CONGRESS

ORAL ABSTRACT BOOK

KONYA

NECMETTİN ERBAKAN
UNIVERSITY

25-27 April 2018
Necmettin Erbakan University
MOLBIOTECH 2018

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April 25-27, 2018 - Konya

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CONTENTS

Preface

Dear colleagues,

It is my pleasure to welcome you to the 7th International Molecular Biology and Biotechnology Congress held in Konya, Turkey, from April 25 to 27, 2018. This congress is an interdisciplinary platform for the presentation of new and recent advances in researches in the fields of Molecular Biology and Biotechnology. Over 500 contributions from 15 different countries have been submitted and accepted for oral/poster presentations after peer review process.

Global population growth in the 21st century and limited natural resources present major threats and challenges. Recent advances in Molecular Biology and Biotechnology enable scientists and researchers to cope with the problems and to find out the solutions without threatening the natural resources and environment. This congress aims to bring scientists from international communities to highlight the recent advances and developments in Molecular Biology and Biotechnology and their application in Agriculture, Microbiology, Plant, Animal, Aquatic, Environment, Medicine and Industry.

Dear colleagues, it is our mutual purpose to find ways and methods for everyone to get benefit from the applications of Molecular Biology and Biotechnology in worldwide. Throughout the next three days, scientists from 15 different countries will discuss the problems and their solutions through the applications of Molecular Biology and Biotechnology.

I would like to thank to all the authors, reviewers, scientific committee, organizing committee, secretariat, session moderators and colleagues for their help in organizing this scientific event in Konya, Turkey. There is also a great thank for Necmettin Erbakan University for their support and collaboration.

Sincerely,

Prof. Dr. Mehmet KARATAS
Chairman of Congress

Dean of Faculty of Science
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Department of Biotechnology

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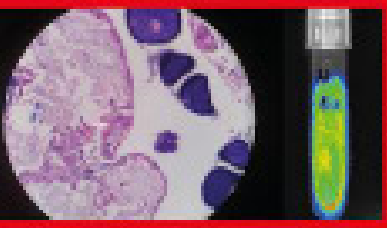


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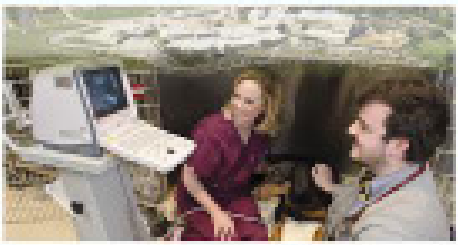


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Heavy metal induced gene expression in metal accumulator plants

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Abstract

Plants are sessile and are susceptible to biotic and abiotic stresses which occurs due to continuous change in climatic conditions. They respond to avoid these detrimental effects in a variety of different ways. In particular, heavy metal stress is one of the major problems in the developing countries which not only affects the plant health, but also severely disturb the whole ecosystem. Prolonged exposure to heavy metals such as cadmium, copper, chromium, lead, nickel, and zinc can cause deleterious health effects in humans. Molecular understanding of plant metal accumulation has numerous biotechnological implications also, the long term effects of which might not be yet known. The amount of heavy metal causing toxicity depends on the type of ion, ion concentration, plant species and stage of plant growth. Tolerance to metals is based on multiple mechanisms such as cell wall binding, active transport of ions into the vacuole and formation of complexes with organic acids or peptides. Here, one of the most important mechanisms for metal detoxification in plants appears to be chelation of metals by low molecular weight proteins such as metallothioneins and a family of peptide ligands, the phytochelatins. For example, glutathione (GSH), a precursor of phytochelatin synthesis, plays a key role not only in metal detoxification but also in protecting plant cells from other environmental stresses including oxidative stress. In the last decade, the tremendous developments in molecular biology and the success of genomics have highly encouraged studies in molecular genetics, mainly transcriptomics, for the identification of the functional genes implied in metal tolerance in plants. These studies have already succeeded in the identification of many genes that largely belong to the metal-homeostasis network. In this presentation I will describe recent advances in understanding the genetic and molecular basis of the metal induced gene expression in plants including the gene expression work which is being carried out in my laboratory on some metal accumulating plant species in *Brassicaceae* family.The heavy metal accumulating species *Brassica nigra, B. juncea* and an industrial crop plant *B. napus* have received attention due to its possible use for phytoremediation of heavy metal-polluted soils. I will discuss the strategies for exploring these immense and valuable genetic and biological resources for phytoremediation of heavy metal pollutants from the environment.

Keywords: Accumulator plants, transcriptomics, proteomics, metal transporters, phytoremediation

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Surface engineering with low-energy ion beams: from ultra-smooth surfaces to hierarchical nanostructures

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Abstract

Low-energy ion beam sputtering, i. e. the removal of atoms from a surface due to the impact of energetic ions or atoms, is an inherent part of numerous surface processing techniques. Beside the actual removal of material induced by atomic recoils and the sputtering of atoms from the surface this surface erosion process often results in a pronounced topography evolution, generally accomplished by a kinetic roughening of the surface. Typically, during ion sputtering, the surface of the solid is far from equilibrium and a variety of atomistic surface processes and mechanisms become effective. It is the complex interplay of these processes that either tends to roughen (e. g., by curvature dependent sputtering or incorporation of surface contaminations) or smoothen (e. g., by surface diffusion or viscous flow of surface atoms) the surface, which, finally, can result in a rich variety of surface topographies. Two prominent examples are the spontaneous formation of well-ordered ripple or dot pattern and the realization of experimental conditions where surface relaxation dominates and smooth surfaces are emerging. Both special cases are of high interest for many potential applications in nanotechnology. For instance, using broad beam ion sources with appropriate beam dimensions an alternative cost-efficient route exists to produce large-area nanostructured surfaces in a one-step process or for polishing of high quality optical surfaces, e. g., for smoothing of surfaces or interfaces of thin films. In this contribution the current status IOM related activities in the field of tailoring the topography of Si surfaces at the nanometer and micron scales by low-energy ion beams will be summarized. Starting from the diversity of pattern that can be formed, two special cases have been discussed (i) the formation of self-organized ripple and dot pattern and (ii) the smoothing of surfaces by using appropriate conditions of low-energy ion beam erosion. In this context, we briefly review potential processes believed to be responsible for pattern formation and smoothing in the low energy regime, especially for materials which are amorphous or become amorphous during the irradiation process. Concerning potential applications of ion beam, it will be demonstrated how ion beam smoothing (IBS) can be used for the finishing of high end optical surfaces with topography and roughness control down to the atomic scale. Finally, a short outlook will be given, especially for future work that is aimed to the combination of patterning by self-organization and conventional lithographic techniques in order to realize a better control of the self-organization process itself together with hierarchical structuring at different length and height scales.

Keywords: nanotechnology, ion beams, surface engineering, self-organization, polishing

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Investigation of apoptotic effect of *Achillea ketenoglui* extract on colorectal cancer cells

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Abstract

Induction of tumor cell death by the use of some phytochemicals that consumed through diet, and derived from medicinal plants opens up new horizons for cancer treatment researches. Our aim in this study is to assess apoptotic changes by way of implementing methanolic extract of the *Achillea ketenoglui* to the colorectal cancer cell line. In our previous study, we demonsrated the cytotoxic on HCT-116 and HT-29 cell lines. IC50 value is determined as 350 uM(48h) and 300 uM (24h) on HCT-116 and HT-29 cell lines, respectively. To examine the apoptotic effects of the extract, total RNAs were isolated from dose group and the control cells firstly, then cDNAs were synthesized. Expression profile of the apoptosis and cell cycle related target genes are determined by RT-qPCR. Apoptotic cells was determined with Annexin V kit. Protein expression were detected via western-blot method. According to the results, when the control group compared with the cells, it was determined that increase in the gene expressions of Bax, Caspase-3, Caspase-7, Caspase-8, Caspase-9, p53, p21, fas, pparg of dose group HCT 116 and HT-29 cells. Based on the obtained data, we believe that methanol extract of the A.ketenoglui induces apoptotic pathway.

Keywords: *Achillea Ketenoglui*, Extract, Apoptosis, Cancer

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The effect of thymoquinone on angiogenesis mechanism of agressive breast cancer cells

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Abstract

Thymoquinone obtained from black seed has been shown to have many positive effects on the organism such as antidiabetic, anti-inflammatory, antioxidant and antitumoral effects. Angiogenesis is one of the most important steps of malignancy and it is considered as a prerequisite for progression, proliferation and metastasis of tumor. Angiogenesis not only supply necessary nutrients for the tumor but also help them to invade the tissue and to metastasize to the distant sites. In this study, it was aimed to investigate the effect of thymoquinone on angiogenesis which is an important step of tumorigenic development.MD-MB231 breast cancer (highly metastatic and aggressive) cells were exposed to thymoquinone at different doses. Expression of angiogenesis- and metastasis-associated proteins, MMP-2 was evaluated by Western blott analysis. In addition, tubulogenesis , which is the last step of angiogenesis, was evaluated by tube formation assay, and invasion was analysed by matrigel invasion assay.It was seen that thymoquinone suppressed MMP-2 expression in aggressive breast cancer cells. In addition, it was observed that thymoquinone suppressed the tube formation and invasion ($p<0.05$). As a result of the study, it reduced the level of proteins involved in both angiogenesis and metastasis and disrupted the tubule formation. This is the first study in the literature indicating that thymoquinone inhibits angiogenesis and tubulogenesis in aggressive breast cancer cells. By inhibiting angiogenesis, thymoquinone may indirectly inhibit proliferation, invasion, and metastasis of tumors. For that reason, thymoquinone can be used as an effective agent in addition to the current treatments.

Keywords: Angiogenesis, breast cancer, thymoquinone, tubulogenesis

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Anti-cancer activity of methanol extracts of *Ranunculus Constantinopolitanus (Dc.) D'urv* on Mcf-7 breast cancer cell line

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Abstract

Ranunculaceae phytometabolites exhibit promising effects against cancer, many of which modulate signaling pathways that are key to cancer initiation and progression, and enhance the anticancer potential of clinical drugs while reducing their toxic side effects. In this study, it is aimed to investigate anticancer activity of flowers, body, leaf, and seed methanol extracts of *Ranunculus Constantinopolitanus (DC.) D'URV* in the MCF-7 breast cancer cell line and L-929 healthy adipose tissue cell line (mouse). Methanol extracts from flowers, body, leaf and seed of *R. Constantinopolitanus (DC.) D'URV* were prepared. The effects of the extracts on cell viability were determined by 3- (4,5-dimethylthiazol-2-yl) -2,5-diphenyltetrazolium bromide (MTT) method MCF-7 breast cancer cells and L-929 cell line were incubated for 96-well plates (100,000 cells / well) with different concentrations of extracts. MTT method was applied when incubation times were completed. The results were analyzed by the GraphPad Prism6 to determine the concentration values (IC50) of the extracts causing 50% mortality in MCF-7 breast cancer cells and L-929. Quantitative measurement of cell death was performed with Hoechst 33258 (HO; Sigma)/propidium iodide (PI; Sigma) staining, which allowed for apoptosis to distinguish necrosis. According to our experimental results, when the flower, body, leaf and seed methanol extracts of *R. Constantinopolitanus (DC.) D'URV* were compared with the L-929 cell, it was found that MCF-7 cell line viability significantly decreased with time and dose. As a result; flowers, body, leaf and seed methanol extracts of *R. Constantinopolitanus (DC.) D'URV* showed cytotoxic effects by reducing MCF-7 breast cancer cell line viability.

Keywords: *R. Constantinopolitanus (DC.) D'URV*, Extract, MCF-7, Hoechst 33258, Propidium iodide

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Toxicity and antioxidant properties of quercetin on 8305C Cells

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Abstract

Quercetin (3,5,7,3',4'-pentahydroxyflavon) is known as one of the best identified flavonoids and due to its strong antioxidant properties it is used in studies. Quercetin located in apples, onions, broccoli, strawberry, peas and green tea and a major phenolic components of plants. Thyroid cancer is a very common type of cancer and thyroid tumors constitute 1% of all tumors. In this study, we aimed to analyze antioxidant activity of Quercetin (QE) on 8305C (human thyroid anaplastic carcinoma) cells for the first time in the literature. Evaluation of cell viability was assessed using MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay. Following cell viability of QE, intracellular antioxidant activity and oxidative stress were establish by measuring malondialdehyde (MDA) levels, superoxide dismutase enzyme (SOD) activity and reduced glutation (GSH) levels. Briefly, 8305C (human thyroid anaplastic carcinoma) cells were seeded to 96 well plates and incubated at given cell culture conditions for 24 hours. After the incubation the medium was replaced with a fresh medium containing series of dilution of QE (0.5-1000 µg / mL) for 24, 48 and 72 hours. According to MDA, SOD and GSH levels the optimum doses of QE were found to inhibit oxidative damage in 8305C cells. Overall our results suggest that quercetin may could be used as a therapeutic support in human thyroid carcinoma cells.

Keywords: Antioxidant, Cytotoxicity, Human thyroid anaplastic carcinoma cells, Quercetin

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Effects of juglone and resveratrol fractions on Ehrlich ascites carcinoma cells

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Abstract

Recent studies demonstrated persuasive evidence that juglone and resveratrol have mighty anticarcinogenic features. However, there is insufficient data on the fractions of these molecules. This study was therefore undertaken to evaluate the anticarcinogenic effects of JRK (Metabolites of juglone and resveratrol by Kefir) against Ehrlich ascites carcinoma bearing mice. Lactobacillus bacterias in kefir yeast were grown in the cell culture (1,5x109/ml). Juglone and resveratrol (1:2) were added to the medium and exposed for 48 hours. The solution obtained after filtration was applied to Bap-c male mice (0.1 ml/day i.p.) given EAC (Ehrlich ascites carcinoma) cells throughout five days. Then; Bax, Caspase-6, 8 and 9 mRNA levels were measured by Real-Time PCR method in the ascites which isolated from the abdomen after decapitation. JRK solution significantly reduced EAC cells. Accordingly, the waist circumference has also decreased. JRK did not change Bax, Caspase-8 and 9 mRNA expressions but caused a tendency towards elevation. However, no change was found in Caspase-6 mRNA expression with JRK. On the other hand, according to the immunohistochemistry study results, reduced Bax levels with EAT were enhanced with JRK; reversely, enhanced Bcl2 levels with EAT were decreased with JRK. These findings indicate that JRK may be an alternative anticarcinogenic agent. In the advanced phase, higher dosing and more compherensive results are likely to benefit from clarifying the effects of JRK solution.

Keywords: Juglone, Resveratrol, Kefir, Ehrlich ascites carcinoma, Bax, Bcl2, Caspase

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Investigation of the effect of medicarpin on head and neck cancer cell line

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Abstract

Medicarpin, a natural pterocarpan, has various beneficial biological roles including inhibition of osteoclastogenesis, stimulation of bone regeneration and induction of apoptosis. Medicarpin has also been shown to inhibit cell growth in various tumor cells via the NF-κB pathway. Although there are several studies analyzing the effect of medicarpin in various tumors, there are limited information about expression and activation of signal pathways in head and neck cancer cells (HNSCC). Therefore, we have investigated the differential expressions of AKT, PDK1 and PTEN at the mRNA and protein levels in HNSCC by using qPCR and western blot techniques. Cell viability was assessed by MTT assay; and, IC50 value of medicarpin was determined as 80 M. The wound healing assay was used to examine cell migration and cell interactions; and, results were statistically significant ($p<0.05$). Expression levels of the PTEN ($p=0.000382$) and AKT ($p=0.000276$) were statistically significant in HNSCC cell line compared to control cells. In conclusion, medicarpin treatment may be inhibited cell growth via PTEN/AKT signal pathway in HNSCC.

Keywords: Medicarpin, HNSCC, PTEN/AKT pathway

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The relationship between idiopathic male infertility and the polymorphisms of genes involved in xenobiotic metabolism

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Abstract

Antioxidant gene polymorphisms are thought to be effective in individual susceptibility to male infertility. This study aimed to investigate the relationship between polymorphisms of cytochrome P450 1A2 (CYP1A2) 734 C→A, cytochrome P450 2D6 (CYP2D6) 1934 G→A, glutathione S-transferase M1 (GSTM1) null, glutathione S-transferase T1 (GSTT1) null and glutathione S-transferase P1 (GSTP1) Ile105Val that play role in xenobiotic mechanism and idiopathic male infertility. A total of 306 azoospermic or oligozoospermic idiopathic infertile men and 129 normozoospermic or fertile controls were included in the study. Peripheral blood samples from all participants were collected and genomic DNA was isolated by salting out method. Genotyping was performed using multiplex polymerase chain reaction (PCR) and PCR-restriction fragment length polymorphism methods. There was a statistically significant relationship between idiopathic male infertility and CYP2D6 GG genotype ($P < 0.001$). CYP1A2 AA genotype was slightly higher in the infertile group ($P = 0.056$). There was no association between idiopathic male infertility with GSTT1 null ($P = 0.068$), GSTM1 null ($P = 0.843$) and GSTP1 Ile105Val ($P = 0.192$) polymorphisms. GSTM1 null polymorphism was higher in azoospermic men ($P = 0.009$). Our results show that CYP2D6 polymorphism may play a role in idiopathic male infertility in our sample population.

Keywords: Cytochrome P450, glutathione S-transferase, idiopathic infertile

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Transcript resolution and functional characterization of a novel gammaherpesvirus long noncoding RNA (lncRNA)

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Abstract

Gammaherpesviruses, including Epstein-Barr virus (EBV) and Kaposi's sarcoma-associated herpesvirus (KSHV) are oncogenic viruses that establish lifelong infections in hosts and are associated with the development of lymphoproliferative diseases and lymphomas. Recent studies have shown that the majority of the mammalian genome is transcribed and gives rise to numerous long noncoding RNAs (lncRNAs). Likewise, it was shown that herpesviruses undergo pervasive transcription, including the expression of many uncharacterized putative lncRNAs. Murine gammaperherpesvirus 68 (MHV68) is a natural pathogen of rodents, and is related to EBV and KSHV, providing a highly tractable model for studies of gammaherpesvirus pathogenesis. Previous tiled microarray studies identified 30 novel "expressed genomic regions" (EGRs) of MHV68 transcription which were not predicted by previous canonical ORF-based annotation analyses. We sought to determine whether EGR1, which lies antisense to at least five MHV68-encoded miRNAs, encodes a bona fide lncRNA transcript. Using strand-specific northern blots, we identified a polyadenlyated nuclear transcript that overlaps the important latency-associated genes M2 and M3. Knockdown of this transcript (M3M2) in lytically infected cells using GapmeRs resulted in expression of M2, strongly suggesting M2-regulatory function of M3M2. Furthermore, infection with a mutant virus lacking two M3M2-antisense miRNA stem loops results in increased expression of M3M2, strongly suggesting its regulation by viral miRNAs. Thus, together these data demonstrate a tripartite relationship between lncRNA M3M2, antisense miRNA, and important latency gene M2. Based on the importance of M2 in latent infection, we hypothesize that this relationship may be a key control point for chronic infection and pathogenesis.

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Role of *Schizosaccharomyces pombe git1* gene in oxidative stress response

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Abstract

Glucose is the preferred energy and carbon source for many organisms. Glucose sensing and signal transduction in yeast is generally accomplished through a system of heterotrimeric G-protein and G protein-coupled cell surface receptors. Signal transduction pathway to cAMP/protein kinase is activated with glucose sensing. In this pathway, *Git1* is a C2 domain protein that is directly linked to adenylate cyclase and it is one of the 6 proteins required for the activation of adenylate cyclase. The 3' end of *git1* gene contains ''Mammalian uncoordinated homology 13, domain 2.'' It plays role membrane trafficking, exocytosis, vesicle secretion. This study aims to find out whether the *git1* gene, which is one of the genes involved in glucose signaling, and the 3' end of git1 gene, are related to oxidative stress response. In this study, *Schizosaccharomyces pombe* wild type (972h-) and git1- (*git1Δ*) mutant with *Escherichia coli DH5α* were used. Genomic DNA of S. pombe 972h- was used as a template to obtain *git1* and 3' deletion *git1* genes. These genes were cloned into plasmid pSGP572 containing the GFP reporter gene in the cloning site. The resulting recombinant vectors were transfected into super-efficient *E. coli DH5α* and then isolated. These isolated vectors were transformed into the *S. pombe git1Δ* mutant. Cell morphologies of transformants in the selective media were stained DAPI and then examined under confocal microscope. Transformants carrying recombinant plasmids were confirmed by GFP luminescence detected in a confocal microscope. There was no statistically significant difference in superoxide dismutase and catalase enzyme activities in H2O2 induced oxidative stress conditions in S. pombe recombinants and *S. pombe git1Δ* mutant. These results make think that cells probably select the different pathway alternatives in the stress response.

Keywords: *Schizosaccharomyces pombe*, *git1* gene, oxidative stress, glucose metabolism.

7th International
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25-27 April 2018, www.molbiotech.gen.tr

Application of CRISPR/Cas9 technique to the NRAS gene Q61K mutation in SK-MEL-30 skin cancer cell line

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Abstract

Malignant melanoma is a neoplasm of melanocytes or of the cells that develop from melanocytes. Although melanoma was once considered an uncommon disease, the annual incidence has increased dramatically over the past few decades, as have deaths from melanoma. The 3 Ras genes in humans (HRas, KRas, and NRas) are the most common oncogenes in human cancer; mutations that permanently activate Ras are found in 20% to 25% of all human tumors and up to 90% in certain types of cancer NRAS mutations in codons 12, 13, and 61 arise in 15–20 % of all melanomas. These alterations have been associated with aggressive clinical behavior and a poor prognosis. Until recently, there has been a paucity of promising genetically targeted therapy approaches for NRAS-mutant melanoma (and RAS-mutant malignancies in general). In this study, it was aimed to correct the Q61K mutation causing malignant melanoma cancer using the CRISPR / Cas9 technique, which is considered as one of the most effective techniques for genome editing. For this purpose, malignant melanoma SK-MEL-30 cell line containing the Q61K mutation was used. Once the gRNAs for the target mutation have been designed, they are transferred to plasmids and cloned. Then, plasmids and donor sequence were transferred to malignant melanoma skin cancer cells using electroporation technique. Successful transformed cells which are GFP + cells, sorted from other cells using fluorescence microscopy and flow cytometry. With the HDR-guided repair mechanism, knock-out and knock-in were targeted respectively. Real-time PCR analysis and deep-sequencing showed successful knock-out and knock-in in target cells in some cancer cells. In addition, end-point analysis supports the results of working successfully. In this project, it has been proved that even a point mutation can be corrected by the CRISPR / Cas9 technique. Using the CRISPR technique, we believe that we have given literature a new vision in terms of studying similar mutations.

Keywords: CRISPR/Cas9, Genome-editing, Malignant Melanoma, Q61K

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QTL mapping of *Ascochyta* blight resistance related DNA Markers on chickpea genome

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Abstract

Ascochyta blight is a fungal disease that causes blight lesions on chickpea plants (*Cicer arietinum* L.). It may cause severe yield losses when the environmental conditions are favorable for the fungus. In order to determine the genetic resistance in Chickpea for *Ascochyta* blight disease , Quantitative Trait Locus (QTL) analysis were performed using DNA markers. A Recombinant Inbred Lines (RIL) population consisting of 77 individuals, generated by crossing parental isolates *C. arietinum* (FLIP84-92C, resistant) x *C. reticulatum* Lad (PI599072, sensitive) was used for marker analysis and greenhouse trials. A linkage map was constructed using polymorphic RAPD markers, resulting 11 Linkage Groups (LG) with a total length of 889,1 cM. Two QTLs explaining 31% of the total phenotypic variation were revealed on LG1 and LG3 related to resistance background,. These markers may have the potential in developing *Ascochyta* blight-resistant varieties directly via marker assisted selection (MAS).

Keywords: Chickpea, *Ascochyta* Blight, linkage map, QTL Analysis

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Effect of galangin on cornea damage induced by acute radiation in rats

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Abstract

We aimed to investigate effect of galangin on cornea damage induced by acute radiation in rats. Thirty Wistar type rats were used in our study. The rats were divided into 3 groups. Groups were regulated as control, radiation and galangin+radiation groups. In the radiation group, the total cranium was exposed to 10 Gy external radiation. Galangin was received intraperitoneally 50 mg/kg dose to rats in the galangin treatment group for 2 weeks then irradiated as if used in the radiation group. In the radiotherapy group, the MDA level as a marker lipid peroxidation was significantly elevated comparing with the control group, and observed that this level was decreased in the galangin treatment group. In histological sections taken from the cornea, It was observed that the increase of endothelial cell folds reflecting vascular collapse was much more in the radiation group, whereas it was less in the galangin+radiation group. Galangin+radiation group showed that less corruption in the general morphological structure and less cellular edema, more dense cellular organs in the cytoplasm, whereas morphological structure in the radiation group deteriorated more severely. these results demonstrate that galangin has been effective in alleviating radiation-induced corneal damage.

Keywords: Radiation, cornea, galangin, rats

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Congress
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Antigenotoxic potential of *Iris taochia* Woronow ex Grossh., an endemic plant from Turkey: *In vivo* wing somatic test (SMART) on *Drosophila melanogaster*

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Abstract

The aim of this study is to determine the antigenotoxic effects of the *Iris taochia* Woronow ex Grossh, which is endemic of Turkey and harvested in Erzurum region, by using four different concentration (10, 5, 2.5 and 1.25 mg/mL) of above-ground methanol extracts of it. SMART (Somatic Mutation and Recombination Test), also known as wing spot test in *Drosophila melanogaster*, was used to achieve this aim. In this test technique, two different mutant strains carrying the recessive flare (flr3) and the multiple wing hair (mwh) determinant genes in the genome of D. melanogaster were used. The 72±4 hour-trans-heterozygous larvae which were achieved through crossing between these two mutant strains were fed with the I. taochia methanol extract and Ethyl methanesulfonate (EMS) which is well known for its genotoxic activity. As a result of study, it was observed that plant extract prepared with methanol was showed no genotoxic effect at any concentration level, whereas somatic mutation and recombination rates were observed to decrease significantly, especially at concentrations of 5 and 2.5 mg/mL when applied with EMS. Especially in this two treatment groups with the highest concentration, inhibition percentage was found to be 52.48% in normal-winged groups and up to 61.25% in serrate-winged subjects. The results obtained were proved to be statistically significant with $p<0.05$. Going forward, the early matters of plant extracts and the antigenotoxic action mechanisms of these extracts need to be uncovered with further studies.

Keywords: *I. taochia, D. melanogaster*, Antigenotoxicity, SMART

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Cloning of common bean LEA gene and functional evaluation in tobacco plant

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Abstract

Plants are frequently exposed to biotic and abiotic stresses such as drought, high salt, and low temperature which have to negative effects on growth and productivity of plants. Dehydrins (DHNs), or group 2 LEA (Late Embryogenesis Abundant) proteins, play a fundamental role in response and adaptation to abiotic stresses of plants. Therefore, it is important to screen and identify candidate genes that can confer resistance to abiotic stresses in plants. The aim of this study was clonning and functional evaluation of two novel dehydrin-like genes; LEA-dehydrin (late embryogenesis abundant-dehydrin) and OeSRC1, identified during the transriptome profiling of common bean (Phaseolus vulgaris L.) and olive (Olea europaea L.) respectively. The cloning of our gene of interest was achieved with the gateway cloning system. Tobacco leaf discs were afterwards successfully transformed with gene of interest by means of the Agrobacterium-mediated transformation method. Drought and salt stress tests were carried out on DHNs transgenic lines. It was observed that DHN expression transgenic lines displayed better growth and physiological performances compared to wild-type plants when grown under drought and salt stressed conditions.

Keywords: LEA-dehydrin, common bean, stresses, tobacco plant transformation

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Investigating microrna expression levels associated with ischemia/reperfusion in coronary artery patients before and after coronary artery bypass graft surgery

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Abstract

The objective of this study was to investigate microRNA (miRNA) expression levels associated with ischemia/reperfusion in coronary artery patients before and after coronary artery bypass graft (CABG) surgery. hsa-miR-21-5p, hsa-mi181a-5p, hsa-miR-199a-5p, hsa-miR-199b-5p and hsa-miR-320a levels were analyzed in 46 coronary artery patients pre-surgery and 60 min post- CABG and 24h post-CABG and 48 healthy controls by Quantitative real-time PCR (qRT-PCR) analysis. Pre-surgery miRNA levels were compared to controls and post-surgery (60 min and 24h) patients. Post-surgery miRNA levels were compared to time manner (60 min and 24h). Receiver Operating Characteristic (ROC) curve was used to evaluate the diagnostic value of these five serum miRNAs combination. It was found that miR-21-5p, miR-181a-5p, miR-199a-5p, miR-199b-5p and miR-320a gene expression levels were significantly lower in pre-surgery patients compared to controls. 24h post-CABG gene expression level of miR-199a was significantly lower compared to 60 min post-CABG (p=0.001). ROC analysis showed that the area under the curve (AUC) of pre-surgery was respectively 0.777 (sensitivity 65.2% and specificity 87.5%), 0.784 (sensitivity 63% and specificity 85.4%), 0.810 (sensitivity 87% and specificity 68.5%), 0.808 (sensitivity 82.6% and specificity 72.9%), and 0.784 (sensitivity 73.9% and specificity 81.2%) for miR-21, miR-181a, miR-199a, miR-199b, and miR-320a. Additionally, altered serum levels of miR-199a and miR-199b could be novel non-invasive biomarkers for coronary artery disease.

Keywords: microRNA, Coronary Artery Disease, Bypass Graft Surgery

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Congress
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Production of polylactic acid nanofiber as drug carrier

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Abstract

A polylactic acid nanofiber have been produced by electrospinning for drug delivery. A colon specific drug called oralac was used to develope a new drug delivery system in terms of loading and release studies. The active compound of the oralac is lactulose. Lactulose loading and release were studied at specific wavelength of the drug by using UV-Spectrophotometer. Absorbsiton of lactulose was detected at 277 nm. Drug loading and release studied were done at 2,2; 4; 6 and 7.6 of pH. According to the data, maximum loading of the drug (30,53 mg at 15 min) was observed at 2,2 of pH and maximum release of the drug (18,89 mg at 15 min) was observed at 7,6. Characterization was done by using FT-IR and SEM methods. As conclusion, the results indicated that polylactic acid nanofiber can be used for drug release related to the pH in basic environment which means it may be effective on human colon. It can be a candidate to use as colon targeted drug system which may reduce the side effect of the lactulose.

