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# Molecular differentiation of Polish and Georgian strains of *Clavibacter michiganensis* subsp. *sepedonicus*

**Agnieszka Maciejewska**
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*Clavibacter michiganensis* subsp. *sepedonicus* (Cms) causes a dangerous bacterial disease called ring rot of potato. This disease is of quarantine status in the European Union, as the cause of loss is its easiness to spread. It means, this disease is controlled by national regulations and there is zero tolerance for this pathogen in the potato crops. This bacterium generates a huge threat for seed material and breeding production and it hinders trading of potato in the EU and other countries. Lack of sufficiently sensitive methods of detection and identification it leads to poor elimination of this pathogen. Up to now, the routine detection techniques, such as immunofluorescence test (IF) and immunofluorescence assay (IFAS) by using poly and monoclonal antibodies are not sensitive enough. Therefore, still the rapid methods of detection and identification of Cms are searched. The information about diversity and structure of Cms populations are indispensable for the development of the most effective methods for detection and eradication of this pathogen. However, the present knowledge about the diversity of strains is very poor. The differentiation of Cms strains would help to define the quarantine risk and devise effective methods for control of ring rot. Analyses observed that the MP-PCR technique showed the genetic diversity among tested isolates of Cms bacteria both in Polish and Georgian. The MP-PCR technique showed the differences between the genomes of isolates Cms which were detected in a reproducible way, therefore this method is widely used in genotyping organisms.

## Biography

Agnieszka Maciejewska is currently a PhD student and works at the Plant Breeding and Acclimatization Institute in Department of Plant Pathology. Her research is connected with quarantine pathogen *Clavibacter michiganensis* subsp. *sepedonicus* of potatoes. She was responsible for the biochemical and molecular characterization genetic variability population of *Clavibacter michiganensis* subsp. *Sepedonicus*.

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## Optimization of regeneration and *gus* gene transferring in Kalanchoe blossfeldiana R.

**Maryam Mehdizadeh Hakkak, Nasrin Moshtaghi and Abdolreza Bagheri**
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Kalanchoe blossfeldiana R. is a pot plant with red, pink and white flowers and fleshy leaves. In this study, the effect of plant growth regulators were investigated on plant regeneration and then gene transferring was optimized. In the first experiment, three types of explants: Leaf, veins and petioles were used for regeneration and different concentrations of BA and Kin (0, 0.5, 1.5 and 3 mg/l) alone or in combination with NAA (0, 0.2 and 0.7 mg/l) were used for regeneration. Results showed that the highest number of shoots (47.33) and leaves (330.33) were obtained on MS medium supplemented with 1.5 mg/l BA and 0.7 mg/l NAA. Maximum length of shoots (1.7 cm) was obtained from petiole explants on MS medium supplemented with 1.5 mg/l BA and 0.7 mg/l NAA. Regeneration rate was 100% in all treatments, but it was 0% in the medium without growth regulators and the medium without BA. Also in medium containing Kin, adventitious root regeneration obtained from leaf explants. Different media: MS, ½ MS, ½ MS with 1 mg/l IAA and ½ MS with 1 mg/l IBA were used for rooting. The most number of roots (21/12) and root length (1.56 cm) were obtained in ½ MS supplemented with 1 mg/l IBA. For acclimation, different substrates such as coco peat, peat moss, coco peat- peat moss and coco peat-perlite were used. The most length increasing percentage (cm) and leaf number increasing percentage (cm) were obtained in peat moss substrate. In the second experiment, optimizing *gus* gene transferring was done by Agrobacterium tumefaciens strain LBA4404. Leaf sections and stems of *in vitro* plantlets were used as explants for co-culturing in 10 and 30 minutes by *Agrobacterium tumefaciens* containing *gus* gene. The highest percentage of gene transferring and its expression (38.46%) was observed in leaf explants by 10 minutes co-culturing. So we can use from this protocol for transferring the useful and interest genes to this plant.

### Biography

Maryam Mehdizadeh Hakkak has completed her Bachelor's degree in Plant and MSc in Plant Biotechnology at Ferdowsi University of Mashhad in 2012 and 2015 respectively.