Keywords: Lactulose, oralac, nanofiber, polylactic acid, electrospinning, pharmaceutical

7th International
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Congress
25-27 April 2018, www.molbiotech.gen.tr

Evaluation of oxidative DNA damage on diabetic Rat by using ethanol extract of *Salvia huberi* HEDGE

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Abstract

The genus *Salvia* (Lamiaceae) comprises nearly 900 species is distributed throughout the world. The genus is represented in Turkish flora by 89 species and 94 taxa 45 of which are endemic to Turkey. In Anatolia, species of the genus known as "adaçayı" are used as diuretic, stimulant and antiseptic. In previous studies, antibacterial effect of *Salvia huberi* HEDGE which is endemic to Turkey is reported. Plant materials used in this study were collected from their natural habitat in Erzurum, Turkey. Two different concentration (0.5 % and 1 % (w/w)) of ointments were prepared from ethanol extract of aerial parts from S. huberi with glycol stearate, propylene glycol and paraffin (3:6:1) for evaluating their wound healing effect on diabetic rats and 0.5 g of the ointments were topically applied once a day for 7 and 14 days. In this study, comet assay was performed to evaluate the oxidative DNA damage in blood of all tested diabetic groups treated with *S. huberi* ointments. *Diabetes mellitus* was induced by STZ. Fito (Tripharma, Turkey) was used topically as the reference drug for positive control. In S. huberi ointments treated groups, genetic damage index (GDI) and damage cell percent (DCP) values were lower than both control and vehicle groups. Additionally, STZ increased statistically significant GDI and DCP values when compared with non-diabetic rats.($p<0.001$). The study results indicated that the ethanol extract ointments of the S. huberi reduced oxidative damage in diabetic rats as compared to the vehicle and negative and positive control groups.

ACKNOWLEDGMENTS;This study was supported from Adiyaman University Scientific Research Center TIPFBAP/2015-0004.

Keywords: *Salvia huberi*, DNA damage, comet assay, diabetes, streptozotocin

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Investigation healing effects of isgın and dandelion against of doxorubicin induced toxicity in *D. melanogaster*

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Abstract

Chemotherapy is one of the effective methods used in the treatment of cancer. The greatest concern during the use of chemotherapeutic drugs is side effects. Increasing free radical formation in cells and tissues and thus oxidative stress is known as one of the most damaging systems. Antioxidants used during chemotherapy may increase the efficiency of treatment by decreasing the formation of radicals due to oxidative stress and prevent healthy cells from being damaged. From this point of view, the therapeutic and protective properties of Isgın (*Rheum ribes*) and Dandelion (*Taraxacum officinale*) giant toxic effects of Doxorubicin, one of the drugs used in chemotherapy, were investigated by conducting survival rate experiments on *Drosophila melanogaster*. For this purpose, in each experimental set, fly larvae (72±4 hours) were placed in the media and individuals who developed from larvae were recorded. As a result of the study, it was determined that the percentage of survival rate decreased in Doxorubicin treated group compared to the control. In addition, while the percentage of survival rates belonging to Isgın and Dandelion treated groups were higher than the control, the values in plant extracts plus Doxorubicin treated groups were close to the control. These differences in survival percentage were statistically significant ($p < 0.05$). This protective effect can be explained by the inhibition of the formation of free oxygen radicals by the antioxidant properties of plants and the removal of them from the biological system.

Keywords: Doxorubicin, *Drosophila melanogaster*, *Rheum ribes*, *Taraxacum officinale*, The survival rate

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The effect of 1800MHz cell phone radiation on COMT, MAO-A, Crybb1 genes expression levels in rat cardiac tissue

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Abstract

Most of the research studies mainly focused on the radiofrequency electromagnetic fields (RF-EMF) exposure and it's health effect. Catechol-O-methyltransferase (COMT), Monoamine oxidase A (MAO-A) and Crystallin, beta B1 (Crybb1) genes could play an important role in congestive heart failure. This study examines the possible effect of 1800Mhz RF-EMF on these gene expression levels in cardiac tissue to eleminate that cell phone exposure may be effect cardiac problems. Twenty-two female wistar albino rats were divided into three groups. Experiment group was exposed 1800Mhz RF-EMF 2h/day along 8 weeks. Control group was kept in their own conditions. Sham group was kept in experiment conditions without RF-EMF exposure. Immediately end of the 8 weeks the rats were sacrified and removed their heart. Stored at -80oC until RNA isolation. RNA isolation was performed from tissue homogenate. COMT, MAO-A, Crybb1 genes expression levels was determined with TaqMan assays. Findings showed that Crybb1 gene (p= 0,015) and COMT gene (p=0,004) expression levels was significantly different between the groups. Further studies should be performed.

Keywords: Cell phone radiation, Cardiac tissue, Gene expression, COMT gene, MAO-A gene, Crybb1 gene

7th International
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Characterization of meniscus scaffolds containing PHBV nanofibers and loofah embedded in chitosan

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Abstract

Meniscus injuries are very important in orthopedic surgery. Meniscus stapling and stitching as well as meniscus prostheses are available for total meniscus injury. However, they do not meet all of the expectations. Tissue engineering techniques have been developed to provide alternative strategies for the repairment of damaged meniscus tissues. In this study, we aimed to develop hydrogel scaffolds that have similar characteristics with the meniscus and show good biocompatibility. For this purpose, three different types of scaffolds were prepared. The first scaffold was made of chitosan hydrogel only. The second scaffold was composed of natural cellulose based Loofah integrated within chitosan matrix. Poly(3-hydroxybutyric acid-co-3-hydroxyvaleric acid) (PHBV) nanofibers and Loofah embedded in chitosan was used as the third scaffold. PHBV nanofibers were prepared by wet-electrospinning method and then mixed with Loofah. This three-dimensional fibrous structure obtained after freeze-drying was immersed into chitosan solution. Second freeze-drying step was applied to obtain composite sponges. Chitosan based hydrogel scaffolds were maintained by using different concentrations of genipin as the cross-linking agent. The morphologies and chemical structures of the scaffolds were characterized by scanning electron microscope (SEM) and Fourier-transform infrared spectroscopy (FTIR), respectively. The swelling ratio test of the scaffolds was carried out in phosphate buffered saline (PBS) solution. Also, the mechanical properties of the scaffolds were assessed under compression loading. Hereby, the new meniscus scaffolds designed could have the potential of healing meniscal tears.

Keywords: Scaffold, Tissue Engineering, PHBV, Loofah, Chitosan

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Niche modelling study on some *Ampedus* species (Coleoptera: Elateridae) of Turkey

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Abstract

The genus *Ampedus* is one of the rich genera of Turkish Elateridae, has 44 species in Turkey. A considerable number of species are morphologically quite similar to each other and there are concerns about making species diagnostics using diagnostic keys. In order to understand phylogenetical relationships of *Ampedus* (*Elateridae*) species in Anatolia, Sequences of CO1 gene region of some *Ampedus* species were compared with CO1 sequence of five other species belonging to the same genus and outgroup species which are three species from the same subfamily, two species from different subfamilies and one species from the nearby family Buprestidae from NCBI database. Three Ampedus species selected for (*Ampedus platiai, Ampedus sanguinolentus, Ampedus samedovi*) for Ecological Niche Modelling (ENM) from the result of phylogenetic analysis. ENM results of current time for the species is consistent with the known distributions of the species. Using IPCC 5 climate scenarios, possible future distributions of species for 2050 and 2070 are estimated using ENM. The results obtained are important in understanding how species in Anatolia will react to climate change and planning conservation strategies. The fact that the *Ampedus platiai* is an endemic species, it increases the significance of results in terms of conservation biology.

Keywords: *Ampedus*, Coleoptera, Elateridae, Phylogeny, Ecological niche modelling, Climate change

7th International
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Molecular phylogeny of the *Thlaspiceras* sensu Meyer species complex of the genus *Noccaea* Moench (Brassicaceae)

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Abstract

Noccaea Moench is taxonomically one of the most problematic genera of the cabbage family (Brassicaceae) and discussions about its generic circumscription are still ongoing. Thlaspiceras species complex which previously considered as a different genus, consist of 11 species (9 of which are endemic to Turkey) mainly distributed Mediterranean part of Turkey. In this study, phylogenetic relationships of the members of this complex are investigated using of Internal transcribed spacer regions (including ITS1 and ITS2) and the 5.8S gene of nuclear ribosomal DNA and encompassing the largest sampled data set (44 populations of 8 species) used so far. This data set were investigated first time based on Bayesian phylogenetical analysis and contrary to previous cpDNA results, the members of the Thlaspiceras complex are monophyletic (posterior probability= 1.00), although their phylogenetic relationships are not concordant with the classical delimitations of the species. 9 haplotypes were detected in the data set, some of which are shared among species. Additionally all species examined were subjected first time a molecular biogeographical analyses. Ancestral area reconstruction analyses based on Bayesian Binary Marcov Chain Monte Carlo (BBM) simulations with 100 % statistical support revealed members of the Thlaspiceras species complex arose in the Amanos mountains. Finally the most important taxonomical character (fruit horn) was assessed by BBM algorithm and results clearly showed that hornless fruit (with % 97 statistical support) is ancestral for this complex.

Keywords: Brassicaceae, ITS, *Noccaea*, Molecular phylogeny, *Thalspiceras*

7th International
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Revealing the potential of plant- and fungus derived chitinase for enhanced fungal resistance in Transgenic Potato lines

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Abstract

Chitinases are strong antifungal enzymes, and their genes can be utilized to engineer crop plants, including potatoes, with tolerance against fungal pathogens. We have evaluated the fungal inhibition activity of Barley derived chitinase gene and chitinase from Trichoderma fungus. The recombinant chitinase proteins were initially expressed in prokaryotic host and subsequently the purified recombinant protein fraction was subjected to in-vitro fungal inhibition assay. The barley derived chitinase inhibited the growth of Alternaria solani from 39.5 to 60.5%; of Colletotrichum falcatum by 52-56%, in a quantitative in vitro assay. While the recombinant chitinase of Trichoderma inhibited the growth of Fusarium oxysporum to 78% in a quantitative in vitro assay. Further, transgenic potato lines were generated expressing chitinase as anti-fungal genes. The transgenes were driven by strong promoter to achieve enhanced transcription and translation. The bioassay of transgenic potato lines revealed varying degree of resistance against inoculated fungi. The transgenic plants remained healthy and green in comparison with the control plants, which turned yellow and eventually died 3 weeks after infection. mRNA expression of the transgene was revealed to be up to 7 folds high in fungus infected transgenic potato lines.

7th International
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Identification of potential transcripts in *Hibiscus sabdariffa* l. expressing under drought and salt stress

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Abstract

A better understanding of genetic and environmental factors affecting plant response to salt and drought stress is necessary to produce economic yield. The current study was performed to assess the effects of NaCl on Roselle (*Hibiscus sabdariffa* L.) by Differential display (DDPCR). By screening of 99 sets of primer combinations, an up-regulation of 34 cDNA transcripts were identified and 24 were gel purified, reamplified, cloned and sequenced. The BLASTX revealed 3 transcripts showed significant homology with known genes, while 6 transcripts for drought stress also showed overexpression. Real-time RT-PCR expression studies revealed significant over expression of transcripts in roots under salt and drought stress respectively. Full length sequence of RMYB gene revealed that it belongs to MYB protein comprises of a single Open Reading Frame (ORF) of 229 amino acids with no intron region. The high level of constitutive expression in stem, leaves and roots was observed. Expression analysis of RMYB gene demonstrated higher level in drought and salt stress followed by cold stress. 3D image of RMYB was predicted by *in silico* 3D homology modeling studies generated by using the *I-TASSER server* based on fold recognition method. Validation of 3D structure was done by Ramachandran plot and calculation was assessed by PROCHECK analysis for reliability. Gene was cloned in plant expression vector under CaMV 35S promoter, with GUS reporter gene and genetic transformation in local cotton variety was done and transgenic plants were confirmed through PCR by using gene specific primers. Identification of the potential and novel transcripts will contribute to understand the molecular mechanism of salt and drought stress.

Keywords: Differential Display, Drought stress, Gene expression, Salt stress

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Rational design of a novel biocatalyst using a gas sensor hemoprotein

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Abstract

Recent advances in molecular biology and protein design have led to the application of biocatalysis as an alternative to chemical catalysis in the synthesis of therapeutics with regioselectivity and enantioselectivity. Hemoproteins contain the heme prosthetic group. Hemoproteins play a large variety of roles in biological systems, making them good candidates for biocatalysis. The Heme-nitric oxide/oxygen binding (H-NOX) protein was identified by homology to the nitric oxide signaling protein. soluble guanylate cyclase. In this research, the H-NOX domain from the methyl-accepting chemotaxis protein, *Caldanaerobacter subterraneus* subsp. *tengcongensis* (TtH-NOX), was tuned into a biocatalyst using rational design. Four mutants of TtH-NOX were characterized. Each mutant was tested for catalase and peroxidase activities. The wild type TtH-NOX catalyzed hydrogen peroxide decomposition and 2,2'-azino-bis(3-ethylbenzthiazoline-6-sulfonic acid (ABTS) oxidation inefficiently. However, the mutation of a distal tyrosine to a histidine resulted in an increase in the oxidative activities as compared to the wild type. On the other hand, the mutations in the proximal pocket of TtH-NOX decreased catalytic activity for these reactions. Taken together, the mutations in the distal pocket and proximal pocket resulted in changes in reaction rates and electronic properties of the heme group. The mutations changed the molecular mechanism of the hemoprotein, showing that both the proximal and distal pocket residues are vital for catalysis. These observations contribute to the understanding of the physiological roles of hemoproteins. This project will pave the way for discovery of novel biocatalysts using H-NOX proteins as scaffold and aid in the design of future biocatalysts.

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Antioxidant properties of pitaya seeds and oxidative stability of seed oils

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Abstract

Two types of pitaya seeds were evaluated on antioxidant activity and stability of seed oils. Each of them have red outer shell but *Hylocereus undatus* have white pulp color while *Hylocereus polyrhizus* have red pulp color. Fruit samples were obtained from Gazipaşa/Antalya in September 2016. The fat content of *H. polyrhizus* and *H. undatus* pitaya seeds were 22.78-23.97 %, respectively. The total phenolic content, α-tocopherol content, γ-tocopherol content, free radical scavenging activity and induction time of H. polyrhizus and H. undatus pitaya seed oils were determined as 12.81-11.90 mg GAE/g dry sample, 3.67-2.75 g/kg oil, 1.29, 1.64 g/kg oil, 46.90 to 51.47% inhibition and 5.37 to 5.07 hours, respectively. Seeds contain significant quantities of phenolic compounds and tocopherols. Percent of unsaturated fatty acids were found to be high in seed oils of both pitaya species. Unsaturated fatty acids detected in H. polyrhizus seed oil was 77.79 % and was 80.67 % in H. undatus seed oils. In both pitaya species, linoleic acid, a polyunsaturated fatty acid, was the dominant fatty acid. Antioxidant activity of seed extracts were found to be at medium level. Resistant of seed oils to oxidation were similar to resistant of sunflower oil. It may be suggested that furthure studies on betalains of pitaya may produce more detailed activity data.

Keywords: *Hylocereus polyrhizus, Hylocereus undatus*, Antioxidant activity, Fatty acids, Oxidative stability

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Seven fungal strains with a potential for the bioethanol production from lignocellulosic materials

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Abstract

Bioethanol has been considered as one of the best candidates for the replacement of the fossil fuels. However, the use of food sources as raw materials in traditional production methods increases social and scientific concerns. This situation has accelerated the search for alternative raw materials, and consequently, lignocellulosic biomass has been approved as a promising non-food source for next-generation biofuel production. Thus, recent research efforts have focused on the microorganisms having lignocellulolytic and bioethanol producing activities. In this context, our present study was conducted to identify seven ethanol producing fungal strains that were previously isolated from decaying woody materials. Ethanol yield rates of the strains grown in the modified BMC medium were 1.72 g/L for MG3, 6.61 g/L for MG8, 26.68 g/L for MG9, 9.93 g/L for MG11, 6.12 g/L for MG16, 4.37 g/L for MG47 and 3.91 g/L for MG50. Conventional microscopic examinations and molecular techniques including ITS-PCR, sequencing of amplicons and their BLAST analysis on the NCBI database were used for identification of the fungal strains. According to the results, the strains were identified as Penicillium brevicompactum (MG3), Trichoderma harzianum (MG8 and MG50), Mucor plumbeus (MG9), Fusarium solani (MG11 and MG47) and Fusarium candidum (MG16). In conclusion, seven fungal strains with a potential for the bioethanol production from lignocellulosic sources were cultured and identified. This data is valuable for the development of next-generation biofuel production technologies using the non-food based raw materials.

This study was supported by Republic of Turkey – Ministry of Food, Agriculture and Livestock: TAGEM-13/ARGE/17.

Keywords: Bioethanol, Fungi, Internal transcribed spacer (ITS), Lignocellulose

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Removal of Cu (II), Co (II) and Ni (II) Ions from Aqueous solutions using modified sporopollenin

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Abstract

Heavy metal pollution is one of the most important environmental problem. Heavy metal accumulation in soil and water has an important influence not only on functions of ecosystem but also on the health of animals and human beings via food chains. The most important source of heavy metal pollution is the industries. Heavy metals in wastewater can be removed by different methods. Particularly the issue of removing heavy metals with solid support sporopollenin is of great interest. The rapid growth of the industry and technology in our age increases the environmental pollution. Sporopollenin used in this study is naturally found on plant walls. Because sporopollenin is a natural substance, it has great resistance to external influences. In this study, (E)-2-((2-hydroxynaphthalen-1-yl) methylene)amino)pyridin-3-ol was covalently bound on the surface of sporopollenin via chemical reaction. Newly synthesized substance was characterized with infrared spectroscopy method. The sorption capacity of such a matrix for Cu(II), Co(II) and Ni(II) in aqueous solutions was studied. Langmuir, Freundlich and Dubinin-Radushkevich adsorption isotherms were calculated. For adsorbent, thermodynamic parameters were calculated and ΔH0, ΔS0 and ΔG0 values were estimated. This investigation reveals a new, simple, environmentally friendly and cost-effective method for removal of metal ions from aqueous solutions.

Keywords: Sporopollenin, Self-Assembled Monolayers, Immobilization, Adsorption, Adsorption Isotherm, Termodynamic

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Molecular binding profile of protoberberine alkaloids on glycogen synthase kinase 3β as a drug candidate for Alzheimer's diseases

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Abstract

Glycogen Synthase Kinase 3β (GSK-3β) is a serine/threonine kinase which has essential roles in Alzheimer's Diseases (AD) processes. AD shows neuropathological markers as tau hyperphosphorylation and accumulation of amyloid β (Aβ) proteins. Aβ proteins are generated from sequential cleavages of amyloid precursor protein (APP). Recent studies show that inhibition of GSK-3β causes to decrease in the cleavage of APP. Thus the accumulation of Aβ was prevented by this process. Due to the therapeutic benefit of the inhibition of GSK-3β it has been a favoured target for scientists. Alkaloids are secondary metabolites which are produced by a large variety of organisms as plants with diverse structures. Protoberberine alkaloids such as berberine, palmatine, jatrorrhizine, columbamine, magnoflorine were found to prevent a progressive neurodegenerative disorder as experimentally, the mechanisms of them are not absolutely clear. In this study, we have aimed to elucidate the binding and affect mechanism of these alkaloids on the GSK-3β. For this purpose, molecular docking studies were applied for these natural products by using CDOCKER module of Discovery Studio 3.5 Client. Binding mechanism was identified by Hydrogen, π bindings' between ligands and GSK-3β.

Keywords: Alzheimer's Diseases, Protoberberine alkaloid, Molecular docking

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Structural and spectroscopıc analysıs of ε-caprolactam molecule and dockıng studıes on NDM-1

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Abstract

Caprolactam, which is soluble in water, some hydrocarbons and many solvents containing oxygen and chlorine, is a colorless crystalline cylic amide with a melting point of 70 °C. Caprolactam as monomers for nylon-6 and engineering plastics is an important basic organic chemical widely used in textiles, automobiles, electronics and other industries. Some lactam derivatives have antibacterial, anticancer and antifungal properties. Caprolactam is readily biodegradable and classified as non-toxic to the environment or aquatic life. In addition to these, it is not classified as carcinogenic. In this study, the optimized molecular structure of caprolactam was determined by density functional theory and the molecular structure has been revealed by comparing the experimental 1H-NMR, 13C-NMR and IR spectra with the theoretical 1H-NMR, 13C-NMR ve IR spectra. Also, the New Delhi metallo-β-lactamase 1 (NDM-1) enzyme is a key enzyme that pathogenic Klebsiella pneumonia uses to hydrolyze nearly all lactam antibiotics. The hydrolysis is the most common cause of resistance among clinically important Gram-negative bacteria. Because of this property, the interaction of ε-caprolactam molecule with NDM-1 enzyme has been investigated theoretically and the position and the orientation of the caprolactam molecule in this enzyme were determined.

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Preparation of gelatin-based electro-conductive hydrogel for biomedical applications

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Abstract

Herein, an electro-conductive hydrogel (ECH) was synthesized by encapsulation of poly(3,4-ethylenedioxythiophene):poly(styrenesulfonate) (PEDOT:PSS) into the gelatin methacrylate (GelMA) hydrogel. Three different amounts of PEDOT:PSS (0%, 1%, 1.5%, 2.5% w/w) were used to invesitgate the effect of amount of PEDOT:PSS on the conductive properties of hydrojel. According to conductivity measurements which were performed by using 4-probe methods, the highest conductivity (1×10^{-2} S/cm) was obtained by encapsulating 1.5 wt.% PEDOT:PSS into the GelMA polymeric network. The presence of PEDOT:PSS in GelMA network was confirmed by FT-IR and SEM analysis. Cytotoxicity test was carried out by using WST-1 assay and L929 cell lines. It was seen that GelMA-PEDOT:PSS hydrogels have no any toxic effect on the viability of L929 cell lines. All results showed that the obtained ECH could be a promising biomaterial for biomedical applications in the future works.

Keywords: Electro-conductive hydrogels, GelMA, PEDOT:PSS.

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Cytotoxicity and cellular uptake of re-labeled magnetic protein cages

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Abstract

Protein cage architectures designed for medical imaging and therapy, provide enhanced stability and biocompatibility under physiological conditions as well as providing functionalization and targeting. In the study, empty ferritin protein was used as a cage architecture and magnetic iron oxide nanoparticles were synthesized within the interior cavity of the protein (magnetoferritin). Non-radioactive Re was incorporated on the protein cage exterior, so that a multifunctional nanoparticle (Re-magnetoferritin) was prepared for localized radiation therapy due to its' magnetic targeting capability while enhancing contrast in MRI signals. In order to evaluate the potential use of these nanoparticles in cancer therapy; cellular uptake, in vitro cytotoxicity, apoptotic potential of nanoparticles were evaluated in both human normal mammary epithelial and breast metastatic adenocarcinoma cell lines. The results showed that the internalization of nanoparticles into the cells is through receptor mediated endocytosis and increased in 4 hours. Cancerous cells exhibited significantly highest uptake as well as highest cytotoxicity compared to normal cells. The mineralization and surface modification of ferritin did not alter the cell viability when compared of the results of the proteins without modifications to a large extent. IC50 values of nanoparticles were calculated as 0.96 mg/mL for cancerous and 1.73 mg/mL for normal cells. The main mechanism of cell damage in both cell types is found to be apoptosis and nanoparticles induced higher apoptotic rates in cancer cells compared to normal cells. At concentrations above 1 mg/mL, NPs induce apoptosis which can also be used for cancer treatments.

Keywords: Magnetoferritin, Cytotoxicity, Apoptosis, Breast cancer cell lines

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Investigation of pre-miR-34a rs35301225 and pri-miR-34b/c rs4938723 polymorphisms in lung cancer

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Abstract

Lung cancer is the most common cancer in worldwide according to GLOBOCAN 2012. A number of factors play a role in lung cancer etiology. Expression level of some miRNAs in lung cancer has been associated with increased risk of cancer. MicroRNAs (miRNAs) are small (19-25 nucleotides), noncoding RNAs and negative gene regulators. miRNAs play a substantial role in the pathogenesis of human cancers. Because of that, miRNA polymorphisms can be important for carcinogenesis. MiR-34 is a family of miRNAs known to have reduced levels of expression in lung cancer and other human cancers (pancreas, colon). It is function like tumor supressors and targeting oncogenes like MET, RET and RAB43. In this study we investigated two polymorphisms (rs35301225 C/A,T and rs4938723 T/C) in miR34a and miR-34b/c from miR-34 family. The study population composed of 100 patient with lung cancer and 100 healty controls. Blood was collected into EDTA-containing tubes and genomic DNA was extracted. Genetic polymorphisms of miR-34a rs35301225 and miR-34b/c rs4938723 were detected by using PCR-based restriction fragment length polymorphism (RFLP). We found that miR-34b/c rs4938723 variant heterozygote CT was associated significantly increased risk of lung cancer compared with their wild-type homozygote TT ($p<0.01$). There was no polymorphism in 100 controls but we found heterozygous polymorhism in 42 patients from 100. However, no significant effects were observed on association between miR-34a polymorphism rs35301225 and lung cancer. In conclusion, rs4938723 polymorphism of miR-34b/c is thought to play an important role in the pathogenesis of lung cancer.

Keywords: Lung cancer, miRNA, miR-34a, miR-34b/c, polymorphism

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Investigation of gene polymorphisms associated with vasodilator effect of leptin in in-vitro pre-eclampsia model

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Abstract

The aim of this study was to investigate whether leptin has both molecular and pharmacological effects on serotonin-induced contraction responses in normal and pre-eclamptic human umbilical artery. Umbilical cord from both normal pregnant women and pregnant women with pre-eclampsia were used which were obtained from Obstetrics and Gynecology Clinic. These tissues were performed as in-vitro experimental model. The in-vitro isolated organ bath was used for evaluation of pharmacological agent effectiveness. Serotonin which is a vasoconstrictor agent available in human plasma, was applied to umbilical artery as 10-9-10-7M concentrations cumulatively. Vasoconstrictor effects of serotonin were examined in the presence or absence of leptin. Leptin, a vasodilator agent, was applied at 10-7M concentration in order to distinguish the responses of vasodilation and vasoconstriction on normal and pre-eclamptic cords by MP36 software. For molecular-based experiments, a modified DNA isolation protocol was created. eNOS and leptin receptor genes polymorphisms were determined by HRM analysis and validated by PCR-RFLP analysis. In conclusion, there is no significant vasodilatation rate of leptin at serotonin concentration of 10-7M ($p>0.05$). However, the difference was significant on vasodilatation rates in normal and pre-eclampsia groups compared to controls at 10-9-10-8M concentrations ($p<0.05$). As for molecular analysis, rs1137100, rs1137101 and rs2070744 single nucleotide polymorphisms (SNP) were determined ($p>0.05$). Student t-test was used as statistical analysis. The results suggest that SNPs in the eNOS and leptin receptor genes may be related to pre-eclampsia. Therefore, future studies are required to reveal the role of eNOS and leptin receptor genes in the pathogenesis of pre-eclampsia.

Keywords: Gene polymorphism, Leptin, Pre-eclampsia, Serotonin, Vasodilatation

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Investigation of gene expression in sciatic nerve ınjury using lithium loaded hyaluronic acid microgel

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Abstract

Recently, studies about peripheral nerve injury treatment have focused on use of various conduits obtained from some biomaterials. These biomaterials may be natural or biodegradable synthetic materials. Especially hyaluronic acid is one of the best-known natural biomaterials and it is known to reduce scar formation at injury site. Therefore, the significance of cellular survival pathways in the effective treatment of nerve injury is great. Ionic agents such as lithium have important roles in these pathways. Lithium is an enzymatic inhibitor of Glycogen synthase kinase 3 beta and it activates the Wnt/ β-catenin signaling pathway. So, it plays an important role in the survival of the cells in nerve injury. It also has great potential therapeutic benefit for some neurodegenerative diseases because of its protective effects. In this study, hyaluronic acid microgel was synthesized and then loaded with lithium. This microgel was used as filling material. The results were compared to the solely lithium treated case. Our study revealed that the treatment of rat with lithium solution or lithium loaded hydrogels after peripheral nerve injury stimulated the expression of vascular endothelial growth factor A, brain derived neurotrophic factor, glial derived neurotrophic factor, nerve growth factor, restored nerve structure and accelerated the recovery. According to our data, this system has proliferative effects of lithium on both axon and Schwann cells and so it may be used as a potential nerve guidance filling material.
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Keywords: Microgel, Nerve injury, Lithium, Gene expression

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Protective effect of hyperbaric oxygen therapy on gentamicin-induced nephrotoxicity

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Abstract

Hyperbaric O2 is a method of using pure O2 at a higher atmospheric pressure in order to treat a medical condition. Hyperbaric oxygen therapy (HBO) for gentamicin-induced nephrotoxicity is thought to be effective in the treatment of renal toxicity in animal models. The purpose of this study is to investigate the effect of HBO therapy on gentamicin-induced nephrotoxicity in rats. Rats were randomly assigned to four different groups of seven rats in each group. The study consists totally 28 male wistar albino rats. Fourteen of the rats were injected with 100 mg/kg intraperitoneal gentamicin once daily for 7 days. The other half of the rats were exposed to HPO 90 min daily for 7 days under 2.5 atm. On the day of eight, the laboratory results were obtained from serum. Moreover, we also performed malondialdehyde, Superoxide dismutase and α–Glutathione-S-Transferases levels of the kidney in all groups. When the gene expression of cytokines in kidney tissue was examined, TNF-α, IL-1β and Kim-1 levels in the gentamicin treatment group were statistically increased compared to the HBO+Gentamicin group (p=0.015, p=0.024, p=0.004). Serum Urea, albumin and LDH levels were found to be increased (p = 0.006, p = 0.224 and p = 0.180 respectively) in gentamicin group compared to HPO + gentamicin group. The HPO + Gentamicin therapy group was not statistically different from the control groups but significantly different from the Gentamicin group for antioxidant parameters. Histopathologic studies have been performed in which hyperbaric oxygen administration significantly reduced the renal damage. Gentamicin administration caused tubular necrosis in kidney. HBO administration may be recommended for treatment of nephrotoxicity originating from gentamicin. Therefore, in this study, we investigated the effects of HPO to discuss the potential role of HPO in nephrotoxicity.

Keywords: Gentamicin, HPO treatment, nephrotoxicity

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Investigation of osteogenic associated genes in a new generation bone substitute: Combination of mesenchymal stem cells and fibrin glue coated ceraform

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Abstract

There are three fundamental elements for bone tissue regeneration: osteogenic progenitor cells, osteoinductive growth factors and osteoconductive scaffolds. Ceraform®, is a synthetic calcium phosphate ceramic and it is biocompatible bone defect filling material. In this study adipose tissue derived mesenchymal stem cells were differentiated into osteoblast cells and loaded on Ceraform®. In order to improve cell adherence, Ceraform® was covered with fibrin glue (FG). The cells were cultivated for a 28-day period by osteogenic induction medium. Days 1, 7, 14, 21 and 28 were selected as specific intervals for incubations. Total RNA was isolated and cDNA was synthesized. Differences in the expression of runt-related transcription factor 2 (Runx2), bone morphogenetic protein-2 (BMP-2), and osteocalcin (OCN), collagen (COL-1) and osteopontin (OPN) were determined by qPCR. The peptidylprolyl isomerase A (PPIA) gene was used as an internal control. According to the qPCR results Runx2, COL-1 and OCN gene expressions were highest on the day 14th and then start to decrease. BMP-2 gene expression was increased on day 14 and 21 then maximum on day 28. On the other hand, OPN gene expression was decreased on days 7 and 14. These findings pointed out that the osteogenic induction was successfully activated on FG coated bone material. Therefore, this new bone substitute is promising in clinical applications.

Keywords: Adipose- derived mesenchymal stem cell, ceraform, fibrin glue, osteoinduction

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CoQ0 (Coenzyme Q0) decreases nitrite levels in IFN-γ activated RAW 264.7 macrophages

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Abstract

CoQ0 is a redox-active ubiquinone compound that accumulates mostly in the mitochondria of cells. Anti-angiogenic, anti-inflammatory and cancer modulatory effects have been reported for CoQ0 in vitro or in vivo. However, the effects of CoQ0 on IFN-γ mediated inflammatory signaling is largely unkown. In this study, we tested CoQ0 for its modulatory effects on IFN-γ activated RAW 264.7 macrophages which are widely accepted models of inflammatory signaling. We used spectrophotometric MTT assay,Griess assay and Western blot for the determination of cell viability,nitrite levels and STAT1 protein levels respectively.Our results showed that, when applied solely,CoQ0 did not significantly effect cell viability at 0.5-10 μM concentration range whereas diminished cell viability at 25 μM and higher concentrations ($p<0.001$). According to these results,non-cytotoxic concentrations of CoQ0 (0.5 μM-10 μM) were applied to IFN-γ stimulated RAW 264.7 cells. To examine the probable anti-inflammatory activity of CoQ0 on macrophages,nitrite levels which are the end product nitric oxide were measured in the medium.For this purpose, RAW 264.7 macrophage cells were treated with CoQ0 for 1 hour before IFN-γ (20 ng/ml) treatment for 20 hours.According to the results, IFN-γ treatment caused a significant increase in nitrite levels compared to cells without IFN-γ stimulation and CoQ0 treatment (1-5 μM) decreased nitrite levels ($p<0.001$) compared to cells treated with IFN-γ only. Cell viability significantly decreased with 10 μM CoQ0/IFN-γ treatment and it is concluded that the inhibitory effect of CoQ0 on nitrite levels is between 1-5 uM concentration range.CoQ0 also decreased p-STAT1 protein levels in IFN-γ stimulated macrophages at its nitrite inhibitory concentrations.These results collectively show that CoQ0 has inhibitory activity in IFN-γ treated macrophages and further studies may contribute to reveal its effects on IFN-γ related signaling to use this compound for IFN-γ associated pathologies.

Keywords: CoQ0, RAW 264.7, macrophage, nitrite

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A new approach to the meniscus damage treatment: Synovium-derived exosomes

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Abstract

In surgical meniscus tear treatment rehabilitation and patient satisfactory rate is low and patients face a strong risk factor of osteoarthritis therefore development of a non-invasive and more efficient treatment is crucial. Exosomes are emerging as a popular cell free candidate with many advantages over cell therapy. In this study we hypothesized that the efficiency of meniscus tear treatment with exosomes enhanced with stem cells derived from join itself can be rather high and the treatment could be forefront and better alternative than cell therapy. Therefore exosomes derived from mesenchymal stem cell isolated from rat synovium tissue (rS-MSC) were isolated, characterized and analysed and its regenerative abilities were analysed and compared with parents. Initially rS-MSCs were enzymatically isolated from synovium tissue and characterized by immunophenotypic and differentiation assays. Afterwards exosomes that isolated from the characterized rS-MSCs were characterized and analysed in terms of cell to cell transplantation and regenerative ability. As a result, MSCs isolated from synovium tissue were positive for CD90, CD54, CD29, MHC Class I and negative for MHC Class II. The cells were also differentiated to adipo-, osteo- and chondrogenically to characterize as MSC. Exosomes were also positive for CD63 and CD81 markers. It was observed that they could be uptaken by target cells in transplantation assays. Wound healing assay proved that regenerative abilities were better than parents. Our study suggests exosomal transplantation; a bioactive cell-free therapy could be a better therapy alternative for meniscus injury.

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Keywords: Rat, synovium, mesenchymal stem cell, exosome

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In vitro shoot regeneration in olive (*Olea europaea* L.) cv. Gemlik

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Abstract

The olive (*Olea europaea* L.) is one of the oldest and most important crops in terms of human consumption, and has economic value due to its nutritive and therapeutic values. In the present study, the effect of plant growth regulators (PGRs) on in vitro shoot regeneration of *O. europaea* was studied. For this purpose, nodal explants were cultured on woody plant medium (WPM) supplemented with cytokinins [6-benzyladenine (BA), kinetin (Kn), or gibberellic acid (GA_3)] at the concentrations of 0.5–4.0 mg/L. The highest shoot regeneration rate (93.33%) and shoot number (1.87 shoots per explant) were observed on WPM containing 4.0 mg/L BA followed by 2.0 mg/L BA and 4.0 mg/L Kin which were statistically placed in the same group. The longest shoot (3.0 mm) was obtained with 1.0 mg/L Kn. WPM supplemented with 2.0 mg/L GA_3 gave the best response regarding the leaf number (2.67 leaves per explant). The highest leaf length (5.4 mm) was recorded on WPM containing 0.5 mg/L BA and 4.0 mg/L GA_3.

Keywords: *Olea europaea*, Shoot regeneration, Cytokinin, *In vitro*

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Application of plant cell and tissue cultures in environmental genetics

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Abstract

The plant cell and tissue cultures were used for investigation of influence of environmental factors including urban pollutions like UV irradiation and low frequency (50 Hz) electromagnetic field (LF EMF) of different density in presence of silica nanoparticles. The changes of cell relative fluorescence in tested cultures were detected with use of BD FACSJazz® cell sorter. Influence of different density LF EMF on cells of several plant species (*Cyclamen persicum, Tilia cordata, Hordeum vulgare* and *Triticum aestivum*) was investigated. In order to use test organisms using most suitable preparations somatic cell culture from callus culture initiated from leaves of flax (*Linum usitatissimum*) was used first of all. Somatic and gametic cell cultures of other species were also established. The relative fluorescence of the somatic cells had large distribution, since the cells differed by many parameters (size, shape, metabolism etc.). Immature pollen cells (one-nucleus stage) as most appropriated for investigation of influence of environmental factors were found. A significant increase in relative cell fluorescence was observed for all mentioned plant species after treatment by UV irradiation and LF EMF with density 400μT. It was found that cell relative fluorescence was dependent on duration of cultivation in SiO2 nanoparticles suspension. The genetically different clone cultures of freshwater macrophyte duckweeds (*Lemna minor*) were established and used as excellent model system for investigation of environmental factor influence on whole organisms.

Keywords: Cell culture, UV irradiation, LF EMF, silica nanoparticles, *Lemna minor*

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Effects of EGFR 19. exon 747-750 deletion on capture of non-small cell lung cancer

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Abstract

The most important factor in the etiology of lung cancer is smoking, which is important in other environmental pollutants and genetic susceptibility. The course of NSCLC is very aggressive, has a high mortality, and constitutes a large proportion of lung cancers, about 80%. Among the gene mutations that are prognostic value in NSCLC, EGFR accounts for the highest rate with 50-80%. EGFR is a transmembrane glycoprotein that exhibits both tyrosine kinase activity associated with normal cell growth and conversion to malignant transformation. In our study; The relationship between EGFR gene 19th exon 747-750 deletion in NSCLC has been examined. A sample of 180 patients who were diagnosed as NSCLC in Mersin University Medical Faculty Oncology Clinic and healthy 192-person control group that was created by considering the same age and gender characteristics. The DNAs were obtained according to the standard salt precipitation method. Mutation detection and genotyping analyzes were determined by polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) analyzes. The mutant genotype ratio of EGFR exon-19 deletion was 15.1% in the control group and 29.5% in the NSCLC group, increasing the risk of NSCLC by 2.68 fold($p<0.001$). Distribution of NSCLC according to major histological types of tissue; adenocarcinoma. 61.8% in squamous cell carcinoma, 28.9% in squamous cell carcinoma and 9.2% in squamous cell carcinoma($p<0.001$). Male gender, smoking, and older age were shown to be important risk factors for NSCLC. ($p<0.001$).

Keywords: KHDAK, EGFR gene, ekzon-19, 747-750 deletion, older age

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Pyrosequencing of KRAS, NRAS and BRAF mutations using in metastatic colorectal cancer cases: The need to establish a NGS based specific cancer panel

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Abstract

Approximately 50–60% of patients diagnosed with colorectal cancer have metastases. The relative 5-year survival rate in metastatic colorectal cancers is 10%.The receptor for EGFR has been reported to be overexpressed in 49–82% of colorectal tumours. In recent years, anti-EGFR therapy has given hope to these patients. Unfortunately, anti-EGFR therapy only affects 10–20% of these cases. The presence of KRAS, NRAS and BRAF (V600E) mutations in the tumour have been associated with anti-EGFR therapeutic resistance. For this reason, it is critical to investigate the mutations of related genes in the tumour tissue of these cases. At our centre, KRAS (exon 2, 3, 4), NRAS (exon 2, 3, 4) and BRAF (V600E) mutations in metastatic colorectal cancer cases were analyzed using a pyrosequencing method. Of 93 analyzed patients, 51 (54.8%) mutations were detected. There were KRAS mutations in 45 (48.3%) patients, an NRAS mutation in 1 (1.07%) patient and BRAF mutations in 5 (5.37%) patients. The KRAS codon 12 mutation was present in 36 (38.7%) of 93 patients and in 70.5% of the patients with other mutations. These findings were consistent with the literature. The reliability of the pyrosequencing technique is quite high; however, the working process is quite laborious. With the NGS analysis system, more patients and genes can be studied in a shorter time and at greater cost efficiency. Therefore, there is need for cancer-specific NGS gene panels.

Keywords: KRAS, NRAS, BRAF

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Parental and Epirubicin-HCl Resistant Lung Cancer Cells Showed Different Sensitivity to Mountain Tea (*Sideritis stricta* Boiss > Heldr.) Essential Oil

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Abstract

Research conducted the consumption and value of herbal teas has increased in recent years. Indeed, in our country "dag çayı" or "yayla çayı (mountain tea)" known by the name *Sideritis stricta*, it is commonly consumed in tea form. Mountain tea is collected and consumed by local people and it is also widely sold to domestic and foreign markets in the form of tea bags prepared by grinding directly or dried plants after being collected and dried by some companies. In this study, the cytotoxic effect of essential oil obtained from Sideritis stricta, endemically grown in Antalya flora, was assessed with using two different tests as 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazoliumbromide (MTT) and CellTiter-Blue® Cell Viability in parental and epirubicin-HCl (drug) resistant non-small cell lung cancer (H1299) cells. After 24, 48 and 72 hours incubations IC50 values were calculated respectively from MTT test results, for essential oil on parental cells, 90, 76 and 60 µg/mL, for essential oil on drug resistant cells 115, 92 and 68 µg/mL. Also, after 24, 48 and 72 hours incubations IC50 values were calculated respectively from CellTiter-Blue® Cell Viability test results, for essential oil on parental cells, 75, 50 and 37 µg/mL, for essential oil on drug resistant cells 106, 84 and 69 µg/mL. Parental H1299 cells were found to be more sensitive to cytotoxic effect of the essential oil according to both tests. It has been observed that cytotoxic effect of the essential oil increased with time and concentrations on parental and drug resistant H1299 cells in both tests.

Keywords: *Sideritis stricta*, Essential oil, Cytotoxicity, Drug resistance, Lung cancer

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Apoptotic DNA fragmentation triggered by combination therapy of 5-FU and CAPE in A549 cell line

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Abstract

Non-small cell lung cancer (NSCL) is a leading cause of cancer mortality over the World. Caffeic Acid Phenethyl Ester (CAPE) is a major active component in propolis. It has been previously identified as a strong antioxidant, anti-inflammatory, antiviral and anticancer molecule. We aimed to investigate the comparative effects of 5-FU, CAPE with single and combine treatment in A549 cells. We investigate to analysis of apoptosis by DNA fragmentation in A549 cells. Thus, we further examined DNA fragmentation to clarify whether CAPE analogues induced apoptosis or not. Cells were cultured in RPMI-1640 in a humidified atmosphere of 5%CO2 at 37°C. Cell viability was determined by MTT assay. The IC50 values were detected for 5-FU, CAPE and combined treatment by 50µM, 4µM and 12,5µM +1µM respectively. We compared the effect of monotherapy and polytherapy of drugs on cells. Cells were treated with determined concentration for 24 and 48 hours. After treatment, cells were isolated according to DNA fragmentation protocol and DNA fragments showed on 3% agarose gel. For cell viability, cells were treated with IC50 value for each drug and combination 24h, 48h of incubation. Combine therapy is more effective than single therapy of these drugs. We determined that DNA fragmentation, a marker for induction of apoptosis, increased with 5-FU treatment at 48 hours. These results suggest that 5-FU is more effective than CAPE to induction of apoptosis. This study is a basic qualitative study for the investigate of the apoptosis pathway triggered by 5-FU.

Keywords: Lung cancer, CAPE, 5-Fluorouracil, Apoptosis

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Molecular identification of *Citrus cachexia viroid* (CCaVd) in citrus variants

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Abstract

Citrus growing has a remarkable production potential in our country and particularly in our region. Recently, a number of studies have been carried out on viroids; one of the disease agents in citrus fruits. In addition, 12 major viroids have been reported in citrus fruits. *Citrus cachexia viroid* (CCaVd) disease, which causes the death of trees have a substantial negative economic impact on citrus growing. This viroid has recently become a critical threat as various new rootstocks are introduced in the region. A total of 160 specimens were examined in the citrus viroid samplings held between 2015-2017, including 50 oranges (Citrus cinensis), 50 mandarins (*C. reticulata*), 50 lemons (*C.limon*) and 10 sub-notches. In some of the mandarin trees examined in Adana, Mersin and Hatay of Cukurova Region, the characteristic gummy bark formation symptom of CCaVd was observed. In samples of Satsuma and Rize mandarin varieties, the presence of CCaVd was detected by molecular applications with specific primers. By implementation of the PCR products on a 2% agarose gel, banding was recorded at 283 bp in CCaVd-infected samples and at 220 bp in non-cachexia-ethnic infected samples. Among the examined samples, 125 of them were reported to be contaminated with CCaVd while the 115 samples were contaminated with non-cachexia. PCR products were purified and transferred to the sequence analysis. Blast analyses and dendogram generation of the obtained sequences were performed using the Mega 7 program. The BLAST analyses of the selected specimens displayed 99% similarity to the registered isolates (AF213493, AB054605, DQ014514, KC584013, AJ490824, KX156936, etc.) when compared to the NCBI database isolates.

Keywords: Citrus viroids, *Citrus cachexia disease, Hop stunt viroid*, molecular detection, Phylogenetic analysis

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Use of gaseous ozone for reduction of ochratoxin A and fungal population on sultanas

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Abstract

Contaminated dried vine fruits including sultanas are regarded as an important source of ochratoxin A (OTA), a fungal secondary toxic metabolite, in human diet. In this study, sultanas were treated with gaseous ozone at 12.8 mg/L to evaluate the effects of ozonation on OTA level, fungal viability and total phenolic content. Sultanas were exposed to continuous stream of gaseous ozone up to 240 min in a treatment chamber at ambient laboratory conditions. The initial OTA level on spiked sultanas, determined as 16.7 µg/kg, decreased by 60.2 and 82.5% after 120 and 240 min of ozone exposures, respectively. Exposure to gaseous ozone for 120 min yielded more than 2.2 log reduction in the fungal population naturally present on sultanas. Ozonation did not cause a significant ($P>0.05$) change in the total phenolic content of sultanas up to 120 min of treatment. The results obtained indicate that over the 60% reduction in the level of OTA on sultanas can be achieved by gaseous ozone treatment without causing a significant decrease in total phenolic content. This study shows that gaseous ozone has a remarkable potential to degrade OTA and reduce fungal viability on sultanas.

Keywords: Gaseous ozone, sultanas, ochratoxin A, fungal viability, total phenolics

7th International
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25-27 April 2018, www.molbiotech.gen.tr

Morphological and molecular characterization of four fungal *Hebeloma* species and identification of *Hebeloma subtortum* as a new record

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Abstract

Hebeloma (Fr.) P. Kumm. (Hymenogastraceae) is an ectomycorrhizal fungus distributing in the temperate zones of the northern hemisphere. The genus was divided into thirteen sections using different characters such as habitat, smell, lamellae, presence of cortina, structure of cheilocystidia and dextrinoid reaction of the spore. 27 *Hebeloma* species have been reported in Turkey and in the current study four of them (*H. subtortum, H. mesopheum, H. cavipes, H. eburneum*) were characterized based on both microscopic/macroscopic features and molecular techniques. Structures of pileus, lamellae, stipe, basidia, spores, pileipellis, hyphae and cheilocystidia were studied as morphological characters. The nuclear ribosomal internal transcribed spacers (nrITS) region was used to determine the phylogenetic relationships among species. In the tree, *H. subtortum* and *H. mesopheum* located in sect. *Hebeloma* with their representatives retrieved from NCBI while *H. cavipes* and *H. eburneum* grouped with their representatives and caused sect. *Denudata*. *Hebeloma subtortum* and *H. mesophaeum* have almost similar spore length and width, but *H. subtortum* is differentiated by mainly ovoid spore; adnate, occasionally subdeccurent lamellae; a pruinose stipe, widened towards the base and smaller basidia. DNA sequence of H. subtortum showed 99% similarities with those of representatives. At the end of the study, we contributed to the documentation of a new record of *Hebeloma subtortum*, supported by a full description and phylogenetic results. *Hebeloma subtortum* has already been recorded in Turkey (under the names *Hebeloma mesophaeum* var. *lacteum* and *H. sordidum*), but this study appears to be the first record which is further confirmed by phylogeny.

Keywords: *Hebeloma*, ITS, Mycogenetics, New record, Phylogeny

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Comparison of 16S rRNA and nosZ denitrification functional genes as molecular markers for assessing bacterial diversity in environments

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Abstract

Bacterial denitrification in agricultural soils is a major source of nitrous oxide, a potent greenhouse gas. Denitrification is a dissimilatory process in which nitrate and nitrite are reduced to gaseous nitric oxide, nitrous oxide and molecular nitrogen when oxygen is limited, which consists of four reaction steps catalyzed by nitrate reductase (napA or narG), nitrite reductase (nirK or nirS), nitric oxide reductase (qnorB or cnorB) and nitrous oxide reductase (nosZ). The aim of this study was to investigate the comparison of 16S rRNA and nosZ genes as molecular markers in the identification of bacteria with denitrification ability. For 16S rRNA, PCR products of 49 bacteria were obtained with 27F-1492R primer pairs. For nosZ, PCR products were obtained with primers 1F-1R (259 bp), 2F-2R (267 bp) and F-1622R (453 bp) of 39 bacteria that the single nosZ band provided on the agarose gel. Following the procedure, PCR products were purified to perform sequence analyses. We compared the 16S rRNA and nosZ gene sequencing results analyzed with the GenBank and EzTaxon. The bacterial 16S rRNA gene clone library was dominated by Gammaproteobacteria and Bacilli. The nosZ clone library did not contain similar to pure culture; these sequences were most closely associated with environmental clones. Our study showed that the nosZ functional gen could be used to identify denitrification abundance in environment but could not be used to identify pure bacterial cultures. It was also found that the nosZ sequences showed uncultured denitrifier species.

Keywords: 16S rRNA, nosZ, Denitrification, Phylogeny, Molecular markers

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Production of γ-poly(glutamic acid) using feather hydrolysate as fermentation substrate

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Abstract

Polyglutamic acid (PGA), which is water-soluble and biodegradable, can be used for numerous applications. One of the significant challenges of producing PGA at an industrial scale is its cost. As an effort to move towards feasible PGA production, feather hydrolysate (FH) derived from enzymatic hydrolysis of feather was used to produce PGA. 30-L fermentation was realized to obtain keratinase using *S.pactum* DSM 40530. Fermentation broth retentate were concentrated after centrifugation by using cross-flow filtration. When the total volume was decreased by a factor of 15, the volumetric enzyme activity increased by 8.75-fold and 8×103 U L-1d-1 of enzyme activity was the optimum for achieving 75% feather degradation per gram of feather. 40 g/L of FH was used with different media compositions using *B.licheniformis* 9945a. Among four different cultivation where L-glutamate, tri-sodium citrate and glycerol were used, highest yields of both γ-PGA and cell dry matter (CDM) were obtained from cultivation 1, at 5.4 and 8.6 g/L, respectively, despite culture media did not contain glutamic acid, an essential precursor for γ-PGA production. In cultivation 2, which was not only missing glutamate but also citrate, the γ-PGA and CDM yields decreased to 3.2 and 7.8 g/L, respectively whereas it was only 1.94 and 4.2 g/L when FH was used as the sole substrate in cultivation 3. When cultivation 4 was adopted where only glycerol was missing, the γ-PGA and CDM yields slightly increased to 2.3 and 5.46 g/L, respectively. This is the first study that achieved the production of PGA from FH.

Keywords: γ-Poly(glutamic acid); γ-PGA; Feather hydrolysate; Keratinolytic activity; Feather

7th International
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Congress
25-27 April 2018, www.molbiotech.gen.tr

Investigation of *las* and *rhl* quorum-sensing systems in clinical isolates of ceftazidime resistant *Pseudomonas aeruginosa*

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Abstract

Pseudomonas aeruginosa is a Gram-negative, common opportunistic pathogen in immunocompromised patients and is responsible for causing a huge variety of infections with important levels of morbidity and mortality. P. aeruginosa has also numerous virulence factors. Production and regulation of these virulence factors depend on intercellular communication systems called Quorum Sensing (QS). Bacterial cell density reaching a certain threshold QS triggers the expression of many virulence factor genes (*lasI, lasR, rhII and rhIR*). This study aims to investigate of ceftazidime resistant *P. aeruginosa* strains from different sources in terms of virulence factors that is, QS capability. For this study, ceftadizime resistant *P. aeuginosa* strains (n:50) causing clinical infections were isolated from patients in intensive care, wound, and infection units of Samsun Education and Research Hospital in Turkey. The strains were analysed for the production of several virulence factors such as N-acylhomoserine lactone, swimming, twitching, and swarming controlled via mediated QS. Then, the capacity of biofilm formation was investigated by microtitration plate method. The existence of *lasI, lasR, rhII and rhIR* genes which are under the control of QS genes for the synthesis virulence factors were investigated with PCR. *P. aeuginosa* ATCC 15692 was served as a positive control. As a result of the study, twenty-nine strains expressed HSL and they were recorded as QS (+). Remaining twenty-one strains were recorded as QS (-). *lasI* gene was shown in 48 strains, lasR gene in 46, *rhlI* gene was shown in 41 QS(+) strains, *rhlR* was shown in 36. Our results show once again that virulence factors have important role in intercellular communication systems.

Keywords: *Pseudomonas aeuginosa*, Virulence factors, Quorum Sensing

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Genetic identification of biocatalysts from functional metagenomic DNA library

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Abstract

Employing metagenomic approach, researchers are able to decode the genetic patrimony of unculturable microbes, but the DNA sequence information alone is not enough to determine the gene function and this is the main challenge of sequence-based metagenomics. Functional metagenomic which consists on the cloning and expression of metagenomic DNA and screening for enzymatic activity can be the right approach to bioprospect known or/and unknown enzymes. Library of over 108,000 EPI300T1R of pCC1FOS fosmid recombinant about 40 K of metagenomic DNA was constructed from microbiota of Malaysian palm oil mill effluent and screened with a cocktail of three fluorescent substrates; methylumbelliferyl-ß-D-glucopyranoside (MUGlc), methylumbelliferyl-ß-D-cellobioside (MUC) and chlorocoumarin-xylobioside (CCX) to detect endo-glucanase, ß-glucosidase and endo-xylanase activities. The high-throughput screening of the library indicated high number of clones with fluorescence signal and 100 high rated clones were selected for sequencing with Next-Generation Sequencing strategy. Over 80 probable cellulose-degrading enzymes and over 30 probable xylan-degrading enzymes were found in the NGS-data.

Keywords: Functional metagenomics, high-throughput screening, next-generation sequencing, cellulose-degrading enzymes, xylan-degrading enzymes

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Pyrethroid resistance and distribution of kdr allele in field populations of *Culex pipiens* in Turkey

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Abstract

Mosquitoes within the *Culex pipiens* complex have been implicated as major vectors for several pathogens responsible for infectious human diseases. Vector control is important in terms of mosquito-borne disease prevention and management. Insecticides are used extensively to control mosquito insect vectors. Due to their high efficacy, rapid rate of knockdown, low mammalian toxicity and less environmental impact, pyrethroid insecticides are currently being promoted worldwide for disease vector. The widespread and improper use of pyrethroid insecticides has resulted in the evolution of resistance in many mosquito species, including *C. pipiens*. Previous studies demonstrated that pyrethroid insecticide resistance is caused by point mutations in the S6 transmembrane segment of domain II of the para-homologous voltage gated sodium channels in the *C. pipiens*. In the majority of cases, an A→T transition at position 1014 is observed, resulting in a leucine to phenylalanine (L1014F) substitution. In this study field collected mosquito specimens were identified individually using Sanger Sequencing with standard insect DNA barcoding primers targeting fragment of cytochrome oxidase I gene. gDNA samples belonging to *C. pipiens* were monitored in order to detect kdr allele frequency by allele-specific PCR. The results showed that the distribution of the L1014F kdr mutation is widespread and the kdr mutant alleles in all populations were mostly in heterozygous condition. These data provide suitable information for the design and implementation of successful resistance management strategies against this species.

Keywords: *Culex pipiens*, pyrethroid resistance, kdr, allele-specific PCR

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Effects of stearic acid on programmed cell death mechanisms of the fission yeast (*S. pombe*)

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Abstract

Stearic acid (SA) is a type of saturated LCFA (long chain fatty acids) including 18 carbon atoms. SA is found in many foods and oils, and also used in pharmaceutical industry. Some researchers have reported significant inhibition of p21 and PI3K/Akt signaling in response to dietary uptake of SA. We evaluated apoptotic effects of SA (300-1500 μM) in the fission yeast (*S. pombe*), which is a uni-cellular model organism and also known as micro-mammal. Effects of SA on cell proliferation and viability were assessed using hemocytometer and methylene blue staining method. For visualizing nuclear morphology, nucleus was stained with DAPI and acridine orange/ethidium bromide (AO/EB) dual stain. DNA fragmentation and nuclear condensation were observed between 900-1500 μM doses. 10-90% of cells showed apoptotic nuclear morphology in correlation with increasing doses of SA. The results were validated with AO/EB dual staining. In addition, expression of *S. pombe* caspases, Pca1 and Sprad9, markedly increased. The potential effects of SA on cell proliferation and programmed cell death mechanisms were shown in unicellular model fungi, *S. pombe*. Besides, *S. pombe* was evaluated as a new model organism in molecular toxicology and cell death research.

Keywords: Stearic acid, apoptosis, DNA fragmentation, *S. pombe*

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Inhibition of nanotoxic effect of zinc oxide by resveratrol

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Abstract

Zinc oxide (ZnO) is a compound that has harmful effects as well as being used in many different areas. Numerous studies have been carried out to minimize the toxic effects of ZnO nanoparticles (NPs). In the present study, the protective role of resveratrol (RSV), a potent antioxidant polyphenol substance, was examined against ZnO-induced nanotoxicity on human pulmonary alveolar epithelial cells (HPAEpiC). In this context, the cytotoxic and genotoxic effects of different concentrations of RSV (5, 10, 20 mg/L) and ZnO NPs on the cells were measured alone and in combination. At the same time, the effects of aforementioned applications on the total antioxidant capacity (TAC) level in HPAEpiC were assessed. The results obtained showed that ZnO NPs alone significantly increased cytotoxicity and genotoxicity on cells compared to negative control (control (-)). In the experiments performed with RSV + ZnO NP combination, cytotoxic and genotoxic activity decreased at the level of $p < 0.05$ especially at 20 mg/L application of RSV. When the level of TAC in cells was examined, a concentration-dependent increase was detected between TAC and RSV. It was determined that ZnO NPs reduced the TAC level statistically ($p < 0.05$) in comparison with control (-). In conclusion, the present study revealed that RSV, a natural antioxidant, showed protective property against genotoxic and cytotoxic damage induced by ZnO NPs on HPAEpiC.