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# Exploration and Purification of Bioactive compound from seaweeds against human bacterial pathogen

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Treatment of Infectious diseases by the use of commercially available drugs are becoming certain limitations due to changing patterns of resistance in pathogens and causing side effects. These limitations demand for improved pharmacokinetic properties which necessitates continued research for the search of new novel drugs. Marine organisms are rich source of structurally novel and biologically active metabolites. The cell extracts and active constituents of various algae and seaweeds have been shown to have antibacterial activity against gram positive and gram negative bacteria. Hence the crude extracts from the seaweeds *Amphiroa foliacea, Chactomorpho tortuosa, Caulerpa scalpelliformis* and *Sargassum sp* were tested for their resistance against multidrug resistance pathogens such as *Staphylococcus aureus, Klebsella sp, Proteus sp*. The extracts were obtained with the solvents methanol, chloroform, ethyl acetate and hexane. A highest zone of inhibition was observed in the hexane extract of *Amphiroa foliacea* and ethyl acetate extract of *Sargassam sp against Proteus sp, Staphylococcus aureus* respectively. An inhibition zone of 8mm was observed in ethyl acetate extract of *Sargassam* sp against *Staphylococcus aureus*. Further the extract of *Sargassum* sp was purified using silica column chromatography. Single compound fractions were separated and each fraction was screened for antibacterial activity against *Staphylococcus aureus*. F4 fraction possessed antibacterial activity of 7mm which is similar to crude extract. Further the F4 fraction is subjected NMR analysis. Ethyl acetate fraction was found to be possess α-hydroxy stearic acid.

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# Compensatory effects of *hOGG1* for *hMTH1* in oxidative DNA damage caused by hydrogen peroxide

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This study aimed to investigate the potential compensatory effects of *hOGG1* and *hMTH1* in the repair of oxidative DNA damage. The *hOGG1* and *hMTH1* gene knockdown human embryonic pulmonary fibroblast cell lines were established by Lentivirus-mediated RNA interference. The messenger RNA (mRNA) levels of *hOGG1* and hM1TH1 were analyzed by the real-time polymerase chain reaction and 8-hydroxy-20-deoxyguanosine (8-oxo-dG) formation was analyzed in a high-performance liquid chromatography-electrochemical detection system. The *hOGG1* and *hMTH1* knockdown cells were obtained through blasticidin selection. After transfection of *hOGG1* and *hMTH1* small interfering RNA, the expression levels of the mRNA of *hOGG1* and *hMTH1* genes were decreased by 97.2% and 96.2%, respectively. The cells then were exposed to 100 mmol/L of hydrogen peroxide ($H_2O_2$) for 12 hours to induce oxidative DNA damage. After $H_2O_2$ exposure, *hMTH1* mRNA levels were increased by 25% in *hOGG1* gene knockdown cells, whereas *hOGG1* mRNA levels were increased by 52% in *hMTH1* gene knockdown cells. Following the treatment with $H_2O_2$, the 8-oxo-dG levels in the DNA of *hOGG1* gene knockdown cells were 3.1-fold higher than those in untreated HFL cells and 1.67-fold higher than those in $H_2O_2$-treated wild-type cells. The 8-oxo-dG levels in *hMTH1* gene knockdown cells were 2.3-fold higher than those in untreated human embryonic pulmonary fibroblast cells but did not differ significantly from those in $H_2O_2$-treated wild-type cells. Our data suggested that *hOGG1* could compensate for *hMTH1* during oxidative DNA damage caused by $H_2O_2$, whereas *hMTH1* could not compensate sufficiently for *hOGG1* during the process.

## Biography

Yuebin Ke is a Professor of the Shenzhen Center for Disease Control and Prevention and an Adjunct Professor of Life Sciences at Shenzhen University. He has completed his PhD from Huazhong University of Science and Technology and Postdoctoral studies from Virginia Polytechnic Institute and State University. He has published 18 papers in the areas of environmental health and molecular biology.

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# Molecular study of intron 2 of calreticulin gene (CALR) in type-2 diabetic patients

**Negin Parsamanesh, Hosein Javdani, Ali Farahi and Maryam Valavi**
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Type 2 diabetes mellitus (T2DM) is a complex polygenic disease. Genetic factors play major role in the pathogenesis of T2DM. Calreticulin is a 46 kD $Ca^{2+}$ binding protein and ER chaperon. Calreticulin (CALR) was found in high concentration in pancreas. Genomic analysis and detection of variants related to type-2 diabetes can help to determination T2DM pathophysiology and its familial pattern of inheritance. Imbalance in $Ca^{2+}$ concentration and dysfunction of the chaperone system are speculated to be linked with type-2 diabetes mellitus (T2DM). Two-dimensional protein profiling of pancreatic beta cells in T2DM subjects has shown that the Ca2+ binding chaperone, calreticulin (CALR), plays a role in the pathophysiology of this disease. In a case/control study design, we performed mutation screening of the promoter region, 9 exons and exon/intron boundaries of CALR by PCR-SSCP and sequencing in 120 patients afflicted with T2DM and 530 controls. Two novel mutations were detected in T2DM patients, which were absent in the control gene pool (Mid P exact <0.01). The first mutation was a G>T transversion in intron 2 conserved polypurine tract at IVSII-142. The second mutation was a 9-bp deletion in the highly conserved exon 9 encompassing amino acids 402-404. Exon 9 encodes the low affinity, high capacity $Ca^{2+}$ binding domain of CALR. This case is the first instance of a microdeletion in a gene coding sequence reported in T2DM. To our knowledge, the current study reports for the first time, CALR gene mutations that co-occur with T2DM.

## Biography

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# Hepatitis C virus core gene polymorphism in cases of hepatocellular carcinoma

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**Introduction:** Hepatocellular carcinoma (HCC) is one of the common sequelae of hepatitis C virus (HCV) infection. It remains controversial, however, whether HCV itself plays a direct role in the development of HCC. Although HCV core protein was reported to display tumorigenic activities in cell culture and experimental animal systems, its clinical impact on HCC development in humans is still unclear.

**Aim:** We mapped sequence differences in the viral core gene which is strongly implicated in cellular transformation and the development of liver cancer to test the hypothesis that core gene sequences from HCC patients differ from those of patients without HCC.

**Methods:** HCV core sequences from HCC patients and controls were obtained and compared with each other. A logistic regression model was developed to predict the HCC risk of individual mutations and other sequence features.

**Results:** Study showed that sequences of HCV in patients with hepatocellular carcinoma differ from those of patients with early-stage liver disease. One polymorphism was particularly strongly associated with liver cancer. Specifically, core amino acid position 71 was present in 33.3% of the full length sequences from patients with HCC but only 6.7% of patients without HCC. Multivariate analysis identified core amino acid polymorphism, elevated α-fetoprotein (AFP) levels, elevated ALT level, elevated alkaline phosphatase level and liver fibrosis as independent factors associated with HCC.

**Conclusions:** HCV core genes from patients with and without HCC differ at several positions. Our findings suggest that HCV core gene sequence data might provide useful information about HCC risk. Prospective investigation is needed to establish the temporal relationship between appearance of the viral mutations and development of HCC.

## Biography

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# Accepted Abstracts

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## First-principles study of siloxene and germoxene: Stable conformations, electronic properties and defects

**Apostolos Atsalakis**
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Recent interest in two-dimensional (2D) forms of Si and Ge has surged recently with a focus on silicene and germanene, the Si and Ge-based analogues of graphene as well as their derivatives. Siloxene and germoxene are 2D materials made of honeycomb Si and Ge backbone sheets that are decorated with H atoms and OH groups. This work uses first-principles calculations to probe the properties of their various conformations. It is shown that the most stable siloxene (and germoxene) polymorph is the so-called washboard structure and not the chair geometry assumed in previous studies. The stability of the washboard configuration relates to the formation of a network of hydrogen bonds between its hydroxyl groups. It is also found with hybrid functional calculations that siloxene and germoxene are wide band-gap semiconductors with gap values of 3.20 eV and 2.64 eV, respectively. Finally, we show that H and OH vacancies introduce spin polarization in these 2D materials and have a tendency to pair up in stable di-vacancies.

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## Evaluation of circulatory RNA based biomarker panel in hepatocellular carcinoma

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The circulating transcriptome (coding and non-coding) plays a critical role in cancer and novel accurate strategies for early detection of hepatocellular carcinoma (HCC) are strongly needed. We chose a HCC-specific RNA based biomarker panel based on the integration of differential lysosomal associated membrane protein 2 (LAMP2) gene expressions with its selected epigenetic regulators using bioinformatic methods. This was followed by RT-qPCR validation in serum of 78 patients with HCC, 36 patients with chronic hepatitis C (CHC) infection and 44 healthy volunteers. We used risk score analysis to evaluate the diagnostic efficacy of the serum profiling system. Moreover, in 20 of the 78 HCC cases involved in the study; we examined the expression of RNA based biomarker panel in both HCC and adjacent non-tumor tissues and assessed their correlation with the serum level of this panel. The 4 RNA based biomarker panel [long non-coding RNA-C terminal binding protein, androgen responsive (lncRNA-CTBP), microRNA-16-2 (miR-16-2), microRNA-21-5-P (miR-21-5p) and LAMP2 had high sensitivity and specificity for discriminating HCC from healthy controls and also from CHC patients. Among these 4 RNAs serum miR-16-2 and miR-21-5p were independent prognostic factors. The circulatory RNA based biomarker panel can serve as a potential biomarker for HCC diagnosis and prognosis.