Keywords: Antioxidant, Nanotoxicity, Resveratrol, Zinc oxide

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Preparation of magnetic CuFe2O4 and reduced graphene oxide nanocomposite for L-Cysteine detection

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Abstract

Graphene has gained tremendous interest as a supporting material due to its large surface area, high conductivity, ionic mobility, and superior mechanical flexibility [1] Graphene can be synthesized by two reactions: (1) chemical oxidation of graphite to graphene oxide (GO) and (2) reduction reactions. GO is a two dimensional (2D) carbon material with a large specific surface area, multiple aromatic regions and hydrophilic oxygen groups [2]. CuFe2O4 has received great attention and is widely used in sensors, electronics and catalysts in recent years [3]. In this study, I report the synthesis of a copper ferrite-reduced graphene oxide nanocomposite. The nanocomposite was characterized by X-ray diffraction (XRD), scanning electron microscope (SEM) and Fourier-transform infrared (FTIR) spectroscopy. Detailed investigations of the detection of L-cysteine was carried out using Cyclic Voltammetry (CV).

Keywords: Copper ferrite, electrochemical, reduced graphene oxide, L-cysteine

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Preparation, caharacterization, biological and sensor application of copper nanoparticles (CuNPs) based on nitrogen-doped porous carbon particles (GQDs)

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Abstract

Nitrogen-doped porous carbon particles, the next generation carbon based nanomaterials, due to their outstanding physical, chemical and biological properties, have potential in revolutionizing the future of nanomedicine and biotechnology. Hence recently, many studies were conducted on graphene quantum dots nanomaterials. Here, copper nanoparticles (CuNPs) were synthesized using nitrogen-doped porous carbon nanomaterials (GQDs) as reducing reagent and stabilizer. Compound was characterized by UV-Vis, FT-IR spectroscopy, transmission electronmicroscopy (TEM) and thermogravimetric analysis (TGA). UV-Vis spectroscopy studies of the interactions between the CuNPs and GQDs with calf thymus DNA (CT-DNA) showed that the compound interacts with CT-DNA via electrostatic binding. The DNA cleavage activity of the CuNPs and GQDs was studied by agarose gel electrophoresis method. pBR322 DNA (0.1 µg µL-1) in Tris-HCl buffer (100 mM, pH:7,2) treated with the compound at 37 ^{0}C for 3 h. DNA cleavage study showed that the CuNPs and GQDs cleaved DNA without any external agents.

Support from Canakkale Onsekiz Mart University, The Scientific Research Commission (ÇOMÜ-FBA: 2018-1291) is greatly acknowledged.

Keywords: Copper nanoparticles (CuNPs), Nitrogen-doped porous carbon particles, Calf thymus DNA (CT-DNA), DNA cleavage, DNA binding

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Preparation and characterization of BSA-gold nanoparticle conjugates

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Abstract

In this study, we have conjugated different ratios of bovine serum albumin (BSA) protein with TAMRA labelled gold nanoparticles (AuNPs) and characterized these conjugates. Gold nanoparticles were synthesized in the aqueous medium using sodium citrate acted as both reducing and capping agent. Polyethylene Glycol (PEG) chains were used for passivation. The particles were then labelled with TAMRA dye. The nanoparticle formation was confirmed with its characteristic surface plasmon absorption band observed at 521 nm. In addition, transmission electron microscopy analysis revealed the average particle size to be about 20 nm. BSA model protein was conjugated to PEGylated AuNPs via EDC/NHS chemistry. The bio-conjugation process was investigated using the measurements of optical density, fluorescence intensity, Dynamic light scattering, gel electrophoresis and zeta potential. Fluorescence intensity was found to be increased in proportion to BSA ratios. After conjugation, the zeta potential of the resulting AuNPs was reduced from -31,2 mV to -20,1 mV in this experiment. The studies on the conjugation of biomolecules onto nanoparticles have been increasing day by day. However, it is extremely important to determine whether these biomolecules maintain their biological efficacy prior to biological or medical applications. Therefore, before assessing the bioactivity of prepared bioconjugates, irrelevant model biostructures such as proteins, antibodies or oligonucleotides whose effects have already been known, should be used. The clarification of conjugation and interaction with the model protein BSA will further enrich the nanomedicine field by developing and conjugating nanotherapeutic agents especially nanoparticle-protein conjugates.

Keywords: Gold nanoparticles, Polyethylene Glycol, Bovine Serum Albumin, bio-conjugation, nanoparticle-protein interaction

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Preconcentration of Al(III) by coriolus versicolor immobilized γ-Fe2O3 nanoparticles prior to its determination by ICP-OES

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Abstract

This study investigated utilization of Coriolus versicolor loaded with γ-Fe2O3 nanoparticles as a biosorbent for magnetic solid phase extraction (MSPE) and detection of trace levels of Al(III) from some environmental and food samples. The surface structure of immobilized *C. versicolor* was characterized by FT-IR, SEM and EDX. The effects of pH, flow rate, quantities of *C. versicolor* and γ-Fe2O3 nanoparticles, eluent type, concentration and volume, foreing ions and sample volume were tested for optimization of the process. The best experimental conditions were found as pH 6.0, 2.0 mL min-1 flow rate, 100 mg amount of *C. versicolor* on 150 mg amount of γ-Fe2O3 magnetic nanoparticles, 5.0 mL of 1.0 mol L−1 HCl as eluent, and 500 mL of sample volume. The limit of detection and preconcentration factor were achieved as 0.03 ng mL-1 and 100, respectively. Accuracy of the recommended process was tested by recovery measurements on the certificated reference materials and high recoveries (≥95%) with low RSDs were obtained. The developed process was successfully applied for quantification recovery of Al(III) in various environmental and food samples.

Keywords: *Coriolus versicolor*, Aluminium, Preconcentration, Biosorbent, Magnetic solid phase extraction

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Molecular and screening assay in nematode-viroid interactions in *Kalanchoe daigremontiana*

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Abstract

Root knot nematodes are important nematode group cause crop losses in many plant species. They also interact with many organisms including viroid. As a minuscule pathogen, Viroid consists of a short strand of circular, single-stranded RNA without protein coat. They are inhabitants of higher plants and cause diseases. Hop stunt viroid (HpSVd) and *Potato spindle tuber viroid* (PSTVd) are most damaging viroid disease agents. Nematode and viroid interactions may cause devastating effect on plants. However, the effect of both pathogen interactions on plants have not been fully understood. Therefore, this study was conducted to determine the nematode and viroid effects on an indicator plant, *Kalanchoe daigremontiana* using molecular and screening assay. For this aim, *Meloidogyne incognita* with HpSVd and PSTVd viroid were inoculated to *K. daigremontiana* to determine the interactions among them. The experiment was set up as the infection of nematode, PSTVd, HpSVd, nematode+HpSVd, nematode+PSTVd, PSTVd+HpSVd and PSTVd+HpSVd+nematode. RNA extraction and screening assay were achieved following the mechanical inoculation, and specific primers were used for the detection of viroids. Results revealed that the replication of circular RNA of viroid in all infected plants were detected in PSTVd, HpSVd, nematode+HpSVd, nematode+PSTVd, PSTVd+HpSVd, PSTVd+ HpSVd+nematode samples apart from control and solely nematode infected plants. Decreased plant growth was observed in both nematode and viroid inoculated plants, and molecular and screening results showed parallelism. Results indicate that this study is a leading research on nematode-viroid interactions in *K. daigremontiana* that may provide a useful resource for future studies.

Keywords: *Melodiogyne incognita, Hop stunt viroid, Potato spindle tuber viroid, Kalanchoe daigremontiana*

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Biological activity evaluation of most popular edible plant-Salsify (*Tragopogon porrifolius*) in Sivas

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Abstract

Tragopogon porrifolius belongs to the Asteraceae family, is an annual or biennial plant. It is subdivided into three subspecies as *T. porrifolius* subsp. *australis*, *T. porrifolius* subsp. *cupani* and *T. porrifolius* subsp *porrifolius*. The plant is known as ''Yemlik'' among the local people. The roots, leafy shoots, and open flowers of this plant is consumed in Southern and Central Europe, North America and United Kingdom and further is also used to treat cancer in Lebanese folk medicine. *Tragopogon porrifolius* has antioxidant activity due to some phenolic acids and flavonoids. In addition to that this plant has monounsaturated and essential fatty acids, vitamins and polyphenols components. The plant materials were collected from natural habitat before flowering stage. In this work, GC-MS was used for characterization of the chemical composition of ethanol extracts from *T. porrifolius*. In vitro antioxidant activity as well as some enzyme inhibitory activities such as α-glucosidase, α-amylase, acetylcholinesterase and butyrylcholinesterase have been examined on the extract. GC-MS results indicate that T. porrifolius ethanol extract have 4H-Pyran-4-one (15.0%), Benzeneacetaldehyde, Isosorbide as major constituents. The extract demonstrated potent antioxidant activity in a concentration dependent manner. The *T. porrifolius* extract exhibited higher levels of ABTS scavenging activity than ABTS. The total phenol and flavonoid content assay results demonstrated that T. porrifolius contains quercetine equivalent 12.33 ± 0.23 mg/g flavonoid and gallic acid equivalent 74.71 ± 5.59 mg/g phenolic constituents. As for the enzyme inhibition activity, the extract exhibited strong inhibition activity on the tested five enzymes such as AChE, BChE, α-glucosidase, α-amylase and tyrosinase at concentration of 2 mg/mL. The identification of AChE, BChE, α-glucosidase and α-amylase inhibitory activity in *T. porrifolius* support the possible use of the plant as functional food for the management of Alzheimer's and diabetes. The study results support the traditional use of this plant among the Turkish people scientifically. The results of this study will shed light on the further study on the plant as well as on the carrying out of biological activity guided isolation of active compounds.

Keywords: *Tragopogon porrifolius, in-vitro*, antioxidant, enzyme-inhibition, GC-MS

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High-throughput genomic simple sequence repeat (SSR) marker development and construction of a high resolution physical map in chickpea (Cicer arietinum L.) genome

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Abstract

Chickpea (Cicer arietinum L.) (2n=16) is the second most important legume crop in the world, cultivated globally in arid and semi-arid regions. It is a rich source of protein, dietary fibers, carbohydrates and minerals. In addition to its high nutritional value, the crop supports soil fertility via fixation of atmospheric nitrogen. The present research reports the development of 29,207 novel simple sequence repeat (SSR) markers specific to C. arietinum genome. A bioinformatic approach with high-stringency repeat identification criteria was utilized in order to mine C. arietinum chromosomes for simple sequence repeats, resulting in 67,816 identified repeat loci. Repeat loci were further converted to 29,207 PCR markers using high-stringency marker design parameters. The markers are well-distributed among the eight C. arietinum chromosomes with an average distance of 16.5 kb between adjacent markers. A physical distance map of the C. arietinum genome was constructed based on absolute marker positions. A set of newly developed markers that represent all eight C. arietinum chromosomes was validated with laboratory experiments in order to prove the amplificability of markers generated through bioinformatic analyses. As a result of the present research, C. arietinum genome was saturated with almost 30,000 genome-specific markers with known absolute positions along physical chromosomes. The large number of, C. arietinum specific DNA markers introduced in the present work constitute a valuable resource for molecular genetic research in chickpea, including germplasm characterization and preservation, and mapping genes/QTLs (Quantitative Trait Loci) that control relevant traits (e.g. disease resistance, drought and salinity tolerance) in chickpea production.

Keywords: Molecular genetics, Bioinformatics, Sequence-specific markers, DNA markers

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Development of novel sequence-based markers linked to CMV (*Cucumber Mosaic Virus*) resistance in pepper (*Capsicum annuum L.*)

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Abstract

CMV (*Cucumber Mosaic Virus*) is one of the earliest known plant diseases with a broad host range, affecting hundreds of crop species including members of the *Solanaceae* family. CMV resistance in *Capsicum annuum* has long been identified as a multigenic trait, requiring the introgression of multiple loci from resistance sources for breeding toward CMV resistance. In 2010, a single dominant gene was identified in the short arm region of *C. annuum* LG2 (Linkage group 2), at a location syntenic to ToMV resistance locus in tomato genome. Yet, a tightly linked marker was not defined and the closest marker was mapped at 2 cM away from the CMV resistance locus. In the present work, bioinformatic analysis was performed in order to saturate the short arm region of *C. annuum* LG2 with simple sequence repeat (SSR) markers and primers were designed that flank the repeat loci. A total of 10 primer pairs were used amplify the SSR loci from a set of CMV resistant and susceptible *C. annuum* genotypes. Among the 10 SSR markers, three displayed polymorphisms among the tested *C. annuum* genotypes. More importantly, allelic distribution of two polymorphic SSR markers correlated with the CMV resistance status of the tested genotypes. Thus, the two markers developed specific to the short arm of pepper chromosome 2 represent candidate loci for the selection of CMV resistance in pepper breeding programs.

Keywords: Marker assisted selection, Molecular breeding, Simple sequence repeat markers, Disease resistance

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Identification and regulation of antiporters in strawberry (*Fragaria X ananassa*) under salinity stress

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Abstract

New oral anticoagulants including dabigatran are commonly used in the prophylaxis and treatment of systemic embolism and deep vein thrombosis worldwide. Cancer patients have also an increased risk of developing venous thromboembolic events or may have other indications for anticoagulation, such as atrial fibrillation. However, several data suggest that anticoagulant drugs may have an effect on tumor development and progression. In this study, we aimed to investigate the cytotoxic effects of dabigatran on a cancer cell line HeLa cells derived from human cervical cancer. Cells were placed in 96-well culture at an initial density of 50.000 cells/ml in six replicates and incubated in the Dulbecco's Modified Eagle's Medium (DMEM)/Ham's F12 supplemented with 10% fetal bovine serum (FBS). Following incubation, the cells were treated with six dilutions of the test material [Pradaxa (dabigatran etexilate, 150 mg)™]. Test material was prepared in culture medium supplemented with 1% dimethyl sulfoxide. Stock solution were prepared as 0.30 g/20 ml for the initial dose. Stock solution underwent serial dilution and were prepared in five dilutions and only DMEM/F12 medium was served as control groups. The cell viability was determined by MTT assay. At 24-hour incubation, the cells exposed to all dilutions of dabigatran showed a significant difference compared to normal fibroblastic morphology. The cells displayed cellular alterations including nuclear condensation, rounded morphology, and cell degeneration. The viability of HeLa cells was examined at 24 and 48 post-incubation hours. At 24 and 48 hours, dabigatran showed a cytotoxic effect in all dilutions. The results showed that dabigatran may reduce proliferation of cancer cells.

Keywords: Dabigatran, Cell Culture, Cytotoxicity, HeLa cells

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Synthesis of sericin capped silver nanoparticles for use as bioactive agent in wound dressing materials

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Abstract

In this work, silk sericin was used as reducing and coating agent to prepare sericin capped silver nanoparticles (S-AgNPs) which can be used as antibacterial agents in wound dressings. The sericin, which makes up 25-30 % of the silk, has properties such as excellent oxygen permeability, antioxidant effect, moisture regulating ability and antibacterial activity. Because of these properties, studies on silk sericin for wound healing give promising results. For S-AgNPs synthesis, 1 mM, 5 mM and 10 mM AgNO3 solutions were prepared and 10 mL of each AgNO3 solution was transferred to a 50 mL Erlenmeyer. While the AgNO3 solutions were stirred at high speed with magnetic stirrer, 10 mL of 1 % sericin solution was added to each AgNO3 solution after the pH of the sericin solution was adjusted to 11 with NaOH. The mixture was stirred at room temperature overnight. The transparent solution which turned yellow-brown indicated the formation of S-AgNPs. AgNPs formation was also determined by measuring the absorbance spectra of S-AgNPs between 300 and 600 nm using UV-Vis spectrophotometer. The aqueous stability and size of S-AgNPs were investigated by zeta potential measurements. All of the S-AgNPs synthesized were found to have negative zeta potential and their size was found to be within the range of 47.06 to 54.86 nm on average. To determine the antimicrobial properties of S-AgNPs, agar-well diffusion and minimum inhibitory concentration (MIC) tests were performed. 5 mM and 10 mM S-AgNPs groups showed antimicrobial activity on both gram-negative *Escherichia coli* (ATCC 25922) and gram-positive *Staphylococcus aureus* (ATCC 6538). Synthesized S-AgNPs have the capacity to be used as antibacterial agents in wound dressings.

Keywords: *Bombyx mori* silkworm cocoon, Sericin, Silver nanoparticles, Antibacterial agents, Wound dressing

7th International
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Congress
25-27 April 2018, www.molbiotech.gen.tr

The Association between androgen related genes polymorphisms and idiopathic male infertility

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Abstract

Androgens have an unignorable role in the development of reproductive organs, male sexual function, puberty as well as male fertility. Androgen receptor (*AR*), steroid 5-alpha reductase 2 (*SRD5A2*) and tumor necrosis factor-alpha (*TNF-α*) genes are androgen related genes that are involved in androgen biosynthesis and metabolism. In our study, the aim was to investigate the relationship between polymorphisms of *AR, SRD5A2* and *TNF-α* genes, and idiopathic male infertility. In this study, 335 idiopathic infertile male patients and 142 fertile controls were recruited. Peripheral blood sample was collected from each of the participants and the genomic DNA was isolated from these blood samples using salting out procedure. The genotyping of *SRD5A2* and *TNF-α* genes was performed by restriction fragment length polymorphism (RFLP) method, and CAG repeat polymorphism of *AR* gene was evaluated by polyacrylamide gel electrophoresis. We found a significant association between the polymorphisms of *AR* and *SRD5A2* genes, and idiopathic male infertility ($p=0.015$ and $p<0.005$, respectively). However, we found no statistically significant association between *TNF-α* ($p>0.005$) polymorphism and idiopathic male infertility. In view of these findings, the polymorphisms of *AR* and *SRD5A2* genes may take part in idiopathic male infertility and might be contributory factors to its etiology.

Keywords: *AR* gene, idiopathic infertile, *SRD5A2, TNF-α*

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In vitro antimicrobial activity and wound healing potential of wild *Hypericum lydium* Boiss. from Turkey

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Abstract

The genus *Hypericum*, a member of Hypericaceae family, is represented by 100 taxa, 45 being endemic to Turkey. The genus *Hypericum* sp. has been used for the treatment of burns, eczema, ulcers, diarrhea, hemorrhoids as well as wounds in traditional medicine. It is not known whether *Hypericum lydium* Boiss. has any properties related to the antimicrobial effect of oral microorganisms and the healing of wounds. The present study was designed to investigate the in vitro antimicrobial, anti-collagenase, anti-hyaluronidase and anti-elastase activities of the ethanol extract from the aerial parts of *H. lydium*. The ethanol extract of *H. lydium* showed antibacterial as well as antifungal property. In the study, *Streptococcus sanguinis* (ATCC10556) (MIC: 1.0 mg/mL) and *Streptococcus mutans* (ATCC25175) (MIC: 2.0 mg/mL) were relatively sensitive, while *Staphylococcus aureus* (ATCC25923) (MIC: 32.0 mg/mL) and *Candida albicans* (ATCC10239) (MIC: 8.0 mg/mL) were more resistant to the extract. The extract could inhibit collagenase, hyaluronidase and elastase activity with values of 26.3, 14.2 and 80.27%, respectively at 1 mg/mL concentrations. These findings indicate that *H. lydium* can be used as a promising agent in mouthwash for curing periodontal diseases and in dentistry for the healing of oral injuries.

Keywords: *H. lydium*, antimicrobial, wound healing

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Effects of different sized silica nanoparticle on ultrastructure of rat brain

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Abstract

Silica is among the most popular nanoparticles and is used, besides many areas including agriculture, food, cosmetics and medicine. It is not clear how silica nanoparticles affect brain tissue. In this study, we aimed to investigate the ultrastructural effects of SiO2 NPs in rat brain. Twenty eight male Wistar albino rats were divided into four groups (n=7 rats) as group I (control), group II (6 nm), group III (20 nm) and group IV (50 nm). The rats in the experimental group were exposed to intraperitoneally 150 µg/mL per day SiO2NPs for 28 days and the rats in the control group were treated with 1 mL saline for the same period. 24 h after the last exposure, rats were sacrificed and their brains were removed. Brain tissue sections of were examined ultrastructurally. In the control group, neurons, myelinated and unmyelinated nerve fibers, glial cells and perivascular area were found to be normal in structure. In the myelin sheath of nerve fibers and axoplasms of myelinated and unmyelinated nerve fibers that have degenerative changes were observed in 6 nm, 20 nm and 50 nm groups. In addition, perivascular edema, nuclear and intracytoplasmic vacuols in some neurons were observed in the 50 nm group. The findings of this study show that intraperitoneal administration of 6, 20 and 50 nm sized SiO2 NPs cause structural changes in brain cells. This result suggests that SiO2 NPs may be a potential risk for neurodegenerative diseases.

Keywords: Brain, nanoparticle, electron microscopy, neuron, glial cells

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Investigation of microRNAs affected by high fructose diet in kidney tissues

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Abstract

Increase in total sugar and especially fructose intake in daily energy consumption might lead to the development of obesity, type 2 diabetes, metabolic syndrome and fatty liver. Scientific studies have shown that miRNAs might be associated with metabolic syndrome related pathologies; especially in the formation or prevention of insulin resistance which lead us to consider miRNAs as potential therapeutic targets. In this project, it was aimed to investigate some miRNAs associated with insulin resistance and antioxidant systems in metabolic syndrome caused by high fructose diet. Within the scope of the study, animal model of metabolic syndrome was formed by giving rats high fructose (20%) in drinking water and TNF-α, IL-1β, IL-6, IL-10 and NFκB levels, which are markers of inflammation in renal tissues, were determined by ELISA method. In addition, expression levels of miR-135a (5p), miR-200a (3p), miR-125a (5p), miR-195 (5p), miR-103 (3p) which were considered to regulate insülin resistance and antioxidant systems were measured with real time quantitative PCR (qRT-PCR). Gene expression levels of insulin-PI3K-Akt signaling pathway genes (insulin receptor, IRS1 / 2, PI3K, Akt, mTOR) and major antioxidant enzymes (cat, sod, gpx, gst), which are the target genes of these miRNAs were measured and correlated with changes in miRNA levels. The results showed an increase in tissue inflammatory markers in kidney tissues of rats fed with high fructose diet. Besides, almost all the antioxidant enzymes' and expression levels of irs1 and pi3k in the signal transduction pathway of insulin were found to be significantly suppressed as compared to the control group. This suppression in gene expression may be attributed to a significant increase in miR-103, miR-125 and miR-195 levels that likely to regulate these pathways. These results indicate that fructose can regulate antioxidant systems in kidney tissues via miRNA molecules.

Keywords: Diabetes, Kidney, Oxidative stress, Inflammation, Resveratrol, İnsulin signaling Pathway, Gene expression.

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Determination variations encountered on KATP protein encoding genes in Raynaud's phenomenon cases

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Abstract

Raynaud's phenomenon (RP) is a vascular disorder characterized by recurrent vasospastic response of the fingers and toes to cold or emotional stimuli. Classically ischemia, deoxygenation and hyperemia are the sequence of a typical attack. RP is a relatively common disorder in worldwide population with the prevalence of 3.3% to 22%. ATP-dependent potassium channels (KATP) containing Kir6.1 and SUR2A proteins (KCNJ8/ABCC9 genes), particularly in the regulation of vascular tone in the coronary arteries has a critical role and deficiency or defects in the function can cause vasospasm associated with Prinzmetal's angina. It would be important to determine whether variations of KATP genes related to Raynaud's phenomenon is thought to be associated with vasospasm. It is believed that the studies describing mechanisms involved in the pathogenesis of inherited vascular disorders offers the best opportunity for investigation of the early stages of pathogenicity and diagnosis of Raynaud's phenomenon and associated other diseases. The purpose of this study, KATP channel which is gene coding of run across mutations in vasospasm associated with Raynaud's phenomenon in patients to determine the characterization and investigation of mutation frequency. In our study; the cases with Raynaud's phenomenon, the relation between the variation in the KCNJ8/ABCC9 genes (S422L/V734I) was examined. 50 subjects who were diagnosed with Raynaud's phenomenon (patient group) and 50 healthy subjects (control group) were included in the study. Variations were determined using the Tetra-Primer ARMS PCR method. KATP channel protein variants analysed for possible correlations among Raynaud's phenomenon were not observed in patient and control groups.

Keywords: Raynaud's Phenomenon, KATP Channel Proteins, KCNJ8/ABCC9 Genes, S422L/V734I Variants.

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Investigation of miRNA profiles in patient groups with st elevation acute myocardial ınfarction and non-ST elevation acute myocardial ınfarction

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Abstract

Coronary artery disease (CAD) is the leading cause of deaths in our country and all over although improved diagnosis, treatment and prevention methods in recent years, Due to the sudden occurrence and unpredictability, the determination of early diagnostic markers of Acute myocardial infarction (AMI) is very important. Today, alongside the improved diagnostic methods, in recent years another markers which might be associated with a diagnosis of AMI are miRNAs molecules. Certain miRNAs were shown to play a role in the pathogenesis of atherosclerosis and in regulating cardiac functions. In our study we aimed to investigate hsa-hsa-miR-1, hsa-miR-25-3p, hsa-miR-30d-5p, hsa-miR-34a-5p, hsa-miR-92a-3p, hsa-miR-133a-3p, hsa-miR-150, hsa-miR208a-3p, hsa-miR-221-3p, hsa-miR374a-5p and hsa-miR499a-5p expression levels in patients who had been diagnosed with AMI with ST elevation and non-ST elevation. 25 ST-segment elevation and 25 non-ST elevation patients with the diagnosis myocardial infarction and 20 healthy control group were enrolled in the study. Blood samples were taken from patients and controls to 5ml EDTA tubes, centrifuged at 2000xg for 10 minutes and then the plasma was separated. miRNAs were isolated by the plasma miRNA isolation kit. Isolated miRNAs was transformed to cDNA by Reverse Transcription kit and miRNA expression analysis was performed using high capacity Real-Time PCR System from cDNAs on Dynamic Arrays. There is no significant increase or decrease detected in the expression miRNA levels in the patients group compared to control group ($p>0.05$). In conclusion, miRNAs may be an early biomarker for the diagnosis of AMI however further and larger studies are needed.

Keywords: microRNA, acute coronary syndrome, acute myocardial infarction

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Effect of paricalcitol on paroxanase and arylesterase activities in cardiac tissue of rats exposed to radiofrequency radiation (1800 MHz)

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Abstract

The use of wireless technology is increasing day by day in relation to advancing technology. 1800 MHz radio frequency (RF) radiation is used in wireless technology. RF radiation affects the proliferation, differentiation and apoptotic process of the cell by making changes in the plasma membrane function and gene expression of the cell with thermal and non-thermal effects. Different studies have shown that RF waves cause the formation of reactive oxygen species and lipid peroxidation, disrupt biomolecules structure, alter enzyme activities, cause cell damage and cell death. Antioxidants are widely used to prevent the formation of ROS and the damage it causes. The aim of this study was to investigate the possible toxic effects of 1800 MHz RF radiation on the heart and the role of paracalcitol, a vitamin D vitamin analgesic in eliminating these effects. Twenty eight 8-10 week old male Wistar rats were used in the study. Rats; 4 groups were divided into Group I (control), Group II (only RF applied), Group III (only paricalcitol) and Group IV (RF + Paricalcitol). No treatment was performed in Group I (n = 7). Group II received 1800 MHz RF (1 hour / 30 days). Group III was injected subcutaneously with 0.02 µg / kg paricalcitol (3 times / week for 30 days). Group IV received 1800 MHz RF (1 hour per day for 30 days) and 0.02 µg / kg paricalcitol (3 times per week / 30 days). At the end of thirty days of treatment, the heart tissues obtained from the sacrificed rats were homogenized and the paraoxanase (PON) and arylesterase (ARE) enzyme activities were evaluated in these tissues. In Group II, PON and ARE enzyme activities were significantly lower than Group I ($p < 0.05$). In Group III, PON and ARE activities were significantly increased in all groups ($p < 0.05$). PON activity increased significantly in Group IV compared to Group II ($p < 0.05$). ARE activity increased in Group IV compared to Group II, but this increase was not statistically significant ($p > 0.05$). As a result of the study, paricalcitol was thought to play an important role in eliminating the oxidative damage caused by RF waves in the heart, especially by increasing PON enzyme activity.

Keywords: Electromagnetic field, paraoxonase, arylesterase

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The effect of high cholesterol diet on expression of scavenger receptors and kidney damage

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Abstract

Hypercholesterolemia plays an important role especially in cardiovascular diseases, chronic kidney diseases, obesity, metabolic syndrome and neurodegenerative diseases. As a result of increased LDL levels, the amount of oxLDL increases with the oxidation of LDL. The forming oxLDL is taken up by many scavenger receptors (SR). Modified LDL stimulated activation of intracellular signaling pathways might leads an increase in lipid accumulation, foam cell formation, apoptosis, inflammation and fibrosis. The aim of our work; is to investigate if high cholesterol diet effects SRs expressions and various transcription factors that might be related with kidney damage of rabbit. In this purpose, mRNA expressions of well-identified SRs (SCARA3, SRA, SRB1, CD36, CD68, LOX1, SRF1, SRI, SRG) and following transcription factors (LXR, PPAR) were measured by qPCR in addition to the protein levels of transcription factors (ABCA1, SREBP, PPAR) that regulate modified lipid-scavenger receptor signaling pathways in kidney tissue evaluated by western blotting. Damage-fibrosis formations occurs and lipid deposition in kidney tissue evaluated by periodic acid schiff (PAS), masson trichrome and oil red staining under light microscopy. We observed that high cholesterol diet induced CD36, CD68 and SRI expressions. In this context, vitamin E supplementation in hypercholesterolemic rabbits showed its beneficial effect by decreasing PPAR while enhancing ABCA1 levels. Our light microscopy findings, glomerulosclerosis, interstitial fibrosis, tubular atrophy and degeneration were similarly observed in all groups, nevertheless renal tubular vacuolation increased in cholesterol and cholesterol+vit E groups compared to control group. Morover lipid accumulation were not observed in all groups.