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## Synthesis and application of novel tricyanofuran hydrazone dyes as sensors for detection of microbes

**Hatem Elsayed Mohamed Hussien Gaafar**
National Research Centre, Egypt

Acknowledging the need to develop rapid and sensitive bacterial recognition approaches, we functionalized the tricyanofuran hydrazone molecular switch. Of significant interest in relation to the synthesized hydrazones is the formation of two different conjugated structures upon exposure to different pH values. Many bacteria release ammonia gas, which alkalizes environments. Herein we report the synthesis of a tricyanofuran hydrazone having the function of a colorimetric pH sensor. The UV-visible absorption and fluorescence spectra exhibit reversible color changes of the tricyanofuran hydrazone solution in acetonitrile under acid-base conditions. Our results indicate that the tricyanofuran hydrazone probe can identify the bacterial targets quickly with high sensitivity. The infected samples exhibit a significant color change from orange to blue and in the mean time there is a decrease in fluorescence emission as a function of ammonia and volatile amines released from bacterial metabolites. This tricyanofuran hydrazone chromophore is proposed for use in food packaging with a pH-sensing capability.

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## Alpha cyano-4-hydroxy-3-methoxycinnamic acid inhibits proliferation and induces apoptosis in human breast cancer cells

**Lamia Hamdan Ramdani[1], Zoheir Arrarz[2], Yacoub Al Muataz[2], Lutfi Suleiman[2]**
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Breast cancer is the leading cause of cancer death in women worldwide and a critical public health concern. In this context, the present work has examined the in vitro and in vivo antiproliferative and pro-apoptotic effects of α-cyano-4-hydroxy-3-methoxycinnamic acid ACCA on human breast cancer cell lines, MDA-MB-231, MCF-7 and T47D. Treatment with ACCA resulted in dose and time-dependent decrease of cell proliferation, viability in colony formation assay and in induction of programmed cell death (apoptosis) with minimal effects on non-tumoral cells. The ability of ACCA to suppress growth in cancer cells not expressing or containing defects in p53 gene indicates a lack of involvement of this critical tumor suppressor element in mediating ACCA-induced growth inhibition. The stimulation of breast cancer cells with ACCA would increase the expression of the ratio of Bax to Bcl-2, a process widely involved in the stimulation of apoptosis. Besides, we have demonstrated the ability of ACCA to inhibit the migration and invasion of MDA-MB231 cells. Additionally, tumor growth of MDA-MB-231 breast cancer cells was dramatically affected in vivo by ACCA. Therefore, these results show that ACCA might be very promising therapeutic targets in the treatment of breast cancer.

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## Amyloid-like protein membrane: A natural polymer based biosensing material

**Gozde Kabay**
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Electrospinning has been a popular technique to obtain nanofibrous membranes from synthetic and natural polymer sources. In contrary to synthetic polymers, the production capabilities of natural polymer membranes failed at some extend by means of electrospinnability and efficiency also biocompatibility. In this paper, we introduced an enzyme immobilization platform from a natural polymer membrane. For this purpose, a model protein bovine serum albumin (BSA) was chosen mainly for enhanced supporting property. To procure electrospinnable solution of BSA, beta-mercaptoethanol (β-ME) was used to induce tertiary structure and low ratio (1.5:1.0 TFE:PBS (pH: 7.4)) of 2,2,2-triflouroethanol (TFE) was added as a stabilizing agent, respectively. The electrospun membranes were activated with RF plasma treatment by employing ethylenediamine (EDA) as a precursor to incorporate amino (-NH2) groups on the surface. Those surfaces were cross-linked with glutaraldehyde aqueous solutions at concentrations between 0.01 and 5% wt. which followed by the covalent attachment of glucose oxidase (GOD). The performance of enzyme immobilized membranes was tested by employing amperometric measurements against various glucose concentrations in terms of response time, enzymatic activity and linearity. The effects of plasma parameters and cross-linking conditions on the performance of protein membrane based enzyme electrode were also studied.

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## Phenotypic and genotypic characterization of *Staphylococcus aureus* strains from some food stuff of animal origin

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Methicillin resistant *Staphylococcus aureus* (MRSA) is one of the most important public health problems in many countries. In recent years, the existence of MRSA in foodstuff of animal origin and its transfer among farm animals, foodstuff of animal origin and human beings have been shown with molecular typing studies. The objective of this study was to investigate existence, methicillin resistance (MR) and clonal relationship of *Staphylococcus aureus* (*S. aureus*) strains from foodstuff of animal origin consumed in the Samsun region of Turkey. In this study, a total 175 coagulase positive staphylococci (CPS) strains were isolated from meat (n=110), milk (n=56) and fishery products (n=9). From these, 62 *S. aureus* strains were identified from meat (n=44), milk (n=9) and fishery products (n=9). Identification and MR properties of the isolates were confirmed by PCR technique in which appropriate primers for *nuc* and *mecA* gene were used. For detection of MR, we also used minimal inhibitory concentration (MIC) technique. We compared two techniques; although 21 isolates were determined as MRCPS using MIC (≥12 μg), 18 isolates were detected MRCPS using PCR assay. Among these, 15 isolates were identified as MRSA using PCR technique. We investigated only MRSA isolates for the clonal relationship using PFGE method. PFGE typing of the 15 MRSA strains yielded 6 PFGE patterns. Pattern A and E were found to be dominant types in our study. Pattern E consisting of 7 strains was from fishery products. Pattern A consisting of 4 strains was from meat and fishery products. Patterns B, C, D and F were single isolates from milk, meat and milk products, respectively.

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## Aroma compounds in cooperage oak wood (*Quercus pyrenaica* Willd.): Effect of site and silvicultural parameters

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Aging wines is a long-used technical step. This praxis was set up for the storage, but later, it has been used because of its unique effects on wine organoleptic properties. The role of wood during this process is crucial. From the sensorial point of view, wood is capable of transmitting aroma-responsible volatile compounds. Also, a reduction of astringency and color changes are produced as a result of the phenolic compounds extraction. Different woods have been used in cooperage (chestnut, cherry-tree); however oak is the most common for its chemical composition and for both its mechanical and physical properties. But, the aging carried out by means of barrels entails a time-consuming and expensive practice. On the one hand, aged wines have to be left in the barrels during a long period before they can be brought to market because of the slow extraction process of aroma compounds. On the other hand, it implies some problems, such as their sanitization and handling. The volatile composition of *Quercus pyrenaica* from NW Spain were analyzed on a wide sample set of more than 100. The relationship between some silvicultural and site parameters and volatile composition was studied. Altitude appeared to be the most significant parameter. However, other factors such as annual precipitation and number of trees per hectare whose effects on the volatile compounds were not significant. The influence of geographical location seemed to have a more specific impact. The content of extractable compounds permitted a separation of samples according to their origin.

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## Moving away from non-uniform biologicals in healthcare: Development of synthetic embryo culture and sperm cryopreservation media solutions

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Embryo Culture (ECM), Cell, Gamete and Embryo Cryoprotectant (CM), Stem Cell Culture (SCCM), Cell-based Vaccine Production Media (VPM), etc., contain donor serum proteins (DSPs) which carry risk of disease transmission to patients/their babies/healthcare workers. The European Union recommends avoidance of non-uniform biologicals in healthcare products (EU Tissue Directive No.2004/23/EU) by April 2007. Most manufacturers of healthcare products have not fully complied with this directive. Available embryo culture media (ECM) for human ART supplemented with human serum albumin (HSA) contain contaminants such as hazardous pathogenic agents, micro DNA/RNA strands and undeclared proteins, all of which has the potential to cause adverse events for the baby including epigenetic effects, possible genetic crossovers with embryonic genome affecting the genetic constitution of the embryo and batch variation in quality of media. Late onset adverse events cannot be ruled out. The author developed synthetic human embryo culture media (Synbios™) devoid of DSPs. A clinical trial was performed successfully and patented in USA (US Patent 8415094)/PCT protected in Canada, EU, Australia, Russia, Israel and many nations worldwide. A synthetic spermatozoa cryopreservation medium (SCM) has also been developed and successfully applied with pregnancies achieved. More efforts are needed to develop culture media for the stem cell and vaccine industries. Efficacious synthetic ECM and SCM have been developed which eliminates disease transmission, is safe and even culturally acceptable as it was certified "Halal" or permissible by the EU Halal authority. It is anticipated to comply with regulations.