Keywords: Hypercholesterolemia, Scavenger receptor, Kidney damage

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The effects of myo-inositol on biomass, phenolic compound composition and antioxidant activity in basil callus exposed to drought stress

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Abstract

Myo-inositol (inositol) has several functions in plant metabolism such as cell wall biosynthesis, transport and storage of auxins and stress metabolism. In this study, the effects of inositol on some biochemical and physiological parameters in basil callus culture exposed to drought stress. For this purpose, antioxidant capacity, total phenolic content, flavonoid content, average callus weight and individual phenolic composition (12 phenolic compound) of callus obtained from in basil (*Occimum basilicum* L.) plants were investigated. Explants were cultured in a medium prepared by adding 2 mg/L naphthaleneacetic acid (NAA), 0,1 mg/L 6-benzylaminopurine (BAP), 30 g/L sucrose and 2 g/L phytagel to the basal MS medium (Murashige and Skoog) for callus inductions. The calluses were applied drought stress, inositol and drought stress together with inositol. PEG (polyethyleneglycol6000) was used to generate drought stress in callus cultures. The distribution of phenolic compounds containing 4-hydrobenzoic acid, salicylic acid, vanilic acid, ferulik acid, rosmarinic acid, chicoric acid, caffeic acid, caftaric acid, epicatechin, rutin, quercetin, and kaempferol were determined by HPLC-DAD. The antioxidant activities of the callus were measured by ABTS (2,2-azino-bis(3-ethylbenzthiazoline-6-sulfonic acid) radical scavenging, DPPH (1,1-diphenyl-2-picryl-hydrazyl) free radical scavenging and FRAP (ferric reducing antioxidant power) methods. Drought stress caused a significant reduction in callus weight and inositol increased the average weight of callus. Drought stress significantly increased the total phenolic compound content and antioxidant activity. Inositol reduced the total phenolic compound content and antioxidant activity of the callus. According to HPLC results, individual phenolic compound contents were differently affected by inositol and drought stress. The amount of some phenolic compounds increased, but the amount of some compounds decreased significantly.

Keywords: Antioxidant activity, Myo-inositol, Occimum basilicum, PEG, Phenolic compound

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Pomegranate seed oil: Uses, remarkable benefits and chemical properties

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Abstract

The pomegranate (*Punica granatum* L.) is generally grown in tropical and subtropical regions such as Near and Far Eastern countries and the Mediterranean area, including Turkey. Pomegranate seeds are a waste product obtained after the processing of beverages, juices and sauces. Pomegranate seeds contain vary valuable oil about 6.3-12.2 % on dry matter basis. The oil contained in pomegranate seeds (PSO) consists of 65–85 % conjugated linolenic acids (CLnAs), the most important of which is 9-cis, 11-trans, 13-cis, octadecatrienoic acid, so-called punicic acid. A range of nutraceutical components such as tocols, sterols, fat-soluble vitamins as well as conjugated unsaturated fatty acids are rich in pomegranate seed oil. In this study; triglyceride, tocol composition and fatty acid, sterol profiles of pomegranate seed oil were evaluated by newly developed methods in high performance liquid chromatography (HPLC) and gas chromatography (GC), respectively, and were investigated the chemical and nutritional properties of cold pressed pomegranate seed oil. Different compositions of the mobile phase and flow rates for the HPLC system were used to obtain better separation for accurate quantitative analysis. The dominant triglyceride was found to be PuPuPu and γ-tocopherol was predominant tocopherol in pomegranate seed oil. For fatty acid composition analysis, triglyceride fractions were derivatized into their respective methylesters which were injected into GC-MS to identify and GC-FID to quantify the conjugated fatty acids of each fraction of triglycerides. Punicic acid was found to be dominant followed by catalpic acid and β-eleotearic acid, while β -sitosterol was the most abundant phytosterol form.

Keywords: Pomegranate seed oil, triglyceride, tocopherol, fatty acid and sterol

7th International
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Application of SSR and SRAP markers for genetic diversity of some *Origanum* species from Turkey

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Abstract

Origanum L. is one of the high value medicinal and aromatic plant species used for various purposes in the field of food, pharmaceutics, cosmetics and health. Within the study, 21 different specimens belonging to 18 species of the genus Origanum which comparises 10 endemic species in this study (*Origanum amanum* Post, *O. bilgeri* P.H. Davis, *O. boissieri* Ietswaart, *O. brevidens* (Born.) Dinsm., *O. haussknechtii Boiss.*, , *O. husnucan-baseri* H.Duman, Z.Aytaç & A.Duran, *O. minutiflorum* O.Schwarz & P.H.Davis, *O. saccatum* P.H. Davis, *O. solymicum* P.H Davis, *Origanum vogelii* Greuter & Burdet) naturally grown in our country were used in this study. SSR (Simple Sequence Repeat) and SRAP (Sequence Related Amplified Polymorphism) markers were used to assess molecular genetic diversity among Origanum genotypes. The genetic relationship of 21 *Origanum* genotypes was analysed by SRAP and SSR markers yielding 91 polymorphic alleles among the tested lines. The DARwin (http://darwin.cirad.fr/product.php) computer program was used to determine a Dice coefficient dissimilarity matrix for clustering analysis. The average polymorphism information content (PIC) of marker loci was 0.3. Clustering analysis with NJ (Neighbor Joining) showed that minimum and maximum dissimilarity values are 0.127 and 0.882, respectively. According to the obtained data, *Origanum onites* L. is more closed to *Origanum vulgare* L. subsp. *hirtum* (Link.) A.Terrac. among the 18 species.

Keywords: *Origanum*, Genetic diversity, SSR, SRAP

7th International
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Congress
25-27 April 2018, www.molbiotech.gen.tr

The relation between bioavailability and physicochemical properties in coconut oil and curcumin combinations

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Abstract

Coconut oil contains saturated and medium chain fatty acids (MCFA) by 92% and the other fatty acids like lauric, palmitic, capric, linoleic, stearic and oleic acids which make the coconut oil an important ingredient for health respect. As the MCFAs can metabolize fastly in intestines and they do not take parts in cholesterol biosynthesis and transfer directly to the liver, therefore it is claimed that coconut has HDL lowering effects in the body. Coconut oil also is known for its cardioprotective, antidote, antioxidant, antidiabetic, antimicrobial, antiaging effects and it is very popular with different usage alternatives in cosmetics and drug formulations. Curcumin is an active substance which can be found in the turmeric plant's root which is used in traditional medicine applications for many years for treatments of inflammations, metabolic syndromes, anxiety, hyperlipidemia, rheumatoid arthritis with its antimutagenic, antimicrobial and antioxidant effects. But when the curcumin is used as monopreparate because of the lipophilic structure its absorption is very limited and the bioavailability rate is very low. As a result of the fast metabolization and elimination, the expected health effects cannot be achieved. The studies showed that if the curcumin is combined with the oils which are rich in unsaturated fatty acids like coconut and flaxseed, the bioavailability rate increased considerably. In this study, we started to evaluate the stability (at 25 C %60 RH), shelf life, in vivo-in vitro correlation and physicochemical properties of curcumin and coconut oil combination if there is any relation with chemical stability and bioavailability.

Keywords: Curcumin, coconut oil, bioavailability, chemical stability

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Congress
25-27 April 2018, www.molbiotech.gen.tr

Proteome analysis of sunflower leaf responses to drought stress and recovery

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Abstract

With its global significance, sunflower (*Helianthus annuus* L.) have an important position in oilseed production and is in danger of drought in recent years. To investigate the role of proteins in the drought stress response, 40-day-old sunflower plants were subjected to drought for 9 days followed by 5 days of re-watering. In this study, it was used proteomic approach to study the responses of three genotypes which are showing different levels of tolerance to water deficit (tolerant, sensitive and wild type) before flowering stage. 720 30 spots were identified in each of three genotypes in MALDI-TOF/TOF MS/MS analyses. The analysis showed that 63 differentially expressed proteins identified in water-stressed and recovery plants when contrasted to well watered. More than half of the significantly changed proteins belong to primary metabolism as photosynthesis and carbohydrate metabolisms; besides other proteins function in energy and respiration, defense, arginine, nucleotide, fatty acid and glycolipid, protein and signal metabolisms and cell wall biogenesis. Different expression of proteins in metabolisms and the reductions in nucleotide and protein metabolisms, as well as protein involved in signal transduction of sensitive cultivar, made Tunca less successful in stress resistance compared to other genotypes. Tolerant genotypes were found to exhibit better performance in terms of photosynthesis and carbon metabolism, as well as protein expression in energy and respiration and fatty acid and glycolipid metabolisms in the same way, and increased the expression of 14-3-3 like protein in signal transduction pathway may increased resistance to drought conditions.

Keywords: Cultivated and wild sunflower, drought, recovery, proteomics, tolerance

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Effect of different parameters on facile synthesis of chitosan/poly (n-isopropylacrylamide) microspheres

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Abstract

Microspheres have currently attracted great interest for a wide variety of potential applications such as controlled drug delivery systems in regenerative engineering. Poly (N-isopropylacrylamide) (PNIPAM) is one of the most widely investigated synthetic polymer due to its thermo responsive properties. Chitosan has a variety of applications in biomedical area owing to its good stability, low toxicity, excellent biocompatibility and biodegradability. The combination of PNIPAM and chitosan has been studied as a pH and thermo sensitive material. Herein, we reported a facile synthesize of Chitosan/PNIPAM composite microspheres via water in oil (w/o) emulsion polymerization under different conditions such as initiator [2,2′-Azobis (2-methylpropionitrile)-AIBN] and crosslinking agents (glutaraldehyde and N,N'-Methylenebisacrylamide-MBAm) concentration. Obtained microspheres were successfully synthesized and characterized. Optical Microscopy determined the surface morphology and diameter of the microspheres. Our results revealed that the concentration of initiator and crosslinking agents was affected the surface morphology of synthesized composites. Chitosan/PNIPAM multiresponsive microspheres can be utilized as a potential material for use in biomedical applications.

Keywords: Fabrication, Chitosan, Poly (N-isopropylacrylamide), Responsive polymer, Microsphere

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An experimental and theoretical epr study on molecule and radical structures of metronidazole

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Abstract

Metronidazole (MZ), a synthetic antibacterial and antiprotozoal agent of the nitroimidazole class, is used against protozoa such as Trichomonas vaginalis, amebiasis, and giardiasis. Metronidazole is extremely effective against anaerobic bacterial infections and is also used to treat Crohn's disease, antibiotic-associated diarrhea, and rosacea. Metronidazole is a prodrug. Unionized metronidazole is selective for anaerobic bacteria due to their ability to intracellularly reduce metronidazole to its active form. This reduced metronidazole then covalently binds to DNA, disrupt its helical structure, inhibiting bacterial nucleic acid synthesis and resulting in bacterial cell death. In this study, to obtain molecular structure, conformational analysis of MZ was performed by Spartan 08 program. Consequently, ten conformers have been obtained. Geometry optimization calculations were performed and stable conformer was detected. Also this stable conformer parameters and XRD parameters were compared. For this conformation, seventeen possible radicals were modelled by using density functional theory (DFT) computations with respect to molecular structure. And then Electron Paramagnetic Resonance (EPR) parameters were calculated for these modeled radicals using the DFT/B3LYP method TZVP basis set. EPR parameters which were obtained from gas phase experiment of MZ were taken from literature. Experimental g value is good agreement with model radicals..

Keywords: DFT; EPR; Molecular modelling, Radical models, Metronidazole

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Investigating the antioxidant and cytotoxicity characterıstics of a silver nanoparticle system (agnps) prepared with *curcuma longa*

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Abstract

Drugs used in cancer patients systematically and negatively affect the entire body. In order to minimize or eliminate these effect, scientists have been turning towards the anticancer properties of natural products and try to carry out treatment methods directly on the cancer cells or tissues by targeting. For this, the focus is mostly on nanoparticle systems. Usage of metal particles, especially silver and gold nanoparticle systems, have become prominent. However, chemical synthesis of these NPs creates toxic effects. In recent years, scientists have resorted to synthesis by natural products. This type of synthesis is known as "GREEN SYNTHESIS" in the literature.

This study prepared a silver nanoparticle system with turmeric and compared the antioxidant and anticancer activities of this system to synthetic antioxidants. The resulting particles were characterized by UV spectrophotometer, scanning electron microscopy (SEM), X-ray diffraction (XRD) and Fourier transform infrared spectroscopy (FTIR). Antioxidant activities were determined in vitro and comparatively with those of synthetic antioxidants using parameters of DPPH, metal chelating and total antioxidants. Cytotoxic effects on the PC-3 prostate cancer cell line were determined using XTT test.

The NPs system was created based on the results. The UV, SEM, FTIR, XRD results indicated this in the characterization trials. The bond releases of the plant extract are seen in the FTIR results of the NPs. An activity of close to 45% was observed in DPPH removal with a concentration of 1mg/ml, while 30% metal removal activity was seen in metal chelating. The cell vitality test results were analyzed in the range of 1000 µg/ml-62.5 µg/ml, and all concentrations showed activity. The highest activity was in the highest concentration and activity decreased along with reduced concentrations.

Keywords: Anticancer, Antioxidant, Curcuma longa, Silver nanoparticle, PC-3

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Pectin extraction from lemon peels for the production of chitosan/pectin cryogels

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Abstract

Pectin is a natural, biocompatible, biodegradable and water-soluble heteropolysaccharide that exists in the cell walls of many plants. In the cell walls they serve as one of the main agents cementing the cellulose. Pectin has potential to be used as a biomaterial in tissue engineering field. This study aims to extract pectin from lemon (Citrus limon) peels and its use in the production of chitosan/pectin cryogels for tissue engineering applications. Pectin was extracted using alcohol precipitation method from the albedo of lemon peels. The extracted pectin was then subjected to qualitative and quantitative analyses. Functional groups present in the pectin were investigated using Fourier Transform Infrared (FTIR) spectroscopy. Chitosan/Pectin scaffolds were produced by cryogelation method with different ratios of chitosan and pectin (100:0, 80:20, 60:40 and 40:60, w/w). Interactions between pectin and chitosan, and crosslinking of cryogels with glutaraldehyde were verified by using FTIR. The weight loss of the cryogels was demonstrated as a result of the in vitro degradation test during 21 days. The swelling ratio of cryogels was measured and the duration of the equilibrium was determined as approximately 60 min. The fabricated and characterized cryogels can have potential to be used in tissue engineering applications.

Keywords: Pectin, Extraction, Chitosan, Cryogel, Tissue engineering

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Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Antibacterial activity of recombinant azurin against *Escherichia coli*

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Abstract

Azurin is an anticancer bacteriocin secreted by *Pseudomonas aeruginosa*. Bacteriocins are antibacterial proteins produced by bacteria that kill or inhibit the growth of other bacteria. The antibacterial proteins are used to improve food safety and quality. In this study, the antibacterial activity of recombinant azurin against *Escherichia coli* was determined. After genomic DNA was isolated from *P. aeruginosa*, azurin gene was amplified using forward and reverse primer containing restriction enzyme recognition sites. Amplified gene was ligated with plasmid digested by same restriction enzyme. The resulting plasmid was transformed into food-grade *Lactococcus lactis*. Azurin gene was expressed by the induction of nisin after verified by DNA sequencing analysis. Extracellular production of azurin was determined by Western blot analysis. The well diffusion method was used to determine the antibacterial activity of recombinant azurin against *E. coli*. Lyophilized cell-free supernatants containing azurin were applied at three different concentrations to nutrient agar plates with *E. coli*. After the application, diameter of inhibition zone was observed as 23 mm and 30 mm for 5 mg/ml and 10 mg/ml concentrations respectively. It was also determined that the diameter of zone increased as the concentration increased. Antibacterial azurin produced by food-grade *L. lactis* can be used as protective food additive agaist *E. coli.*

Keywords: *E. coli*, Expression, *P. aeruginosa*, Recombinant product

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Determination of the changes on the small intestine of the pregnant mice by histological, enzymehistochemical and immunohistochemical methods

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Abstract

We aimed to determine the changes on the small intestine in mice during different periods of pregnancy using histological, enzyme histochemical, and immunohistochemical methods. Twenty four mice were divided into four groups as non-pregnant control, at the middle of the first, second, and the third week of the pregnancy. Tissue samples were taken from duodenum, jejunum and ileum regions of the small intestine. Sections were stained with Crossmon's triple staining. Alkaline phosphatase (ALP) was demonstrated with simultaneous azo-coupling method and PCNA protein was demonstrated with the strept-avidin-biotin-peroxidase complex (S-ABC) method. In the last week of pregnancy in duodenum, jejunum and ileum were decreased the villus height, villus width and the rate of villus height/crypt depth. The crypt depth was decreasing in jejunum and ileum while it was increasing in duodenum with pregnancy. The muscle width was also found to increase in each section of the small intestine in the later weeks. It was identified that the reaction intensity of relative ALP statistically significant increased in duodenum, jejunum and ileum in the later weeks of pregnancy compared to control group. In duodenum, jejunum and ileum PCNA positive cell number was found to increase in the first and second weeks of the pregnancy whereas it was determined to decrease in the third week compared to the control group. According to the data obtained in the pregnancy period, though there are some differences among the gestational periods, it was concluded that pregnancy affected villus parameters of small intestine, intensity of ALP and PCNA positive cell number.

Keywords: ALP, mice, PCNA, pregnancy, villus

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Development of lipidic nanocarrier for gene delivery

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Abstract

Gene therapy is generally defined as the transfer of genetic material to cells to treat a disease or at least to improve the clinical condition of a patient. The most commonly used vectors in gene therapy are viral (retroviruses, adeno-associated virus, adeno virus, etc) and non-viral cationic liposomes, polymers and solid lipid nanoparticles etc.) vectors. The aim of this study was to develop a gene carrier system based on Cationic Lipid Nanoparticles (cLNs) and to evaluate the physicochemical properties (zeta potential, particle size, DSC, pH), cytotoxicity, DNA binding properties, serum stability and also transfection to cells. For this purpose, cationic formulations were formulated using glycerol dibehenate (Compritol® ATO 888) and Geloil™SC as a lipidic phase with DOTAP as a cationic agent. These formulations were produced by using oil-in water emulsification technique. GFP was selected as the genetic material to be loaded into the formulations. GFP was adsorbed to formulations via electrostatic interactions. According to results, the cLNs prepared showed considerably small particle sizes (285 nm) and high zeta potential (+44mV). Based on the MTT assay the cytotoxic effect of formulation on the NIH 3T3 cell line showed dose dependant pattern. Prepared formulations was bind to DNA effectively and protect the DNA against to nuclease in serum. It was concluded that cLN formulations can be prepared as pDNA-cLNs complex can be used as gene delivery system. Further studies are going on in vivo experiments on animals.

Keywords: Lipidic Carrier, Nanoparticle, Gene Delivery, GFP

7th International
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Congress
25-27 April 2018, www.molbiotech.gen.tr

Determination of milk/plasma rate and, the milk and plasma pharmacokinetics of amoxicillin in dairy cattle

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Abstract

The present study was conducted to determine the passage ratio of amoxicillin into milk and, pharmacokinetics of amoxicillin in milk and plasma after intramuscular administration. In this study, a total of 5 healthy dairy cattle (Holstein, 450-500 kg, 2-4 years) were used. Animals received a single intramuscular amoxicillin trihydrate at a dose of 14 mg/kg bw. Blood samples were collected from the jugular vein into tubes with EDTA prior to drug administration (0) and at 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, and 12 h after administration. Milk samples were collected prior to antibiotic administration (0) and at 0.25, 0.5 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, and 12 h after administration. The plasma and milk concentrations of amoxicillin were determined using HPLC system with UV detector. The passage ratio of amoxicillin into milk was determined by both AUC-based calculation and using milk and plasma concentrations at sampling times .The milk/plasma ratio of amoxicillin was found 0.46-052. The terminal half-life and MRT parameters of amoxicillin in plasma and milk were determined 6.05 and 2.62 and 8.60 and 5.35 h, respectively. The Cmax levels of amoxicillin in plasma and milk were measured as 1096 and 457.25 ng/mL, respectively. In conclusion, it may be stated that amoxicillin exhibits similar pharmacokinetic behavior profile in plasma and milk.

Keywords: Amoxicillin, Milk, Plasma, Pharmacokinetic

7th International
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Congress
25-27 April 2018, www.molbiotech.gen.tr

Molecular and biochemical characterization of a novel pectate lyase from *Bacillus amyloliquefaciens* BS-6

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Abstract

Microbial Pectate lyase have been commonly used in a variety of industrial applications including waste papers, prebiotic production, and wine clarification, of which the largest bacterial enzyme share has been taken by the *Bacillus*. Although there has been a large amount of research on bacterial pectinases, the challenge is to make a unique enzyme that can work at a wide range of pH and temperature with a stable enzyme activity so that the cost of industrial process to maintain in a specific temperature and a pH for various enzymes used in a single process could be reduced. Here we report cloning of a pectate lyase gene from *Bacillus amyloliquefaciens* BS-6, and biochemical characterization of the recombinant pectate lyase. PEL BS-6 is identical with *B. subtilis* 168 pel enzyme with 100% amino acid and 71% nucleotide sequence homology. Although, they are genetically very close, they are distinctly different in physiology. The pel gene from BS-6 encodes a 421-aa protein with a molecular mass of 65.75 kDa. Specific enzyme activity increased from 12.8±0.3 to 49.6±0.4 units/mg after cloning. The relative enzyme activity percentage of the recPEL BS-6 ranged from 80 to 100 at pH between 4 and 14. It was quite stable at different temperature values ranging from 15 to 90°C. The recPEL BS-6 showed a maximal activity at pH 10 and at 60°C. 0.5mM of CaCl2 is the most effective metal ion on the recPEL BS-6 by increasing the activity with 473%. recPEL BS-6 was stable at -20 °C for 18 months. Additionally, recPEL BS-6 increased the clarity of the juices. This study introduces a novel bacterial pectate lyase enzyme with its ability been thermostable, thermotolerance, and active over wide range of pH meaning that can work at both acidic and alkaline environment, which is the most required properties in the industry.

Keywords: Pectate lyases, pH-thermotolerance, pH-thermostable, *Bacillus amyloliquefaciens*

7th International
Molecular Biology and Biotechnology
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25-27 April 2018, www.molbiotech.gen.tr

Effects of *Agaricus arvensis* mushroom extract on erythrocyte fargility and antioxidant parameters against CCl4-induced oxidative stress in rats

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Abstract

Carbon tetrachloride (CCl4) is a toxic chemical that causes generation of free radical species (ROS) in many tissues such as liver, kidney, testis, brain and blood. The present study was designed to establish the protective effect of *Agaricus arvensis* extract on CCl4-induced oxidative stress in rat. After the toxicity test, rats were divided into four experimental groups: Control, CCI4, CCI4+*A. arvensis*-100 mg/kg and CCI4+*A. arvensis*-500 mg/kg groups. At the end of experiment, the roles of the orally administrated *A. arvensis* extract against CCl4-induced oxidative stress were evaluated by measuring, erythrocyte fragility and antioxidant defence system enzymes such as reducte glutathione (GSH), glutathione peroxidase (GPx), superoxide dismutase (SOD) activities and malondialdehyde (MDA) content in erythrocyte cells of rats. According to the results, erythrocyte hemolysis were significantly increased whereas GPx enzyme activity decreased in erythrocyte of CCl4 treated rats. *A. arvensis* lyophilized extract (100 and 500 mg/kg) successfully debilitated these effects of CCl4. In conclusion, our study demonstrated an improved the erythrocyte fragility and protective effect of *A. arvensis* in CCl4 induced oxidative stress in rat. This protective effect of *A. arvensis* can be correlated to its direct antioxidant effect.

Keywords: *Agaricus arvensis*, Antioxidant, Malondialdehyde, Erythrocyte fragility, Rat

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Molecular structure and the spectroscopic calculation of the allyl alcohol molecule

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Abstract

As a consequence of detailed conformational search of the Allyl Alcohol, five different conformers of molecule have been obtained. Then geometry optimizations of all of the possible conformers was performed by Becke's three-parameter hybrid-exchange functional combined with the Lee-Yang-Parr correlation functional (B3LYP) of Density Functional Theory (DFT) and standard 6-311++G(d,p) basis sets in liquid phase. Conformational energy of most stable conformer is -121227.8552 kcal/mol. Using this conformer, 14 possible radicals were modeled for the same level of DFT. Later, Electron Paramagnetic Resonance (EPR) parameters were calculated for these modeled radicals using the DFT/B3LYP method and TZVP basis set and they were compared with the experimental counterparts. The calculated g value of model radical (Rad 3) is 2.00311 and calculated hyperfine constants of model radical (Rad 3) are H: 12.96 G, H: 3.35G, H: 13.01 G, H: 13.89 G and H: 2.94 G. The calculated and experimental values were good agreement in that study.

Keywords: Molecular Modelling, DFT; EPR; Conformational Analysis, Allyl Alcohol

7th International
Molecular Biology and Biotechnology
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25-27 April 2018, www.molbiotech.gen.tr

Synthesis, structral elucidation, ın vitro anticancer activity and molecular docking studies targeting RXRα with some novel retinoid analogues

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Abstract

Bexarotene which is currently used as a drug in cancer disease and has reached phase II / III stages in various cancer types.In this study, novel six bexarotene-like retinoid derivatives were synthesized, characterized and evaluated their in vitro anticancer activities. These retinoid analogues (6-11) were synthesized in six steps. Compounds were purified by column chromatography using suitable solvent system. The purity of the compounds was controlled by TLC followed by determining of the melting points. The chemical structures of the compounds were explained with their elemental analysis, mass and 1H-NMR spectral data.It is focused to analyze the activity of compounds against target, Retinoid X Receptor (RXRα), to identify binding properties of compounds to active site of enzyme and to evaluate relationships between biological activity and binding affinities of compounds by using Autodock 4.2. program. The sulforhodamine B (SRB) assay, a colorimetric assay based to measure of cellular protein amount was employed for evaluating cytotoxic activity of the compounds on human cancer cell lines (A549, lung cancer; HeLa, cervix cancer; MCF-7, breast cancer and WiDr, colon cancer cell lines). Cells were treated with the test compounds for 48 h. Absorbance of wells was measured at 490nm following cell fixation and SRB staining. IC50 values of compounds were calculated from cell growth (%)-data by S-probit analysis. Data in this study were presented as mean values obtained from three independent experiments.

According to the obtained cytotoxicity results, compounds 6, 8, 11 were found having the highest cytotoxic activity against four cancer cell lines. Among these three compounds, compound-11showed the highest anticancer activity. In addition to these results, it was revealed that compound-11showed slightly higher cytotoxicity against WiDr colon cancer cell line with the IC50 value of 2.38μM than CPT-a routine anticancer drug (IC50: 2.57 μM). Furthermore, when compared to the anticancer activity with molecular docking results, it was found that the better RXRα binding affinities of the compounds, the higher the anticancer activity of them. Considering the results in this study, the highest active compounds can be used as the lead-compounds targeting RXRα for anticancer compounds for further studies.

Keywords: Bexarotene, Retinoid derivatives, SRB, Molecular docking, RXRα

7th International
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25-27 April 2018, www.molbiotech.gen.tr

miR-378 target gene TGFB2 in the Stage II colon cancer tissue

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Abstract

Colorectal cancer (CRC) is the third leading cause of cancer-related deaths in worldwide. The sensitivity of markers used in the early and painless diagnosis of various cancers, including CRC, remains low. Recent studies have shown that non-protein coding small RNA molecules, called microRNAs (miRNA), play a key role in the mechanism of both the development of cancer and its treatment. miRNAs have tumor suppressing and oncogenic effects in the development of certain types of cancer, including CRC. The aim of the present study was to determine the profiles of oncogenic and tumor suppressing miRNAs affecting cancer development and miRNA target gene in the tumor tissue as well as in normal tissues of patients with Stage II CRC. Arrays analysis belong to this study have demonstrated been for the first time in colorectal tumor tissues, 6 of the 8 miRNAs of the miR-378 family (hsa-miR-378i, hsa-miR-378c, hsa-miR-378d, hsa-miR-378e, hsa-miR-378f and hsa-miR-378g) showed that targets TGFB2. It is possible to suggest that early diagnosis of mir-378 colon cancer may have an important role in early diagnosis .

Keywords: microRNA, Stage II colorectal cancer, TGFB2

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7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Molecular screening of *potato virus y* resistance and genetic characterization of local potato cultivars from niğde province

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Abstract

Potato (*Solanum tuberosum* L.) is the world's fourth largest food crop plant with its nutritionally valuable food and has been dispersed from their origin to many countries around the world. *Potato virus* Y (PVY) is a plant pathogenic virus of the family *Potyviridae*, and one of the most serious virus diseases of potatoes worldwide. Niğde province has an importance for potato cultivation and ranked first in potato production in Turkey. The aim of the study was screening of local potato cultivars for PVY resistance and genetic characterization of them via microsatellites. The most common potato cultivars of the region (Agria, Granola, Hermes, Sante, Agata, Alegria, Aurea, Banba, Belmonda, Borwina, Brooke, Concordia, Estrella, Infinity, Jelly, Medeleine, Marfona, Melody, Orchestra, Provento, Soraya, VR-808, Pomqueen, Natascha, Bettina, Nectar and Marabel) were collected and germinated at greenhouses. Nucleic acids (DNA) were extracted from the young leaves based on CTAB method. For determination of virus resistance, STM0003 marker system for potato Rysto resistance gene was used and PCR analyses were performed. Based on the results, Madeleine, Melody, Orchestra, Provento, Belmonda and Estrella are the promising cultivars for PVY resistance. For the genetic diversity analysis, highly polymorphic 8 SSR markers were used and the results indicate that the local cultivars have narrow genetic base. These findings can help growers and breeders to improve PVY resistant potato cultivars.

Keywords: Potato, PVY, Rysto, Microsatellite, Cluster analysis

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

The apoptotic activity of juglone and juglone-ascorbate combination in pancreatic cancer cells

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Abstract

Discovery of new therapeutic agents is mandatory to overcome the challenge of Pancreas Cancer treatment, due to its aggressivity, insufficiency of current therapeutics and acquired resistance to conventional chemotherapy and radiotherapy Juglone, as a naphthoquinone, is a secondary metabolite produced naturally in walnuts type trees having allelopathic feature in its native environment. It was shown that juglone prevents cell proliferation and induce ROS-mediated mitochondrial apoptosis. Ascorbate with both antioxidant and oxidant feature shows selectively cytotoxicity in cancer cells. In this study, according to our hypothese that cytotoxic and apoptotic effects of juglone could be increased by ascorbate, we aimed to evaluate the expression levels of proapoptotic Bax gene, mitochondrial apoptotic pathway related antiapoptotic bcl-2 gene and an important apoptosis inhibitor gene Birc5 (Survivin). Expression levels of Bax, Bcl-2 and Birc5 genes were determined by qPCR following treatment of PANC-1 cells with different Juglone and juglone-ascorbate combination doses during 24 hours. For Bax gene, a statistically significant 2,9 -1,78 ve 2,89-fold increase were determined after 20μM juglone and 10μM and 20μM juglone with 1mM ascorbate treatments, respectively ($p < 0,05$). In the BIRC5 gene expression, there was a significant decrease as 2.93 fold after 20 μM juglone-ascorbate administration. Also, we observed a statistically significant decrease in the expression of Bcl-2 gene as 1.4 and 1.69 folds after 15 μM and 20 μM juglone and 2.93 folds decrease after 20 μM juglone-ascorbate aplications ($p < 0,05$). Taken together, our results suggest that juglon is a promising anticancer agent that has a stronger activitiy in combination with ascorbate.

Keywords: PANC-1 cells, juglone, ascorbate, apoptotic activity

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Expression profile of alpha-glucosidase in sunn pest, *Eurygaster maura*

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Abstract

The sunn pest, *Eurygaster maura*, is a sap-sucking pest of cereal crops in Turkey and neighboring countries. This univoltine species has two biological forms, including active (feeding) stage and passive (non-feeding) stage, a combination of overwintering and aestivating periods. Prior to diapause, adults accumulate lipid reserves in order to meet their energy demand during hibernation in cold winter. Later these adults migrate to grain fields during spring where they feed, mate, lay eggs and die. New generation adults, the major destructive individuals on grains, keep feeding to store fat body for winter until migration. Finally, hibernation period starts after grain harvest and the insect completes its life cycle. Carbohydrates have significant importance with being the large source of stored fat in insect body. Alpha-glucosidase also known as maltase and beta-glucosidases hydrolyze the glycosidic bonds between sugar residues. In this study, Real time PCR analyses were conducted to examine alpha-glucosidase expression in pre-migrated, migrated, feeding, aestivated, pre-hibernating and hibernating stages of sunn pest adults. Results showed that expression of alpha-glucosidase transcript was higher in the feeding stage, while no transcript was found in the migrated stage. These results suggest that this enzyme is likely used during feeding stage to consume carbohydrates and store energy when preparing for hibernation.

Keywords: Alpha-glucosidase, carbohydrate metabolism, diapause, *Eurygaster maura*

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

In vitro cytotoxic activity of a novel oral anticoagulant on hela cells

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Abstract

New oral anticoagulants including dabigatran are commonly used in the prophylaxis and treatment of systemic embolism and deep vein thrombosis worldwide. Cancer patients have also an increased risk of developing venous thromboembolic events or may have other indications for anticoagulation, such as atrial fibrillation. However, several data suggest that anticoagulant drugs may have an effect on tumor development and progression. In this study, we aimed to investigate the cytotoxic effects of dabigatran on a cancer cell line HeLa cells derived from human cervical cancer. Cells were placed in 96-well culture at an initial density of 50.000 cells/ml in six replicates and incubated in the Dulbecco's Modified Eagle's Medium (DMEM)/Ham's F12 supplemented with 10% fetal bovine serum (FBS). Following incubation, the cells were treated with six dilutions of the test material [Pradaxa (dabigatran etexilate, 150 mg)™]. Test material was prepared in culture medium supplemented with 1% dimethyl sulfoxide. Stock solution were prepared as 0.30 g/20 ml for the initial dose. Stock solution underwent serial dilution and were prepared in five dilutions and only DMEM/F12 medium was served as control groups. The cell viability was determined by MTT assay. At 24-hour incubation, the cells exposed to all dilutions of dabigatran showed a significant difference compared to normal fibroblastic morphology. The cells displayed cellular alterations including nuclear condensation, rounded morphology, and cell degeneration. The viability of HeLa cells was examined at 24 and 48 post-incubation hours. At 24 and 48 hours, dabigatran showed a cytotoxic effect in all dilutions. The results showed that dabigatran may reduce proliferation of cancer cells.

Keywords: Dabigatran, Cell Culture, Cytotoxicity, HeLa cells

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Screening and production of protease enzyme from *Bacillus sp.* strains and ıts dehairing application

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Abstract

The leather processing industry contributes significantly to the country's economic development. In this industry, in the conventional dehairing process, chemical pollutants such as lime, sodium sulphide, chrome salts and formaldehyde are used. Presence of these chemicals in tannery waste is responsible for tremendous pollution, causing health hazards to the tannery workers. Chrome salts and formaldehyde contain a special substance that causes bronchial asthma in tannery workers. Chrome intoxication, liver and renal disorders are seen. In recent years, microbial enzymes are used as an alternative technology to the conventional methods. Enzymatic dehairing process was accomplished by proteolytic enzymes of great commercial importance. Proteases have dehairing properties, and used in the leather processing industry. Due to the increasing demand of enzyme in the leather industry, there arises a need for new proteases. Therefore in this study, we have performed screening of protease producing *Bacillus sp.* in Turkish soils. The morphology and biochemical tests were studied for *Bacillus* genus. Most efficient a isolate selected. The nucleotide sequence of the 16S rRNA gene of this new isolate E10-2 was 100% similar to *Bacillus cereus* SL1. The protease produced by *Bacillus* was studied for its dehairing application against beef and goat skins pieces for different soaking periods (12, 24, 36 h). The removal of hairs was found efficient and the quality of dehaired skin was satisfactory after 12 h of treatment. It reported that protease produced by new isolate E10-2 had a potential for dehairing, and might be used as dehairing agent in tanning industries.

Keywords: *Bacillus*, Protease, Beef and Goat Skin, Dehairing

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Discovery of the first microRNA-like species in human pathogenic fungus *Aspergillus fumigatus*

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Abstract

Aspergillus fumigatus is not grouped in the most prevalent fungi in the world however it is one of the most common one in the atmosphere. *A. fumigatus*, an opportunistic pathogen causing fungal lung diseases, is recently shown to harbour several different types of mycoviruses. The miRNAs were first discovered in *Caenorhabditis elegans* and have been identified in animals and plants, however no miRNAs have been reported in fungi apart from microRNA-like (milRNAs) have been identified in *Neurospora crassa, Sclerotinia sclerotiorum, Metarhizium anisopliae* and *Trichoderma reesei* to date. However none of those fungi were infected with viruses. Therefore, here we investigated the existence of A. fumigatus microRNAs in the presence and absence of three mycoviruses: *Aspergillus fumigatus* partitivirus-1 (AfuPV-1, PV), *Aspergillus fumigatus* chrysovirus (AfuCV, CV) and *Aspergillus fumigatus* tetramycovirus-1 (AfuTmV-1, NK). Small RNA-sequencing (sRNA-seq) libraries of virus-free and virus-infected isolates were created using adapters and sequenced using Illumina HiSeq2500. The data was analysed in order to identify miRNA-like reads differentially expressed between virus-free and virus-infected samples using bioinformatic methods. MicroRNA identification was performed using three different approaches; (i) similarity search using miRBase database and known fungal miRNA-likes, (ii) miRNA prediction using the miRCat program, (iii) searching the differentially expressed sRNAs for annotation and folding the flanking regions. Five predicted miRNA-like candidates were checked by northern blotting and out of five candidates, three of them namely Folded-1, Folded-2 and miRCat-2 were detected in related isolates. To our knowledge, this is the first study reporting miRNA-like species in *A. fumigatus*.

Keywords: *A. fumigatus*, microRNAs, miRNA-likes, fungi, mycovirus, sRNA-seq, bioinformatics

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Why molecular modelling and spectral analysis are important for drug design? Key applications: novel thiosemicabazone complexes

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Abstract

Molecular modelling and spectroscopic methods are the rapidly advancing and exciting fields in the physics, chemistry and biology today. Electronic structure methods used for molecular modeling calculations are successful methods for calculating and predicting molecular parameters and molecular properties using the laws of quantum physics principles and some mathematical approaches. By means of spectroscopic techniques, information about the structure, electronic and magnetic properties of the compounds forming the substance can be obtained. The biological activity of metal complexes, popular compounds used for the next generation of drug design, depends intimately not only on the metal and its oxidation state, but also on the type and number of coordinated ligands, and the coordination geometry. This provides a rich platform in pharmacological space for structural and electronic diversity. In the case of the determination of a metal complex structure with spectroscopic (EPR, UV, IR, NMR etc.) and molecular modelling calculations (HF, Post HF and DFT etc.), the anticancer and antimicrobial activity potential of the complex can be revealed out because the electronic structure parameters provides information about the chemical activation properties. In this study, the importance of combined spectroscopic techniques and molecular modelling analysis for drug delivery will be emphasized by the examples of novel thiosemicarbazone metal complexes that have various biological activities such as antifungal, antibacterial, anticancer activities.

Keywords: Metal complexes of thiosemicarbazones, molecular modelling, spectroscopic techniques

7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Quantitative determination of sunset yellow, allura red, fast green, erythrosin-B and quinoline yellow using Fe3O4 modified with *Elaeagnus angustifolia* based on solid phase extraction and HPLC

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Abstract

In this study, we developed and optimized a method for simultaneous quantitative determination of sunset yellow (SY), allura red (AR), fast green (FG), erythrosine-B (Ery) and quinolone yellow (QY), which have toxicological important. The Fe3O4 modified with *Elaeagnus angustifolia* as an adsorbent was used for solid phase extraction (SPE). Optimum conditions such as pH, adsorption and elution time, volume elution were optimized for SPE. Optimum quantitative analysis conditions for adsorption time, elution time, eluent type and elution volume were observed for 5 minutes, 3 minutes, methanol / 1 M NH3 (6/4, v / v) and 2 mL respectively. These optimum values were obtained 25 mL of 10 mM HCl which includes 10 mg/L dye solutions. After SPE extraction, dyes were quantitatively determined by HPLC coupled by an ultraviolet detector. The separation was performed gradiently on a Zorbax C18 reverse phase analytical column (4.6 x 250 mm, 5 µm) with 20 mM ammonium acetate buffer/acetonitrile/methanol as a mobile phase mixture, at 30°C. Mobile phase rate was 1 mL/min. Detection wavelengths were set to 500, 600, 530, 410 nm for SY and AR, FG, Ery and QY, respectively. The retention times were 7.1, 9.2, 10.0, 12.4, 14.8, 22.1 min for SY, AR, FG, Ery and QY, respectively.

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7th International
Molecular Biology and Biotechnology
Congress
25-27 April 2018, www.molbiotech.gen.tr

Investigation of selected exons from RPE65 gene in syndromic Retinitis pigmentosa

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Abstract

Retinitis pigmentosa (RP), also known as night blindness which cause loss of vision, has syndromic forms accompanied by systemic anomalies in many organs. One of the most common is Bardet-Biedl syndrome. Retinal pigment epithelium-specific protein 65 kDa (RPE65) is expressed in the retinal pigment epithelium. Mutations in RPE65 cause autosomal recessive RP. In our study, we aimed to analyze 50 patients who had autosomal recessive inheritance in RP by sequence analysis. Two individuals had syndromic RP (Bardet Biedl syndrome). 4-5-10-11-13 exons which are 14 exons of RPE65 gene were selected for the mutation screening. DNAs which were isolated with DNA isolation kit from blood samples, were amplified with PCR. PCR products were sequenced with ABI Prism 310 Genetic Analyzer instrument and were analyzed with Sequencing Analysis, SeqScape and BioEdit softwares by using the mutation tables. As a result of the sequence screening in individuals who had syndromic RP, G>A E352E polymorphism in Exon 10 of RPE65 gene were detected.

Keywords: Retinitis pigmentosa, Bardet Biedl syndrome, RPE65, Sequencing Analysis

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25-27 April 2018, www.molbiotech.gen.tr

Fabrication of hydrophilic thin films with enhanced electroconductive and mechanical properties by the reinforcement of RGO

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Abstract

In this study, novel hydrophilic films with enhanced mechanical and conductive properties were synthesized by the incorporation of reduced graphene oxide (RGO) at different concentrations into the polymeric network forming hyaluronic acid, gelatin and poly ethylene oxide (HyA/Gel/PEO) by solvent-casting method. The fabricated films were characterized by FT-IR analysis. Mechanical performance was analyzed by universal mechanical tester. The results verified that mechanical properties of RGO-reinforced HyA/Gel/PEO films enhanced significantly with respect to that of unreinforced HyA/Gel/PEO films. To determine biocompatibility of the films, L929 (Murine Fibroblasts) cell lines were used. Water-uptake capacities were measured by swelling tests. 4-prob method was used to measure conductivity features. According to the conductivity results, HyA/Gel/PEO film bearing 20 v.% RGO has the highest electrical conductivity value (1.832x10-6 S/cm). All of the results demonstrated that the obtained electroconductive, durable, biocompatible and hydrophilic films can be used for many applications especially controlled drug release systems and tissue engineering in the future.

Keywords: Electroconductive hydrophilic film, reduced graphene oxide, solvent-casting method

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Production and characterization of biomimetic hydroxyapatite coated polyvinyl alcohol/chitosan composites

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Abstract

Nowadays, hundreds of people of all ages are treated due to accidents or various disabilities resulting from organ or tissue loss. Tissue engineering is a science that aims to increase and restore tissue and organ functions in order to improve quality of life for all these treatments. Cryogels are open cell structured matrices that are produced from frozen solutions of monomeric or polymeric initiators and they are typically composed of interconnected macropores. Hydroxyapatite, which forms the inorganic structure of bone tissue, is a calcium phosphate based bioceramic material used for the construction of various prostheses as artificial bone due to its biocompatibility, for the repair of defected bones, and for the coating of metallic biomaterials. Biomimetic hydroxyapatite coating is a unique method carried out in biomimetic conditions at 37ºC, which is the human body temperature, and 7.4, which is the human body pH value, by using a "Synthetic Body Fluid (SBF)" which has almost equal ion concentration of human blood plasma. In this study, PVA / Chitosan composite cryogels coated with biomimetic hydroxyapatite were produced to be used for the renewal of bone tissue, and characterization was achieved. In the results, it was observed that as the chitosan ratio and the coating duration increased, the coating efficiency increased. The effect of PVA/Chitosan ratio and coating duration on the final properties of the coated cryogels were analysed. For characterization studies, swelling test, porosity analysis, weight increase after coating, in vitro degradation, FT-IR, SEM, EDXS analysis were performed.

Keywords: Cryogel, Polyvinyl Alcohol (PVA), Chitosan, Hydroxyapatite (HA), Synthetic Body Fluid (SBF)

Acknowledgement: This work was supported by the Scientific Research Projects Unit of Mersin University, Project No: 2018-1-TP2-2733

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Determinations of protein secondary structural alterations in the aging of bloodstain on cotton fabric using fourier transform infrared spectroscopy study

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Abstract

Accurate assesment of bloodstains age has great importance for forensic investigator in the determination of time range of crime. The utility of various methods such as HPLC, EPR in the identification of this age were indicated. However none of them are not fast and reliable. The ability of Fourier Transform Infrared (FTIR) Spectroscopy in the estimation of bloodstain age was indicated with recent studies. In these studies the effects of various temperatures on this determination has not been clarified yet. Therefore, the current study was established to elucidate these effects. IR spectra of bloodstain on cotton samples were collected at different temperatures (10°C, 20°C, 30°C and 40°C) and times (1, 24, 48, 72 hours and 5,10 days). Quantitative spectral analysis including the determination of wavenumber of amide I band, area ratio of amide I/amideII bands and secondary structures of proteins were performed the protein region (1740-1475 cm^{-1}). These analyses results implied that there is a structural alteration in the proteins of bloodstain in time and temparature dependent manner especially in the protein secondary structures (α-helix, aggregated β-sheet and random coil). Based on these alterations, Lineer discriminat analysis (LDA) was performed to test the success of FTIR in the estimating bloodstain age. 100 % classification succes was obtained in all cases. These findings proved that this technique together with chemometric analysis can be succesfully implemented in the quick and accurate identification of bloodstain age.

Keywords: ATR-FTIR, bloodsatin age, protein secondary structures, time, temperature

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25-27 April 2018, www.molbiotech.gen.tr

Characterization of a novel esterase obtained from hypersaline lake (acıgöl) by metagenomics approach

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Abstract

Metagenomics is a promising field that enables scientists to discover and identify novel biomolecules from microorganisms, regardless of their cultivability. Here a novel esterase, Est_Ag, was successfully isolated from Acıgol (Denizli) by metagenomics approach. First, environmental DNA from Acıgol sediment was obtained. Then, by targeting the conserved regions among lipolytic enzymes, degenerate PCR and genome walking strategies were applied. The full gene sequence of the protein was analyzed using bioinformatics tools and the putative protein sequence showed a maximum identity of 91% with a previously known esterase. After confirmation of the novelty of the gene sequence, the gene was cloned into expression vector and the protein was over-expressed in *E. coli.* Histidine tagged protein was successfully purified and detailed biochemical characterization was carried out. The enzyme showed optimum activity at pH 9 and 30°C. In substrate specificity experiments, it was shown that the enzyme prefers short acyl chain length para-nitrophenyl esters as substrate. The effect of NaCl, organic solvents, metal ions, detergents and inhibitors on the activity and the stability of the enzyme were also investigated. The outstanding features such as activity at cold temperatures, stability in the presence of organic solvents and metal ions make this novel esterase a potential candidate for industrial applications.

Keywords: Genome walking, metagenomics, esterase

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Determınatıon of carbamazepıne ın human plasma by hplc ultravıolet method: applıcatıon to a therapeutıc drug monıtorıng study

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Abstract

In this study, a high-performance liquid chromatography method was developed and validated for quantitative analysis of CBZ in plasma. The validity of the method was monitored in real plasma samples of 30 patients under epilepsy treatment using CBZ. The chromatographic separation was carried out with a reverse-phase C18 analytical column (3.9 x 150 mm, 5 µm particle size), at 30 °C. 20 mM KH2PO4 (1% triethylamine), acetonitrile, methanol (6:3:1, v/v) was used as a mobile phase. It applied to the column isocratically at 1 mL/ min flow. Ultraviolet detector was set at 220 nm. Chlorpromazine trihydrate was used as an internal standard. The samples were loaded into the HPLC using a manual injector which have a 20 µL loop volume. Accuracy and precision were found between (-9.71) - 1.65 (RE%) and 4.15 - 1.33 (RSD%), respectively for intraday and interday reproducibility study. The detection and quantification limits were 40.1 and 121.7 ng/mL, respectively. Plasma recovery tests were carried out at concentrations of 1, 5 and 20 ppm and the results obtained ranged from 82.42% to 105.68%. Carbamazepine levels were found in the range of 0.15 to 11.38 ppm (6.15 ppm ± 2.38 (mean ± SD)) in blood samples taken from patients who were under epilepsy treatment with carbamazepine between 200 and 1200 mg/day. The method developed, validated and successfully applied to patient samples is a simple, rapid, reliable method that can be used in both therapeutic drug monitoring study and overdose toxicological analysis of patients using CBZ.

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25-27 April 2018, www.molbiotech.gen.tr

Synthesis of novel chiral tetraoxocalix[2]arene[2]triazine derivatives for enantioselective aldol reaction of aldehydes with acetone

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Abstract

Organocatalysis is a reaction carried out by sub-stoichiometric amounts of organic compounds which do not contain even a small amount of enzyme or inorganic element. Although metal catalyzed reactions have wider substrate scope, they are associated with a few drawbacks such as high cost involved in the preparation of catalysts and toxicity of metals which can be carried over to products. Organic compounds, as compared to metals, are more stable, less expensive, non-toxic, readily available, and environmentally friendly. Besides, organocatalytic reactions are less sensitive to the presence of water or air in comparison to metal catalyzed reactions. Thus, the reproducibility and operational simplicity of these reactions are enhanced. Organocatalysts provide better alternatives not only to metal catalysts but also to biocatalysts. They provide broad substrate scope in contrast to enzymes which are highly substrate specific and cannot tolerate even a minor change in the structure of the reactants. In addition, organocatalysts display another advantage over both metal catalysts and enzymes in that they are easily amenable to solid support, leading to easy recovery of the catalyst and simplification of the reaction work up.

Chiral tetraoxocalix[2]arene[2]triazine functionalized at the lower rim with chiral naphtylethylamine units have been prepared. The structures of these receptors were characterized by a combination of 1H NMR, 13C NMR, FTIR and elemental analysis. These compounds were evaluated as organocatalysts for asymmetric aldol reactions between various aldehydes and acetone. Very good yields and enantioselectivities were achieved in optimal conditions.

Keywords: Enantioselectivity, HPLC, Organocatalyst, Tetraoxocalix[2]arene[2]triazine

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25-27 April 2018, www.molbiotech.gen.tr

Synthesis of a new polyoxometalate copper complex and its use in determination of dopamine by uv spectroscopy

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Abstract

Polyoxometalates are a class of discrete transition metaloxide clusters that are constructed on the condensation of metaloxide polyhedral by a king of self-assembly proces [1]. POMs are attractive in terms of their applications, including in catalysis [2], medicine, biotechnology [3,4], and electrochemistry[5]. POMs also can be activated by UV or visible light, respectively[6]. Dopamine is one of the important catecholamine neurotransmitter distributed in the central nervous system [7]. It also plays key roles in the function of the renal, hormonal, and cardiovascular systems [8]. As a result, dopamine has been given tremendous consideration by neuroscientists and chemists in bio-medical and bio-analytical research and there is a strong need to establish highly sensitive and selective methods for the direct detection of dopamine [9]. Herein, the crystal structure Na[(Cu(bipy)2)2(BMo12O40)] has been hydrothermally synthesized and characterized by single crystal X-ray diffraction, Fourier-transform infrared spectrum (FT-IR), powder X-ray diffraction (XRD). Product was used in dopamine determination by UV spectroscopy. Our results have demonstrated that the compound has an effective feature for determination of dopamine by UV spectroscopy with low costs and easy, fast laboratory performance.

Keywords: Polyoxometalates, Dopamine Detection, UV spectroscopy

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Congress
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A new appraoach for malondialdehyde determination: gold-modified electrode

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Abstract

Living organisms have to use molecular oxygen to maintain metabolic functions and during metabolic events, during the metabolism free radicals form. However, free radicals are harmful and whereas the metabolic system do not maintain its function related with removing radical structures from many sources, number of pathological events can occur. As a result of this, the polyunsaturated fatty acids, present in the membrane structure are oxidized, thus the lipid peroxidation process starts. Malondialdehyde (MDA) as an biological marker used to determine the level of oxidative damage in the systemic circulation forms as a result of the conversion of lipid hydroperoxides to aldehydes and carbonyl compounds. The current analytical methods for the analysis of MDA have limitations. In this study, it was planned to develop a new analytical method that is much faster, more accurate and lower cost process for MDA determination. For this aim, modified electrode surface was prepared by modification of carbon electrode surfaces with gold nanoparticles. Modified surfaces were characterized by cyclic voltammetry, electrochemical impedance spectroscopy, scanning electron microscopy, atomic force microscopy and contact angle measurement techniques. The modified surfaces were investigated as MDA sensor.

Acknowledgement: This paper was supported by the Selcuk University Scientific Research Projects Coordination Project Number: 1522021.

Keywords: Biosensor, Malondialdehyde, Modified Surfaces

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Cystine-selective fluorescent probe based on yellow emitting nitrogen doped carbon quantum dot

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Abstract

Carbon quantum dots (CQDs) have been agreed as fluorescent nanomaterials with unique optical and electronic properties and they have been the potential alternatives for organic emitter in the recent years. In this study, a novel heteroatom (nitrogen) doped CQDs, generated by bottom-up approach between L-ascorbic acid and p-phenylenediamine as carbon and nitrogen source, were synthesized by hydrothermal reaction at 160 ^{0}C for 10 h. After purification by silica column chromatography, the structural (FT-IR, XPS) and morphological (AFM, TEM) characterizations were performed in detail. CQDs exhibited a maximum fluorescence emission centered at 530 nm excited at 356 nm. The obtained strong yellow emitting fluorescent CQDs were used as sensitive and selective fluorescence probe in order to detect cystine among sulfur-containing amino acids (cysteine, homocysteine and methionine) based on its quenching effect in aqueous media around physiological conditions. As a result, CQDs can be evaluated as a promising fluorescent probe for cystine.

We express our thanks to the Scientific and Technological Research Council of Turkey (TUBITAK) for financial support (215Z222).

Keywords: Carbon quantum dot, Yellow emission, Cystine detection

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A BODIPY-bearing pillar[5]arene for mimicking photosynthesis based on multi-fluorophoric light harvesting system

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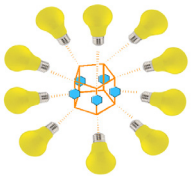
Abstract

Among macromolecules, pillararene compounds linked with methylene bridges at para-positions have multi-terminals and they can be possessed for various applications such as smart polymer, drug delivery, chemosensor, transmembrane. To make a more efficient process of the electron-transfer, various donor and acceptor linked dyads were developed. Among most dyads, Bodipy's have been extensively used as antenna molecule in artificial photosynthetic systems due to their excellent properties such as high molar absorption coefficient, high fluorescence yield, long lifetimes good photostability. Herein, we submit for the original synthesis, characterization, energy transfer mechanism of the pillar[5] arene based on Bodipy and its reactants by employing of infrared, ^{1}H, ^{11}B, ^{13}C, ^{19}F-NMRs, UV-vis, fluorescence spectroscopy, melting point apparatus, CHN elemental analysis and mass spectroscopy. Preliminary UV-vis, fluorescence and excitation studies in in CH_2Cl_2 as solvent revealed that a novel fluorescence resonance energy transfer (FRET) system based on the interaction of pillar[5]arene and Bodipy derivative was disclosed. ε_{max} of target molecule reached to a maximum value and calculated as 955 000 $M^{-1}cm^{-1}$. This fluorescent macromolecule worked well for mimicking photosynthesis a light harvesting system with highly energy transfer efficiency up to 92%. Therefore, this study not only provided a novel model for fabricating mimicking photosynthesis system but also increased the potential bio-medical applications of pillararenes and Bodipy in the field of optoelectronic materials.



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Congress
25-27 April 2018, www.molbiotech.gen.tr

Detailed chromosome measurements and karyotype asymmetry of *Vicia* L. (Fabaceae) some taxa from Turkey

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Abstract

This study examined the detailed chromosome measurements and karyotype asymmetries of seven taxa, Sect. *Cracca* Gray-*V. articulata* Hornem., Sect. *Cracca* Gray-*Vicia cassubica* L., Sect. *Cracca* Gray-*V. villosa* Roth. subsp. *villosa*, Sect. *Vicia* L.-*V. noeana* Reuter ex Boiss. var. *noeana*, Sect. *Vicia* L.-*V. sativa* L. subsp. *sativa*, Sect. *Vicia* L.-*V. peregrina* L., and Sect. *Ervum* (L.) Gray-*V. caesarea* Boiss. & Bal. which is represented by only these seven taxa in Turkey, in the genus *Vicia*. *V. cassubica*, *V. noeana* var. *noeana*, *V. sativa* subsp. *sativa*, *V. caesarea* have 2n = 12 chromosomes. *V. articulata, V. villosa* subsp. *villosa, V. peregrina* have 2n = 14 chromosomes in somatic cells. Total chromosome lengths are 2.93 µm and 4.99 µm in *V. articulata*, 2.09 µm and 4.73 µm in *V. cassubica*, 1.86 µm and 3.36 µm in *V. villosa* subsp. *villosa*, 4.23 µm and 6.05 µm in *V. noeana* var. *noeana*, 2.07 µm and 3.72 µm in *V. sativa* subsp. *sativa*, 4.32 µm and 7.21 µm in *V. peregrina* and 2.39 µm and 5.78 µm in *V. caesarea. V. articulata* is the most symmetrical karyotype, while *V. villosa* subsp. villosa is the most asymmetrical karyotype in intrachromosomal asymmetry including parameters of MCA, AsK, TF, Syi, A1, and A. However, the asymmetrical karyotypes are different in interchromosomal asymmetries. While *V. noeana* var. *noeana* is the most symmetrical karyotype in CVCL, Rec, and A2. *V. caesarea* is the most asymmetrical karyotype in only CVCL and A2. Unlike all parameters, *V. cassubica* is the most asymmetrical karyotype in Rec value.

Keywords: Asymmetry index, *Vicia*, Fabaceae, Karyotype

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Congress
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Investigation of the relationship between matrix gamma carboxyglutamic acid protein G7A gene polymorphism genotype distributions and diabetic nephropathy development

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Abstract

Diabetic nephropathy (DN) is characterized by without other renal diseases the persistent positive urine albumin rod in a diabetic patient or by albumin excretion of more than 300 mg per day. Matrix Gamma Carboxyglutamic Acid Protein (MGP) is a potent inhibitor of calcification in blood vessels. Human MGP gene is localized on the genomic chromosome 12p (12p12.3). MGP G7A gene polymorphism is localized in the promoter region of the MGP gene and is characterized by a guanine / adenine base translocation. It is known that MGP G7A gene polymorphism is associated with Type 2 Diabetes Mellitus (DM). Therefore, the aim of this study is to investigate the relationship between the development of DN, one of the microvascular complications of Type 2 DM, and genotype distributions of MGP G7A gene polymorphism. Our study was performed by 60 diabetic nephropathy patients and 55 healthy controls. DNA isolation was performed from peripheral blood containing EDTA obtained from patient and control groups. The purity and quality of the isolated DNAs were determined by measuring with a nanodrop spectrophotometer. Polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) methods were used to determine MGP G7A gene polymorphism genotype distributions. The significant difference was not found statistically in MGP G7A gene polymorphism genotype distributions between DN patients and healthy control group ($p>0,05$) (Chi-Square Test). In our study, genotype distributions of MGP G7A gene polymorphism were found to be not a genetic risk factor for the development of DN.

Keywords: Type 2 DM, DN, MGP G7A gene polymorphism, PCR, PFLP

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Congress
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Reversible ımmobilization of laccase for reactive blue-24 removal

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Abstract

Laccase (1.10.3.2) enzyme has gained great attention due to its efficient and low-cost biodegradation ability on organic pollutants including synthetic dyes. Laccases are commonly found in white-rot fungi and they are capable of oxidizing various phenolic compounds. Thus, they have been reported as promising tools in the field of synthetic dye degradation from waste water. In addition, their enzymatic reaction promotes formation of less toxic compounds than selected dyes. New generation polymeric systems, cryogels, were chosen as stationary phases for reversible laccase immobilization in presented study. Cryogels with interconnected supermacropores are easily functionalized to carry different molecules such as drugs, enzymes and cells. In this study, reversible immobolization of laccase from Trametes versicolor on poly(hydroxyethyl methacrylate-N-methacryloyl-L-phenylalanine) (PHEMAPA) cryogel discs was performed. Laccase immobilized PHEMAPA cryogel discs was characterized by scanning electron microscopy and swelling tests. Effects of different experimental conditions on reversible immobilization of laccase on PHEMAPA cryogel were investigated. Reactive blue-24 removal with laccase immobilized PHEMAPA cryogel discs was carried out at different pH and temperature conditions. Laccase activity was determined by using guaiacol as substrate. In conclusion, laccase immobilized PHEMAPA cryogels are potentially suitable for Reactive Blue-24 removal with high catalytic activity.

Keywords: Laccase, cryogel, dye removal, reactive blue-24, enzyme immobilization

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Effect of type 1 diabetes and resveratrol on gene and protein expression of renal insulin signaling pathway components

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Abstract

Insulin is a hormone that is produced by the beta cells of the pancreas and regulates the amount of sugar in the blood. Diabet occurs due to loss of function and lack of insulin leading to increase blood sugar and causing glycosuria. In this study, it was aimed to investigate changes in gene and protein expression of renal insulin signaling pathway components with type 1 diabetes and resveratrol which is a potent antioxidant molecule. Male Wistar rats of equal age were divided into four groups as follows; diabetic (n=12), control (n=12), diabetic group supplemented with resveratrol (n=9), control group supplemented with resveratrol (n=12). Diabetes was induced in respective groups with single intraperitoneal streptozotocin (55 mg/kg) administration. One week after the diabetes, resveratrol was given as 20 mg/kg/day throughout 3 weeks. While changes in protein expression were determined by western blot analysis, changes in gene expression were determined by qPCR. The components of insulin signaling elements were up-regulated at gene expression levels in diabetic rat kidney tissues, and this increase in gene expression leads the protein levels to be enhanced. Resveratrol treatment decreased the sensitization of insulin signaling towards the normal levels at gene expression level. The results of this study contain the supplementary data for the molecular mechanisms of the diabetes induced changes in the kidney tissues to put forward to orient new studies searching for new drugs and gene treatments for diabetes.

Keywords: Diabetes, Kidney, Resveratrol, Insulin signaling Pathway, Gene expression, Western Blot

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Enantioselective hydrolysis of racemic naproxen methyl ester with the encapsulated lipases using calix[4]arene derivative containing piperazine

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Abstract

Naproxen, is a non-steroidal anti inflammatory drug and an important member of the 2-aryl propionic acid class of compounds. The anti-inflammatory activity of the S-form of Naproxen is 28-fold greater than that of the R form. For this reason, only the S-form of Naproxen is used as a drug for humans, and significant research efforts have been focused on the production of the S-form of Naproxen as a single enantiomer. Lipases are one of the most widely used enzymes in biotechnology, where they have been used in the organic synthesis and kinetic resolution of racemic com compounds. In particular, Candida rugosa lipase (CRL) is an important industrial lipase, and has been used in a wide variety of hydrolysis and esterification reactions. Lipases also exhibit good selectivity, which has allowed them to be used as important biocatalysts in several other applications, including the synthesis of chiral drug intermediates and nutraceutical lipids. Candida rugosa lipase has been immobilized on a variety of calix[4]arene derivatives using the sol–gel encapsulation technique, and the catalytic activities of the resulting encapsulated lipases towards the hydrolysis of p-nitrophenylpalmitate and the hydrolytic kinetic resolution of racemic Naproxen methyl ester were evaluated using standard techniques. The results revealed that the calix[4]arene-based immobilized encapsulated lipase gave higher levels of enantioselectivity and conversion than the free encapsulated lipase.

Keywords: Calix[4]arene , Candida rugosa lipase , Enantioselectivity , Naproxen , Sol–gel encapsulation

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Investigation of the antibacterial activity and fluorescence properties of N-(2-((pyren-4-yl)methyleneamino)ethyl-5-nitropyridin-2-amine)

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Abstract

Antibiotic-resistant bacteria pose an enormous threat to the treatment of a wide range of serious infections because of their ability to develop resistance mechanisms against virtually all commonly used antibiotics. Fortunately it is now well-known that Schiff base and their derivatives have been shown to exhibit a broad range of biological activities, including antifungal, antibacterial, antimalarial, antiproliferative, anti-inflammatory, antiviral, and antipyretic properties.

Schiff bases have also been widely studied in connection with catalysis of many reactions due to the versatility of their steric and electronic properties and are promising materials for optoelectronic applications and their industrial applications. Correspondingly the aim of this study is to find the novel Schiff base and with good antibacterial activity and to investigate their fluorescence properties. Therefore, this study was carried out to investigate the fluorescence properties of N-(2-((pyren-4-yl)methyleneamino)ethyl-5-nitropyridin-2-amine). Multi-emission spectra of the novel schiff base in tetrahydrofuran were measured by changing excitation wavelengths. Then fluorescence intensities of the schiff base in excitation (λex) and emission wavelengths (λem) were determined and antibacterial activity of the Schiff base was observed by means of disc diffusion method. We studied to determine the biological activities of N-(2-((pyren-4-yl)methyleneamino)ethyl-5-nitropyridin-2-amine) against various bacteria [Staphylococcus aureus ATCC 25923, Staphylococcus aureus ATCC 43300 MRSA, Salmonella enteritidis ATCC 13076, Enterococcus faealis ATCC 29212, Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 25992] and the antibacterial data given for the compounds presented in this study allowed us to state that the novel antibacterial agent had a better activity on the Pseudomonas aeruginosa ATCC 27853 .

Keywords: Shiff base, antibacterial activity, fluorescence properties

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The biological ınvestigation of essential oils of some *achillea* species

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Abstract

Achillea L. (Asteraceae) is a widely distributed medicinal plant in the world and represented by 140 species in the world. *Achillea* L. species are commonly used in Turkish Traditional Medicine for the treatment of wounds, bleedings, headache, inflammation, pains, spasmodic diseases, flatulence and dyspepsia and hemorrhoids for years (1,2). The genus *Achillea* is rich in terpenoids and flavonoids, which are possible bioactive compounds. Monoterpenes were reported to be the major constituents of essential oil of the genus although high levels of sesquiterpenes were quantified (3). Among the monoterpenes 1,8-cineole, found in almost every essential oil, was reported to be the most frequently identified component. Furthermore, it was also reported to be the major compound in about one third of yarrow essential oils. Compounds having bornane skeleton such as camphor and borneol were reported to be the second and third most frequently charactarized components of *Achillea* oil and they were described several times as major compounds. Antioxidant, cytotoxic, antimicrobial, anticholinesterase, urease and tyrosinase enzymes activities of essential oils obtained by hydro-distillation method from *A. biebersteinii*, *A. wilhelmsii* subsp. *wilhelmsii*, *A. aleppica* subsp. *zederbaveri, A. vermicularis, A. monocephala, A. nobilis, A. coarctata, A. teretifolia* species were determined. It has been determined that all essential oils show low-moderate antioxidant activities in ABTS, CUPRAC and DPPH methods. In the method of butyrylcholinesterase, *A. wilhelmsii* subsp. *wilhelmsii* (collected from Niğde) (İnhibition: %77.67±1.03), in the urease method *A. vermicularis* (collected from Mardin) (İnhibition: %55.26±2.01), in the tyrosinase method *A. vermicularis* (collected from Diyarbakır) (Inhibition: 43.07% ± 1.32) was found to be more active. It was determined that all extracts tested showed high-moderate cytotoxic effects against PDF, HT29 and MCF-7 cell lines.

Keywords: Achillea, Antioxidant, Urease, Tyrosinase, Antimicrobial, Cytotoxic activity

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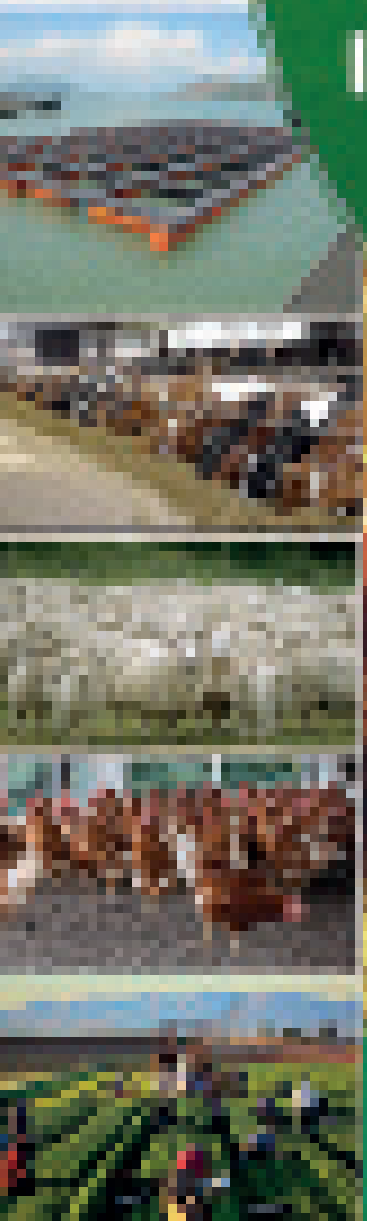

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Quantitative determination of sterols in olive oil deodorizer distillate by gc-ms and gc-fıd

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Abstract

The most important by-product of edible oil refining is the deodorizer distillate (DD) obtained in the deodorization stage. Basically, deodorization is the final key step of the refining process accountable for removing targeted volatile compounds which are liable for producing unacceptable odor, color, taste and flavor in the oil. Increased use of industrial waste and by products fits the requirement of industry to fulfill with environmental rules. The replacement of natural products for synthetic materials has gained worldwide consideration in the food, pharmaceutical and other industries. Therefore, extra virgin olive oil DD (OODD) has been utilized as a natural source of FFAs, tocopherols, sterols, squalene in many fields. In this study, an automated GC-MS and GC-FID system for quantification of sterol compounds in OODD was used. It was seen that OODD has campesterol, stigmasterol and B-sitosterol in the level of 12,73 ± 0,54, 109,92 ± 2,04 and 15,76 ± 0,40 mg/kg distillate, respectively.

Keywords: Deodorizer distillate, Olive oil, Sterol

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Selective separation of acetic acid from its aqueous binary solutions with formic acid

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Abstract

Selective recovery of carboxylic acids from their mixed solutions with high efficiency is a challenging problem in bio-industry. Several methods have been tested for the purpose. Most of them were not successful or expensive due to high energy demands. One of the most advantageous methods is reactive extraction. It contains chemical extraction with physical extraction and previously shown to be successfully used in similar cases. The present study is on the selective extraction of acetic acid (AA-0.25 M) and formic acid (FA-0.25 M) from their binary solutions using a tertiary amine (tri-n-octylamine, TOA) as the extractant dissolved in various organic solvents. Effects of aqueous pH, extractant concentration and solvent type on selectivity and extraction efficiency were probed. At natural pH of the aqueous solution, the extractant is expected to prefer to react with the stronger acid (FA, pKa=3.75). This was also observed and FA was extracted preferably at pH 2.2. The separation factor (α) was about 7-8 using 0.3/0.5 M TOA in all solvents studied. However, purity was low (~58%) which is not desired. The increase in pH decreased the amount of extracted acid but increased the purity. Differently, the weaker acid (AA, pKa=4.75) was preferably extracted most probably due to the higher concentration of undissociated acid. Using 0.5 M TOA, 33% and 5% of AA and FA was extracted, respectively. At a pH of 5, the α and purity were about 10 and 87%, respectively. Therefore only 3-4 consecutive steps will provide high purity FA and AA solutions.

Keywords: Selective recovery, Acetic acid, Formic acid, Reactive extraction, Extractant

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Novel electrochemical sensor based on pillar arene for determination of histamine

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Abstract

The biogenic amine content of a variety of foods has been widely studied because of their potential toxicity. At high concentrations there is a risk of food intoxication, whereas moderate levels can lead to food intolerance. The aim of this study was to develop a modified electrode based on pillar arene for the determination of histamine which is important in food quality. For this purpose, glassy carbon electrode was modified by using pillar arene distributed in gelatin biopolymer. Scanning electron microscope which is a surface imaging technique was used for the determination of surface modification for the modified electrode. Furthermore, electrochemical behavior of the electrodes were examined utilizing cyclic voltammetry and electrochemical impedance spectroscopy techniques. Moreover, optimum working conditions, performance factors and analytical applicability of the modified electrode in real samples were investigated. Pillar-aren–based histamine electrode showed the best performance characteristics; the linear working range was 4.2×10^{-7}–4.2×10^{-5} M, sensitivity was 60 A M^{-1}, limit of detection was 1.12×10^{-7} M and relative standard deviation of the calibration graphs of repeatability was 5%.

Keywords: Modified electrode, biogenic amines in foods, electrochemical sensor

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Fig latices have cytotoxic effects on three different human cancer cell lines

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Abstract

Fig latex (FL) samples collected from *Ficus carica* cultivars Sari Lop (SL) and Bursa Black (BB) were stored at -20°C and lyophilized. Human colon cancer line HT-29 and prostate cancer line PC3 were maintained in RPMI 1460, and human pancreatic cancer line MIAPaCa-2 was maintained in DMEM. Both media were supplemented with 10% fetal bovine serum (FBS) and cells were incubated at 5% CO2 and 37°C. Lyophilized FL samples were resuspended in deionized water and filter sterilized. Cells were treated with 0-100 µg/ml of FL with deionized water as negative control for up to 72 h. Cell viabilities after the treatment were measured with MTT (3-{4,5-dimethylthiazol-2yl}-2,5-diphenyl tetrazolium bromide) assay. Experiments were performed three times in triplicates. The results were analyzed with analysis of variance test. Both FL samples showed cytotoxicity on cancer cell lines in dose- and time-dependent fashion. The BB-FL had statistically higher ($P<0.05$) cytotoxic effects on all three cell lines compared to the SL-FL. The BB-FL had statistically higher ($p<0.05$) cytotoxic effects on HT-29 cells than SL-FL at doses as low as 25 µg/ml for 24 h. However, the difference disappeared as the FL concentration increased to 100 µg/ml and the treatment time increased to 72 h. Same trend was observed on PC3 cells with almost complete cytotoxicity by 100 µg/ml BB-FL in 72 h versus 20% survival rate by 100 µg/ml SL-FL. The SL-FL at 25 µg/ml killed 40% of the MIAPaCa-2 cells while BB-FL at 25 µg/ml killed over 80% of the cells in 24 h.

Keywords: *Ficus carica*, fig latex, cancer cell line, MTT, cytotoxicity

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A novel schiff-base based ‘’turn on” fluorescent sensor for al3+ ıons

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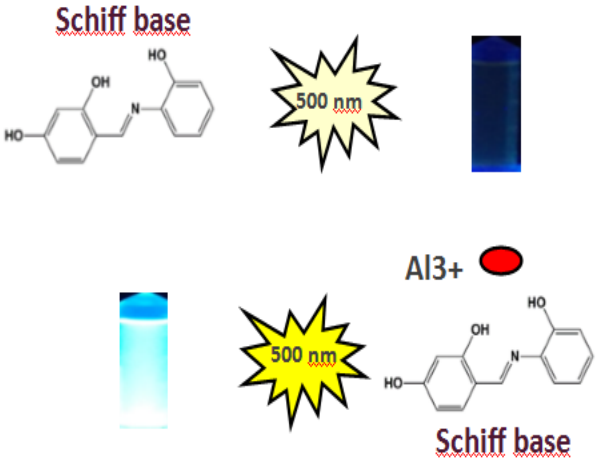
Abstract

Anexcellent chemosensor receptor is prepared by simple Schiff base type reaction and it is exhibited a ‘‘turn on’’ mode with high sensitivity in the presence of Al^{3+} ions. Upon binding of Al^{3+}, a significant fluorescence enhancement with a turn on ratio over 335-fold is triggered. But, other metal ions have no such significant effect on complexation with same receptor. From these spectroscopic data, it is concluded thatthis receptor could be used as Al^{3+} probe. This receptor can also be used as a colorimetric sensor for Al^{3+} ions under UV-light by naked eye ’’from toneless to brilliant fluorescent blue’’.



Keywords: Fluorescence, Schiff-base, Sensor, Aluminium ions

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Saccharomyces cerevisiae SAGA complex is a repressor of TPS1 gene

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Abstract

Trehalose is a nonreducing disaccharide, formed by two glucose units. The synthesis of trehalose is catalyzed by an UDP-glucose-dependent trehalose synthase (TPS) complex composed of catalytic and regulatory subunits. *TPS1* and *TPS2* encoded for trehalose-6-phosphate synthase and trehalose-6-phosphate phosphatase, respectively. *TPS3* and *TSL1* coded for two regulatory subunits of the TPS complex. Trehalose- 6-phosphate synthetase enzyme catalyzes the joining of glucose-6-phosphate with the glycosyl unit from UDP-glucose. *TPS1* promoter includes STRE sequences necessary for stress response, such as nutrient starvation. It is known that chromatin remodeling complexes regulate transcription. SAGA complex is a 2MDa multiprotein chromatin remodelling complex that harbors two known enzymatic modules mediating acetylation and deubiquitination of histones. In our research we tested the effect of Spt7, the subunit of the SAGA complex, on *TPS1* transcription both in normal and stress conditions. We have found that transcription of *TPS1* gene increased by at least 5-fold in *Aspt7* mutants than wild type. This result indicated that SAGA complex is essential for the regulation of *TPS1* transcription. In addition, we found that *TPS1* transcription is was constitutive during nitrogen starvation. Our results showed that SAGA chromatin remodeling complex is essential for the repression of *TPS1* gene expression.

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Keywords: *TPS1*, SAGA complex, Trehalose, *Saccharomyces cerevisiae*

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Inhibition effects of phenolic compounds on biogenic amines formation by spoilage and pathogenic bacteria in ornithine decarboxylase broth

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Abstract

In this study, ornithine decarboxylase broth was prepared using phenolic compounds such as carnosic acid, kaempherol and luteolin. Spoilage bacteria in the flora of aquatic products such as *Photobacterium damselae, Proteus mirabilis, Enterobacter cloacea, Serratia liquefeciens* and *Pseudomonas luteola* and pathogenic bacteria such as *E.coli, Staphylococcus aureus, Yersinia enterocolitica, Salmonella paratyphi* A and *Enterobacter faecalis* have been investigated in ornithine decarboxylase broth. As a result of the study, significant differences were observed by bacteria in terms of ammonia and biogenic amine production ($P<0.05$). It was observed that kaempherol was more effective in the inhibition of biogenic amines than carnosic acid and luteoline in the study. The major amines produced in the ornithine broth were putrescine, cadaverine, serotonin and dopamine. The highest histamine production was achieved by *E.faecalis* supplemented with luteolin from pathogenic bacteria (10.95 mg/L). The highest putrescine production was by the control group of *P.luteola* (246.59 mg/L) and the highest cadaverine production was by the carnosic acid group of *E.faecalis* (71.86 mg /L). The highest production of tyramine and dopamine was observed by *E.cloacea* (751.96 and 156.10 mg / L, respectively), which is a spoilage bacterium. While bacterial production of histamine, putrescine, serotonin and cadaverine was significantly inhibited by kaempferol supplementation, spermine production was largely inhibited by luteolin and dopamine and agmatine production by carnosic acid supplementation.

Keywords: Phenolic compounds, histamine, ornitin, spoilage bacteria, pathogenic bacteria

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Comparison of bone marrow and adipose tissue derived mesenchymal stem cells properties in musculoskeletal tissue engineering

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Abstract

In the recent years, researchers have been searching for more effective ways of bone and cartilage defect treatments in musculoskeletal tissue regenerations. Tissue engineering is a mainly used method for this aim. Tissue engineering techniques will play an important role in the future development of artificial organs and the application of them. Mesenchymal Stem Cells (MSCs) are highly prefered in regenerative medicine due to their multi-differentiation potential, anti-inflammatory effects, safety and ease in harvesting. MSCs also possess paracrine and immune modulating effects through growth factor and cytokine release. Different sources of MSCs are used in regenerative medicine. However, this presentation is based on the usage of MSCs, isolated from human adipose tissue (ASCs) and from bone marrow (BMSCs). ASCs and BMSCs differ in some ways. There are many methods for the isolation of MSCs from bone marrow and adipose tissue. MSC isolation from ASCs has an easier procedure than BMSCs, but the cell density is the least. ASCs are possible to obtain in high amounts from lipoaspirates, have rapid cell proliferation and it has a less invasive procedure than bone marrow. Immunophenotypic and immunohistochemical characterizations should be done for the identification of MSCs. Also, their differentiation capacities into adipogenic, osteogenic, chondrogenic or fibroblastic should be investigated. Some of these studies and the main differences of these two sources of MSCs have been compiled in this presentation.

Keywords: Bone Marrow, Adipose Tissue, Mesenchymal Stem Cell, Musculoskeletal Tissue

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Investigation of the effect of methylenetetrahydrofolate reductase A1298C gene polymorphism on the development of diabetic nephropathy in patients with type 2 diabetes mellitus

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Abstract

Diabetic nephropathy (DN) is one of the microvascular complications of Type 2 diabetes mellitus (DM). Vascular damage to DN, which is not fully understood pathogenesis, is of utmost importance. Among the genes that are effective in the development of diabetic nephropathy from microvascular complications of type 2 DM are the methylenetetrahydrofolate reductase (MTHFR) gene. The human (MTHFR) gene is localized in the telomeric region of chromosome 1 (1p36.3) and encodes the enzyme MTHFR. The MTHFR A1298C gene polymorphism is characterized by a protein Glutamine/Alanine exchange in the C-terminal regulatory region of the MTHFR gene as a result of the Adenine/Cytosine substitution at position 1298 in the 7th exon. The aim of our study was to determine the association between MTHFR A1298C gene polymorphisms and the development of diabetic nephropathy in patients with Type 2 diabetes mellitus. Our study was conducted with a total of 116 people, consists of 61 DN patients and 55 healthy controls. DNA was isolated from peripheral blood, containing ethylenediamine tetraacetic acid. DNA quality was measured with a Nanodrop device at 260 and 280 nanometer wavelengths. The genotypes of MTHFR A1298C gene polymorphism were identified through usage of the Polymerase Chain Reaction (PCR) device and restriction fragment length polymorphism methods (RFLP). The MTHFR A1298C genotype distributions in patient group with diabetic nephropathy did not differ from those in control group (p>0,05) (Chi-Square test). Our study showed that MTHFR A1298C gene polymorphism are not a genetic risk factor for the development of DN.

Keywords: Type 2 DM, DN, MTHFR A1298C Gene Polymorphism, PCR, PFLP

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X-Ray diffraction study of *Scytalidium thermophilum* catalase in the complex with Aminotriazole

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Abstract

Catalases (EC 1.11.1.6) are redox enzymes responsible for the dismutation of hydrogen peroxide into water and molecular oxygen. The crystal structures of fifteen heme catalases, including one isolated from the thermophilic fungus *Scytalidium thermophilum*, have been solved at high resolution. These structures show that the heme catalases are tetramers and each of the four active sites consists of a pentacoordinated-iron protoporphyrin IX prosthetic group with a tyrosinate axial ligand. Some catalases also contain a NADPH cofactor tightly bound at the periphery of each subunit. Our previous studies have indicated that, in addition to the hydrogen peroxide degrading catalatic activity, the catalase from *S. thermophilum* (CATPO) possesses an oxidase activity. This enzymatic activity is oxygen-dependent and is inhibited by classic catalase inhibitors, including 3-amino-1,2,4-triazole (3TR). In order to better understand this oxidative reactivity, we determined the crystal structure of the enzyme-inhibitor complex of CATPO with 3TR at 1.95 Å resolution. Surprisingly, and in contrast to other structural reports of 3TR complexes with catalases, we do not observe the 3TR bound at the heme. Instead, the inhibitor occupies a surface pocket to one side of the heme. This structure has led us to hypothesize that this binding site is in fact the binding site for substrates for CATPO's oxidase activity. In addition, we propose that the oxidase activity of CATPO may represent an alternative protection strategy for catalases, where small organic molecules are used in place of NADPH.

Keywords: *S. thermophilum*, catalase, oxidase, aminotriazole, NADPH

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Assessment of ion channel activity from action potentials: skewness

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Abstract

Membrane potential recording from excitable cells by microelectrode is an essential but still mostly preferred method to determine ion channel behavior that contributes to action potentials (AP) shaping. This information is provided by analysis of AP and interpretations of some essential parameters, but remains incapable to explain some unusual situations. We aimed to investigate the availability of the skewness parameter which is used for analysis of statistical distributions for detailed examination of AP depolarization phase. For this reason, APs evoked by supramaximal stimulations were recorded from diaphragm muscle cells of adult Sprague-Dawley rats. APs recorded from diaphragm preparations that 4-aminopyridine (0.3 mM) applied to block potassium currents (4AP group, N=4) and N-Methyl-D-Glucamine (NMDG) replaced medium (NMDG group, N=4), to block sodium currents. APs recorded also from control preparations (CON, N=10) without any application. By choosing the membrane potential (mV) as an independent variable, membrane potential change (mV/ms) were plotted only for depolarization phase then the skewness value were calculated for each group. Skewness were found positive in CON which means skewed to the left, while this positivenes were found to be increased in 4AP and decreased in NMDG group. These changes were significantly different ($p<0.05$). We have shown that, with the skewness parameter which reflects the behavior of time independent but membrane potential dependent change of depolarization velocity, changes in channel kinetics can be investigated.

Keywords: Diaphragm, action potential, rat, skewness

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IBA induced shoot regeneration during rooting of lentil cultivars

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Abstract

Both rooting and shoot proliferation of lentils is a complex process and difficult during *in vitro* cultures due to leaching out of phenols, oxidative stress or injury due to interaction of plant growth regulators with the explants, ensued by necrosis, deformation, abnormal regeneration behavior etc.. This cause inhibation to growth and propagation of lentil under aseptic conditions. The main objective of the study was promote proliferation of multiple clones of the lentil genotypes Ali Dayı and Kayı 91 without involving long and complex tissue culture steps using *in vitro* regenerated shoots of the newly germinated seeds using different concentrations of (0.25, 0.5, 1.0 and 2.0 mg/l) IBA. Although the exact mechanism of IBA functions is largely unknown and is very complex; genetic evidence suggests that it converts into IAA through β-oxidation of fatty acids and promotes rooting both under *in vitro* and *ex vitro* conditions. IBA is also known to regulate different aspects of plant growth and development such as cell elongation, division and differentiation. The results showed that 0.25 mg/l IBA had high potential to induce shoots during rooting. However, the treatments with 0.50, 1.0 and 2.0 mg/l IBA were inhbitive. These shoots of both cultivars induced callus of variable size at the basal tips ensued by variable shoot regeneration. It was concluded that IBA undertake bipolar functions of rooting and shoot proliferation at lower concentrations. However, bipolarity of the IBA is lost at >0.50 mg/l concentration. This aspect of *in vitro* plant regeneration in lentil tissue culture and biotechnology provides a rapid and cost-effective plant proliferation protocol for *in vitro* studies. The protocol could facilitate biotechnological studies in effective manner for genetic transformation, genomic and proteomic studies.

Keywords: lentil, shoot induction, rooting, IBA, *in vitro*

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Molecular analysis of elastin gene mutations in autosomal dominant cutis laxa

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Abstract

ADCL is characterized by loose and inelastic skin, pulmonary emphysema, aortic root dilation, and peripheral pulmonary aortic stenosis. Our goal was to evaluate the impact of elastin gene mutations on TGFβ signaling and molecular pathology of ADCL. Dermal fibroblasts from four patients with ELN mutations in exon 34 or exon 30 and controls were used. Increased intracellular TGFβ signaling was found in patients with exon 30 mutations, despite unchanged extracellular TGFβ activity. TGFBR1 levels were increased at the protein and the RNA level. Patients with exon 34 mutations had normal TGFβ signaling. Our results indicate mutation-specific TGFβ signaling changes in ELN-related cutis laxa patients, which may influence to disease severity. SMAD6 and SMAD7 mRNA expression levels were decreased in ADCL patients. Elastin assay showed decreased elastin deposition in ADCL cells and long-term TGFβ treatment improved elastin deposition. Semi-quantitative RT-PCR experiments showed increased expression of the mutant compared to the wild-type allele in ADCL cells under baseline conditions. Long-term TGFβ treatment normalized this allelic imbalance in expression. Therefore, we conclude that increased TGFβ signaling is a protective mechanism in ADCL at the molecular level. Uncovering the nature of connections between elastin and TGFβ may help developing treatments for cutis laxa. Our findings are relevant to complex diseases characterized by elastin degradation and TGFβ dysregulation, including aneurysms and chronic obstructive pulmonary disease that have major public health impact.

Keywords: Autosomal dominant cutis laxa, elastin, TGFβ

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Investigation of the distribution of two polymorphisms of *Hif1 Alpha* (rs11549465, rs11549467) in children with cleft palate/lips and their mothers

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Abstract

Palatogenesis is a metabolic event that occurs in the early stages of fetal life. This phenomenon involves many signaling molecules that including transcription factors. Most of the genes leading to the formation of syndromic cleft palate and lips are also found to lead to the formation of non-syndromic cleft palate/lips. It is known that environmental factors play a role in the formation of these pathogens by interacting with genetic factors. The ability of the cell to respond to changes in the oxygen pressure depends on the activation of a transcription factor family known as Hypoxia-inducible factors (HIFs). Studies have suggested that HIF activity must be induced via hypoxia for embryo and placenta development. It has been found that mouse embryos with homozygous *HIF-1 alpha* mutation can not survive and exhibit neural tube defects and cardiovascular anomalies. It has been reported that cleft lip/palate development to be associated with maternal hypoxia in humans. In this study, we aimed to determine the role of two polymorphic structure of *Hif1 alpha* (rs11549465, rs11549467) of child and mother on the development of cleft palate/lip by using the PCR-RFLP method. In G1790A polymorphic structure, we didn't observe any difference between mother and child and their controls. In C1772T, there were statistical differences in the comparison of maternal and child genotypes with control groups. In allelic comparisons, there was a statistically significant difference between mothers of cleft palate/lips children and control group. Although this was different in children, it was not as high as in mothers.

Keywords: *Hif1 alpha*, cleft palate/lip, rs11549465, rs11549467

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Interaction of nanofiber-producing amino calix[4]arene with caco-2 cell

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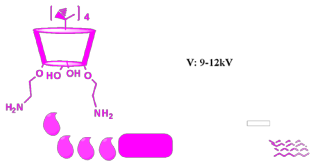
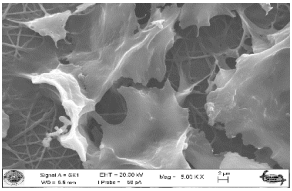
Abstract

Calixarenes, represent a third generation of supramolecular hosts. Their unlimited preparation of derivatives have provide opportunity to their use in various fields of research. They have been utilized such as ion-selective electrode, , sensor, chiral, column packing material and achiral catalyst, membrane, enzyme-mimic, ion and molecular transport and preparation of nanofibers of them by electrospinning. Due to having a cyclic structure and large surface area, calixarenes, can be functionalized easily with polar and apolar groups, be a good carrier for cations, anions and neutral molecules. Synthesis of new calixarenes nanofibers will arise innovative approaches in biomedical applications. In present study, electrospinning of p-tert-butylcalix[4]arene nanofibers with different functional groups and their cytocompatibility behaviour on one aspect of cell function: adhesion. Adhesion of anchorage-dependent cells is a necessary criteria for subsequent functions for different cell attacment. In this study, the biosynthesis of the calix [4] arene complex with the amino group of the produced amino group with the Caco-2 cancer cell is investigated by synthesizing and characterizing the calix [4] arene compound with the amino group and electrospinning this component. Cell attachment kinetics revealed that the Caco-2 attached to the nanofibers functionalized amino groups calix [4] arene at the same rate as to tissue culture plates. In this report, it was shown that 3D cultured systems designed with calixarene nanofibers provide excellent in vitro models, allowing the study of cellular responses in a setting that resembles in vivo environments.





Keywords: Calixarene; Electrospinning; Nanofiber; 3D-Cell Culture; Surface Funtionalization

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Chiral calixarene coated qcm sensor to sense alanine in aqeous media

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Abstract

Proteins are complex compounds which play vital role in human life. It is formed by smaller unit which is called Amino acids. Biosensors are analytical device which is can be investigated interaction between bioanalyte and sensing material. In biosensor application, there are various methods such as electrochemical, calorimetric, optical, acoustic. Among these methods, Quartz Crystal Microbalance (QCM) is acoustic sensor system which is used for antigen-antibody, enzyme-substrate interaction, drug carrier, detection of pollutants. QCM technique is defined as frequency change according to mass change on quartz crystal. In sensor application, there are many studies with regards to the polymeric and macromolecules materials which can be used as sensing material. Among these molecules, calixarene can be used for host-guest chemistry for construction of various receptors for charged or neutral molecules. In this study, a modified QCM sensor by means of coating a calixarene derivative onto QCM surface was used for sensing of alanine in aqueous media.

Keywords: Alanine, Chiral Calixarene, Quartz Crystal Microbalance, Sensor

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Recovery of lactic acid from aqueous solutions using an environmentally-friendly organic phase diluent and trioctylamine

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Abstract

Lactic acid (LA) is one of the most widely used low volatile carboxylic acids in the industry. Today most of the industrial demand is provided via bio-based productions. However, its recovery from fermentation broth is still a challenging problem. Reactive extraction is a promising method for the recovery of carboxylic acids, i.e., LA. It is preferred due to its high efficiency, low energy demand and process simplicity. However, use of organic solvents in the organic phase preparation is one of the main problems of the technique. To eliminate this, vegetable oils were shown to be efficient alternative to replace the toxic solvents. In this work, reactive extraction of lactic acid from aqueous solutions ([LA]0=0.1-1.5 M) using trioctylamine ([TOA]0=0.2-2.0 M) as the extractant dissolved in an environmentally-friendly solvent, safflower oil, was studied. Results showed that extraction efficiency increased with the increase in both TOA and LA concentration. Highest extraction values were obtained at pH 2, where the dissociation of LA is very low. In the ranges of the parameters studied, the highest efficiency and distribution coefficient were 83.1% and 4.9, respectively, with safflower oil. It is an acceptable value compared to the one (97%) obtained with 1-octanol, which is the state of the art organic phase diluent in the literature. Temperature negatively influenced the recovery, thus room temperature was the highest among the ones tested. This study presented how an environmentally-friendly solvent can be effectively used in the industry for the separation processes.

Keywords: Lactic acid, Reactive extraction, Safflower oil, Environmentally-friendly diluent, Trioctylamine

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Pre-yield analysis of kronos durum wheat using speed breeding technology

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Abstract

Development of a new variety takes five or six years in classical breeding. Speed breeding technology gives an opportunity to harvest five or six generations per year for spring wheat. Our aim was to evaluate pre-yield analysis of first generation of spring durum wheat cultivar Kronos (*Triticum turgidum var. durum*) grown by speed breeding technology in plant growth chamber. Seeds were sowed in 9 mm petri dishes. Germinated seeds were planted in pots including soil and torf mixture supplemented with fertilezer. Plants were incubated in plant growth chamber (24 hours light 12000 lux strength, 25+1 °C temperature and 60-70% moisture). Plants were watered by regularly and supplied with macro and micro nutrients. Tiller number, spikelets number, grain number, grain/spikelet, seed/spikelet, yield/spike and thousand grain weight were calculated for Kronos grown in speed breeding. Although average tiller number was 3, most of them did not have a seed. Spikelets number was 11.5, grain number was 18.5, seed/spikelet was 1.6, yield/spike was 0.67 g and thousand grain weight was 36.5 g for main tillers of first generation. The shorter tillers and weaker seeds were observed. As a coclusion, Kronos seeds were harvested at the 70th day after sowing instead of six months.

Keywords: Speed Breeding, Pre-yield analysis, Grain number, Durum wheat

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Isolation of bacteriorhodopsin-producing archaea from lake tuz-Turkey and their molecular ıdentification

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Abstract

Bacteriorhodopsin is one of the most valuable proteins of halophilic Archaea and acts as a proton pump. Due to its unique crystalline structure, it has a great variety of nanotechnological applications. In the present study, salt crystals collected from the shores of Lake Tuz in Turkey for isolation of halophilic archaea. The crystals dissolved in sterile water supplemented with NaCl and spread on agar plates containing Sehgal and Gibbons (SG) selective medium. The plates were incubated at 37 °C for 15 days for preparation of pure archaeal cultures. Then, each of archaeal isolates was grown in 100 ml culture medium containing 25 g NaCl, 2 g MgSO4•H2O, 0.3 g sodium citrate, 0.2 g KCl, 1 g bacteriological peptone with pH adjusted to 7.2. The cultures were shaken at 150 rpm, 37 °C under continuous illumination (40 W) for 48 h. After the incubation period, bacteriorhodopsin producing isolates were determined by purification of delipidated BR by aqueous-three-phase system from purple membranes of the isolates. According to the results, six of isolated archaea (NES-3, NES-4, NES-6, NES-8, NES-9 and NES-11) were determined as BR-producing microorganisms. 16S rRNA gene sequencing results of the isolates showed that NES-4 and NES-8 were assigned to *Haloarcula sp.*, NES-3, NES-6 and NES-11 to *Halobacterium sp.*, and NES-9 to *Halorubrum sp.* Consequently, the results of the present study contributed to widen understanding of bacteriorhodopsin-producing archaeal diversity of Lake Tuz.

Keywords: 16S rDNA, Bacteriorhodopsin, Halophilic archaea, Lake Tuz

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Structural analysis of lipid membranes with flavonoids

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Abstract

Alterations in membrane physical properties i.e. fluidity and thickness inevitably influence the membrane structure and its potential therapeutic applications. Flavonoids which are plant secondary metabolites have been shown to induce membrane lipid structure. This is of significance importance since many epidemiologic and clinical studies suggest a relationship between high flavonoid intake in diet and reduced risk of several diseases such as cardiovascular heart diseases, cancer and diabetes. One great focus of the food industry is to formulate functional foods with increased bioavailability. This is possible with understanding the details of how flavonoids interact with lipid self-assemblies and encapsulating them efficiently. Recent data regarding the influence of flavonoids on membrane structure and their encapsulation using curved membrane mesophases are presented.

Keywords: Flavonoids, phospholipids, synthetic membrane, interactions

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Fluorescent sensing of Zn2+ ions in living cells by a novel probe based Bisphenol A-Pyren

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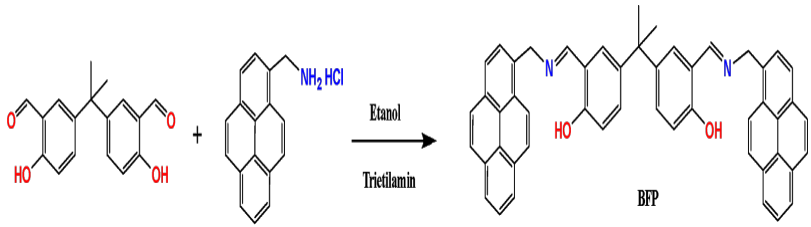
Abstract

The preparation of fluorescent sensors with high selectivity and sensitivity for heavy and transition metal ions has received noticeable attention because they play an important role in living systems. As the second most abundant metal ion, zinc is actively involved in diverse biological activities, such as structural and catalytic cofactors, neural signal transmitters or modulators, regulators of gene expression and apoptosis. Minute quantities of zinc are necessary for the living organism, but excessive amounts may damage the organism. Therefore, it is essential to discover efficient methods which can selectively and sensitively detect Zn2+. Of the many modes of detection available, fluorescence-based methods have attracted increasing attention in recent years owing to real-time detection, operational simplicity and good sensitivity.



In this study, we have designed and synthesized a novel bisphenol A-pyren (BFP) based sensor for Zn2+. The receptor BBP did not show any remarkable emission alone when the excitation wavelength was at 365 nm. But, Zn2+treatment resulted in a large increase in intensity at a wavelength of 485 nm in ethanol-water (95/5, v/v). In contrast, no fluorescence enhancement was detected after adding other metal ions. The detection limit of BFP for Zn2+ was 17.5 nM, which presented a pronounced sensitivity toward Zn2+. Also, possible utilization of BFP as bio-imaging fluorescent probe to detect Zn2+ in human prostate cancer cell lines was observed by confocal fluorescence microscopy

Keywords: Fluorescent, Sensor, Zinc, Living cell

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Pillar[5]arene based nonenzymatic tyramine sensor

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Abstract

Tyramine, which one of the best-known biogenic amine, is found in cheese, chocolate, beer, fermented foods and soya products. Because it is used as quality marker of food, analysis of tyramine is very important in food industry. In this work, nonenzymatic sensor for tyramine detection was constructed based on pillar[5] arene. The pillar[5]arene was dispersed in the gelatin (GEL) solutions and dropped on glassy carbon electrode (GCE) surface. Tyramine detection is based on the oxidation process of tyramine on the sensor surface at a 0.6 V. Surface properties of modified electrode were investigated by scanning electron microscope (SEM), atomic force microscope (AFM), electrochemical impedance spectroscopy (EIS) and cyclic voltammetry (CV). Optimum working conditions such as pH, working potential, amount of pillar[5]arene for GCE/Pillar[5]arene were examined. The linear working range of the sensor was 0.041-1.429μM with sensitivity of 7.613 nAμM-1 and limit of detection of 0.04 μM. The sensor shows good repeatability, reproducibility,stability and anti-interference property. The prepared nonenzymatic sensor was applied to determination of tyramine concentration in food samples.

Keywords: Tyramine, nonenzymatic sensor, pillar[5]arene

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Imaging of Al^{3+} ion in living cells by new fluorescent sensor based Bisphenol A-Hydroxyquinolin

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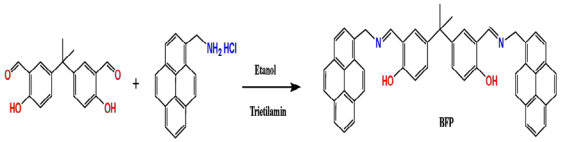
Abstract

Fluorescent probes for selective recognition of numerous biologically and environmentally relevant cations. An important natural metal element aluminum, is used widely in industrial fields for example water treatment, food additives, and medicines, naturally the production of light alloy. The toxicity of Al^{3+} induces damage of the central nervous system and is suspected in neurodegenerative diseases such as Alzheimer′s and Parkinson′s. Also, the high concentrations of aluminum appear in acidified lakes. Thus, detection of Al^{3+} is crucial in monitoring and controlling the concentration levels in the biosphere and in reducing its harmful effects on human health. Recently, great attention has been paid to the development of fluorescent and colorimetric chemosensors for the detection of Al^{3+} ion since they offer several advantages over other analytical methods from the point of high sensitivity, high selectivity, fast response times which can be used for real time monitoring, as well as non-destructive detection.



In this study, a novel fluorescent sensor based bisphenol A-hydroxyquinolin (BHQ) was synthesized and characterized by combination of 1H, 13C, APT, COSY NMR, FT-IR, and elemental analysis. The behaviours of BHQ toward different metal ions were determined by UV–vis and fluorescence spectroscopy. The fluorescence spectra changes showed that BHQ is highly selective for Al3+ over other metal ions in EtOH–H2O solution. Also, BHQ was successfully applied in fluorescence imaging of living cells and the confocal microscopy images indicated that cell-permeable BFA can visualize the changes of intracellular Al3+ in living cells.

Keywords: Fluorescent; Bisphenol A; Aluminum, living cell

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Development of antibiotic resistance in *Acinetobacter baumannii strains*

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Abstract

Acinetobacter strains have been the most frequently isolated factor in hospital infections, especially intensive care units in recent years. Increasing resistance to antimicrobial agents used in the treatment of infections due to Acinetobacter baumannii strains necessitated the search for new options for treatment. In this study, strains of Acinetobacter baumannii were isolated from samples obtained from different clinics in Amasya University Sabuncuoğlu Şerefeddin Training and Research Hospital Medical Microbiology Laboratory for 8 years (January 2010 - December 2017). Antimicrobial resistance and resistance to Acinetobacter baumannii strains have been researched for years, and it has been aimed to benefit in promoting rational antimicrobial use, in helping to identify empirical treatment options. The resistance of 1389 Acinetobacter baumannii strains against to colistin, tigecycline, amikacin, gentamycin, imipenem and meropenem were examined. The automated system VITEK2 (bioMerieux, France) was used for identification and antibiograms. Sensitivity results in suspicious cases were made with Mueller-Hinton (RTA) agar with Kirby Bauer Disk Diffusion Method and the results were evaluated according to CLSI criteria. Strains isolated from different services, high resistance was observed in carbapenem group antibiotics between 2010 and 2017, while 2% Acinetobacter baumannii was resistant to colistin on average. It is important to ensure that resistant strains are spread in the hospital environment and infection control measures are strictly adapted to prevent the transmission of antibiotic resistance to other bacterial strains. Resistance development should be monitored continuously and rational antibiotic usage policies should be applied.

Keywords: *Acinetobacter baumannii*, Antibiotic resistance, Colistin

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Genetic diversity of malus sieversii naturally growing wild apple species in Kyrgyzstan

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Abstract

There are many plant species in the Central Asian region that are endangered and must be protected. M. sieversii and M. niedzwetzkyana were reported as endangered species and were taken on the red list. These species are reported to be endangered due to loss of habitat and degradation, opening of agricultural fields and genetic erosion. It is generally thought that the origin of this apple primarily comes from M. sieversii, also known as Central Asia wild apple. The second significant contribution is thought to be supplied by M. sylvestris. In this study, genetic diversity of 22 M. sieversii genotypes collected from Kyrgyzstan were investigated. Fifteen ISSR primers were evaluated and 78 fragments were obtained. A certain level of genetic diversity has been identified among the genotypes. All genotypes except two were distinguished from each other. This study has established that the M. sieversii genotypes contain significant variation and they should be protected.

*This study funded by Scientific Research Unit of Erciyes Uni¬versity with the project num¬ber of FOA-2014-5037.

Keywords: Central asia, endangered species, wild apple

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The inhibitory impact of *Enterococcus gallinarum* and *Lactobacillus plantarum* on bacterial growth and biogenic amine accumulation in fermented striped piggy

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Abstract

The influences of two lactic acid bacteria (LAB) strains (*Enterococcus gallinarum* and *Lactobacillus plantarum*) on bacterial growth and chemical parameters (total volatile basic nitrogen-TVB-N and biogenic amine) in fermented striped piggy (*Pomadasys stridens*) inoculated with *Photobacterium phosphoreum* were investigated in ambient temperature. Fish were divided into three groups, control group without any LAB inoculation with brine solution (4% glucose and 3% NaCl), treated groups 1 and 2 with same brine solution inoculated with *E. gallinarum* and *Lb. plantarum* at doses of 1% (108 cfu/ml), respectively. Total viable count in fermented fish reached maximum level for control group (8.6 log cfu/g) at 6 days. Initial TVB-N content of fish was 15.39 mg/100 g and significantly increased after inoculation with *P. phosphoreum*. TVB-N content of fermented fish was the lowest by *Lb. plantarum* inoculation at 6 days. Histamine content in raw striped piggy was 2.56 mg/100 g and remained below 8 mg/100g in all groups during storage. Tyramine was main amine accumulated in fish meat during fermentation. The highest tyramine production was found for control and fish inoculated with *E. gallinarum*, with respective value of 1067.55 and 1036.10 mg/100 g at 4 days. *E. gallinarum* induced lower putrescine and cadaverine accumulation at 4 days, whilst fish inoculated with *Lb. plantarum* had the lowest putrescine and cadaverine content at 6 days. LAB inoculation also significantly reduced ammonia and TMA formation. LAB used, mainly *Lb. plantarum* seemed to have inhibitory effects on bacterial growth and some of the chemical parameters of fermented fish.

Keywords: *Enterococcus gallinarum, Lactobacillus plantarum, Photobacterium phosphoreum*, Biogenic amines, Fermentation

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Congress
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Cytotoxic effect of lover rim-functionalized calix[4]arene-based imidazole on the lung cancer cell line

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Abstract

Calixarenes, cyclic oligomers of phenolic units linked through the ortho positions, are a fascinating class of macrocycles. They are synthetic macrocycles readily available by condensation of p-tert-butylphenol with formaldehyde under alkaline conditions. They have generated considerable interest as useful building blocks for the synthesis of hosts for cations, anions and neutral molecules. Due to this ability to form host–guest type complexes with a variety of organic or inorganic compounds, the calixarenes have received increasing attention during the last two decades. The increasing interest in these compounds is stimulated by the simple large-scale synthesis of calixarenes, and the different ways in which they can be easily functionalized both at the phenolic OH groups (lower rim) and, after partial removal of tert-butyl groups, at the para positions of the phenol rings (upper rim). Calixarenes find applications as selective binders and carriers, as analytical sensors, as catalysts and as model structures for biomimetic studies. Especially in recent years, it has been investigated whether calixarenes can be used as drug-solubilizing agents, either as anti-cancer reactivities or as an enhancement of controlled release and water solubility of drugs via host-guest type complexation. The aim of this project is to investigate the anticarcinogenic effects of the compound imidazole-derived calix[4]arene [C-I] on human lung cells. The cytotoxic effect of [C-I] on the A-549 cell line from human lung cancer cells was evaluated using the Alamar blue test from spectrophotometric methods. The A-549 cells were treated with [C-I] at different concentrations (1-100 μM) after reaching 80% density in DMEM growth medium and the IC50 value was calculated as 31.5 μM, calculated from the sigmoidal plot resulting from the inhibitory effect on the growth curve. The compound [C-I] demonstrates that high antiproliferative effect on the cell lines.

Keywords: Calixarene, cytotoxic effect, anticarcinogenic drugs

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Pinpointing the importance of compounds in human metabolic network

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Abstract

The methods like Degree Centrality, Betweenness Centrality and Closeness Centrality provide superficial information about structure of the complex networks, moreover they are impractical for detecting important nodes. Although there are various approaches which give deep knowledge to measure the significance of nodes by combining fundamental network topology parameters such as clustering coefficient and node neighbourhoods, they fail to properly identify bridge nodes. L-value is a recently developed measure which can detect significance of nodes in complex networks based on not only local information but also considering the importance of bridge nodes. Proposed approach considers total number of triangles in network, degree of nodes and their neighbours. In light of this, we implemented aforementioned L-value method in human metabolic network to reveal significant nodes, i.e. critical compounds. Our findings has potential to provide novel and insightful aspect on gene expression profile analysis. By our method, DEG lists derived from RNA-Seq or microarray studies can be revisited in terms of their impact on key compounds.

Keywords: Metabolic network, Network topology, DEG, RNA-Seq

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Evaluation of in vitro antiviral activity of *Agaricus blazei* murril against human respiratory syncytial virus

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Abstract

This study was conducted to investigate the anti-RSV activity of crude methanol and aqueous extracts obtained from *Agaricus blazei* Murrill which is used for medicinal purpose in the World, especially in Asian countries. Extracts were tested by means of the colorimetric XTT assay. The EC50 was defined as the concentration required to achieve 50% protection against virus-induced cytopathic effects, and the selectivity index (SI) was determined as the ratio of CC50 (concentration of 50% cellular toxicity) to EC50. Results showed that methanol extract of *Agaricus blazei* (EC50: 5692.31 µg/mL and SI: 3.81) and its aqueous extract (EC50 = 4433.28 µg/mL and SI = 10.85) had anti-RSV activity in comparable rates to ribavirin (EC50 = 4.19 µg/mL, SI = 27.92) used as a positive control against RSV. The cell cytotoxicity test showed that both of the extracts tested had higher CC50 values than the EC50 values. As a result, we can say that both extracts deserve further study in order to be developed as an alternative to ribavirin, which is commonly used clinically against RSV. This is the first report on the anti-RSV activity of *A. blazei*.

*This research is a part of Hadeel Abduljabbar Abbas Al-Mafrachi's Master Thesis.

Keywords: *Agaricus blazei*, methanolic extract, aqueous extract, anti-RSV activity

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Isolation and molecular characterization of thermophilic bacteria with potency to produce enzymes which are ındustrially ımportant

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Abstract

Thermophilic organisms are the only group of microorganisms which adapt to live at high temperatures and have tolerance to extreme temperatures. Thermophilic microorganisms are naturally ocurring in hot springs, feces and in garbages. Enzymes obtained from the microorganisms resistant to extreme conditions are preferred more because they are used for a longer period of time due to their high catalytic activity. Within the scope of study, water and sludge samples were taken from the thermal facilities and isolation and identification of thermophilic bacteria groups were carried out. The bacteria identified by conventional methods were then identified by genotypic methods. For this purpose, 16S rRNA-PCR analysis was used and the bacteria were identified. Afterwards, genotypic profiling of the bacteria was carried out by rep-PCR and genotypic similarities with each other were designated. According to the sequencing results, bacteria belonging to *Bacillus licheniformis, B. paralicheniformis, Paenibacillus dentritiformis, Aeribacillus pallidus, Anoxybacillus geothermalis* species with a similarity rate of 99 % were detected in 30 samples. After obtaining the sequence data, a phylogenetic tree was drawn to determine the phylogenetic affinity of the bacteria with each other by using the MEGA Clustal W program. Subsequently, bacteria strains producing industrial essential enzymes (lipase, amylase and xylanase) were identified and it was detected that bacteria of *Bacillus lichenifırmis*, *B. paralicheniformis, Anoxybacillus geothermalis* species would be important producers of these enzymes.

Keywords: Thermophilic Bacteria, Isolation, Identification, Thermostabile Enzyme

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Preparation of nanohybrid pectin lyase (PL) from *Bacillus licheniformis* and it's clarification of fruit juices

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Abstract

Pectin lyases are widely used in many industries, especially in fruit juice industry. In this study, isolated and identification of *Bacillus licheniformis* bacteria from tomato samples using chemical and molecular methods. Chemical identification was determined by gram staining and catalase tests. Sequence analysis of 16S rRNA gene and 16S-23S rDNA intergenic spacer regions (ISR) was performed for molecular identification of the isolated bacterium. PL enzyme, which was developed in a solid-culture fermentation medium, was produced from identified bacterial and was firstly purified using three phase partitioning method (TPP). This enzyme, Chitosan / Calcium Pyrphosphate hybrid nanoflower structure was prepared. Vmax and KM values of the free and nanoflower PL enzyme were determined by using Lineweaver-Burk method for pectin, locust bean gum, chitin, substrates. Also, the effects of Ca^{2+}, Cu^{2+}, Fe^{3+}, Mg^{2+} , Zn^{2+} and Hg^{2+} metal ions on pectin lyase enzyme activities were determined. Free and nanohybrid pectin lyase enzymes was used for clarification of some fruit juices such as grapes (black), pomegranate, cranberry. It was determined that the nanohybrid PL enzyme was extremelly effectively in the yield and clarification of fruit juice compared to the control sample.

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Keywords: Pectin lyase, *Bacillus licheniformis*, Three phase partitioning method, Nanoflower

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Effects of *Plagiomnium undulatum* on protein amount and antioxidant enzyme activities of wild mustard and wild oat

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Abstract

Wild oats (*Avena sterilis* L.) and wild mustard (*Sinapis arvensis* L.) are quite common in agricultural areas of our country. Biological control of such weeds is becoming increasingly important. It was aimed to determine the effect of the extracts of *Plagiomnium undulatum* (Hedw.) T.J.Kop. at different concentrations (0, 25, 50 mg. mL^{-1}) in different solvents (distilled water, ethanol, ethyl acetate) on total protein amount and antioxidant enzyme activities (SOD, PO and CAT) of *A. sterilis* and *S. Arvensis*. In total protein amount, reduction in 50 mg . mL-1 dH2O, 50 mg . mL^{-1} ethanol and 25 mg . mL^{-1} ethyl acetate treatments, increase in the others in wild mustard, decrease in 50 mg . mL^{-1} ethanol, treatments ($p<0.05$), increase in other treatments in wild oat to the control was detected. SOD activities reduced in all treatments in wild mustard ($p<0.05$), increased in ethanol, ethyl acetate and 50 mg. mL^{-1} ethanol treatments, but decreased in other treatments ($p<0.05$) in wild oat to the control. PO activity decreased in all groups except 25 mg . mL^{-1} ethyl acetate treatment in wild mustard to the control. In oats, an increase was detected in all treatment groups. CAT activity increase in the treatment of 50 mg . mL^{-1} dH2O, 50 mg . mL^{-1} ethanol, 25 mg . mL^{-1} ethyl acetate, decrease in the other groups in *S. arvensis*, while increase in all groups in at significance level in *A. sterilis*. In conclusion, it has been determined that *P. undulatum* extracts affect wild mustard and wild oat plants. Additional studies are required to be use it as organic herbicides.

Keywords: *Avena sterilis*, CAT, PO, SOD, *Sinapis arvensis*

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Preparation, characterization, anti-microbial, DNA binding and DNA cleavage studies of nitrogen doped graphene quantum dots (GQDs)

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Abstract

Graphene quantum dots (GQDs) as a new species nano materials have attracted great interest in recent years due to their very interesting features. GQDs have potential for use in biomedical applications because they have a smaller nano size and higher biocompatibility. Studies on biological activity and biomedical applications is quite low. It focuses more on the biosensor applications. In in this study, graphene quantum dots (GQDs) containing N atoms were synthesized using hydrothermal reaction of citric acid and 4-aminophenole. Nano materials were characterized by UV-Vis, FT-IR spectroscopy, transmission electronmicroscopy (TEM) and thermogravimetric analysis. The antimicrobial activity of the compound was investigated for its minimum inhibitory concentration (MIC) to bacteria and yeast cultures. Surprisingly, UV-Vis spectroscopy studies of the interactions between the GQDs and calf thymus DNA (CT-DNA) showed that the compound interacts with CT-DNA via both intercalative and electrostatic binding. DNA cleavage study showed that the GQDs cleaved DNA without any external agents.

Keywords: DNA binding, DNA cleavage, Graphene quantum dots (GQDs), UV-Vis

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