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## Optical processing for the analysis of genetic data sequencing

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An optical image processing technique can be proposed for analyzing and exploring DNA sequences. The approach uses the powerful computing capability of optical correlation to provide real-time processing of the genomic data. Instead of representing the symbolic DNA sequences in numeric form, they are converted and treated as 2-D images for matching purposes. Both images of the reference and the target DNA sequences are combined and presented as a joint input image to an optical joint transform correlator (JTC) set-up for a real-time processing. This JTC-based approach is capable to search for similarity/dissimilarity between two tested DNA sequences. The optical approach can facilitates the exhaustive search algorithms for locally and/or globally DNA alignment. Simulations experimental results on actual DNA sequences will be presented to demonstrate the effectiveness of the proposed optical approach.

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## Identification and verification of QTL associated with frost tolerance using linkage mapping and GWAS in winter faba bean

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Frost stress is one of abiotic stresses, which cause a significant reduction in winter faba bean yield in Europe. The main objective of this work is to genetically improve frost tolerance in winter faba bean by identifying and validating QTL associated with frost tolerance to be used in marker-assisted selection. Two different genetic backgrounds were used a biparental population (BPP) consisting of 101 inbred lines and 189 genotypes from single seed descent (SSD) from the Gottingen Winter Bean Population (GWBP). All experiments were conducted in a frost growth chamber under controlled conditions. Both populations were genotyped using the same set of 189 SNP markers. Visual scoring for frost stress symptoms was used to define frost tolerance in both populations. In addition, leaf fatty acid composition (FAC) and proline content were analyzed in BPP as physiological traits. QTL mapping (for PBB) and genome wide association studies (for GWBP) were performed to detect QTL associated with frost tolerance. High genetic variation between genotypes and heritability estimates were found for all traits. QTL mapping and GWAS identified new putative QTL associated with promising frost tolerance and related traits. A set of common 54 SNP markers in both two different genetic backgrounds showed a high genetic diversity with polymorphic information content ranged from 0.31 to 0.37 and gene diversity ranged from 0.39 to 0.50, indicating that these markers could be used for genotyping any faba bean population. Five SNP markers showed a significant marker-trait association with frost tolerance and related traits in both populations. Moreover, synteny analysis between *Medicago truncatula* (model legume) and faba bean genomes was performed to identify candidate genes of these markers. Collinearity was evaluated between the faba bean genetic map constructed in this study and the faba bean consensus map, resulting in identifying possible genomic regions in faba bean which may control frost tolerance genes. The two genetic backgrounds were useful in detecting new variation to improve frost tolerance in winter faba bean. Of the five validated SNP markers, one (VF_Mt3g086600) was found to be associated with frost tolerance and FAC in both populations. This marker was also associated with winter hardiness and high yield in earlier studies. This marker is located in a gene of unknown function.

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## Bacteriophage-antibiotic synergism to control planktonic and biofilm producing clinical isolates of *Pseudomonas aeruginosa*

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**Introduction:** Pseudomonas aeruginosa is a highly resistant opportunistic pathogen and is capable of forming biofilms on medical devices. Bacterial biofilms, which are micro-colonies encased in extracellular polysaccharide material are so difficult to be treated by conventional antibiotics. During the last decade, *P. aeruginosa* phages have been extensively examined as an alternative to antimicrobial agents.

**Aim:** The aim of the study was to assess bacteriophage-antibiotic combination on planktonic and biofilm states of *P. aeruginosa* isolates.

**Materials:** In this study, we isolated 6 lytic phages from hospital effluents; they were tested against 50 *P. aeruginosa* strains, isolated from different clinical specimens delivered to the Diagnostic Microbiology Laboratories, Faculty of Medicine, Alexandria University.

**Results:** Out of the 50 isolates, 15 were susceptible to these phages. So the biofilm forming capacity of these 15 isolates was investigated. The results showed that 14 isolates (93.33%) produced detectable biofilm. The minimum inhibitory concentration (MIC) and minimum biofilm eradication concentration (MBEC) assays were used to evaluate the antibiotic sensitivity patterns of these *P. aeruginosa* isolates in their planktonic and biofilm phases to amikacin and meropenem. Also, the effects of phage on the planktonic and biofilm states of isolates at different multiplicities of infections (MOI) were tested. On the planktonic state, Amikacin-phage combination showed synergistic effect (P=0.001) and Meropenem-phage combination showed synergistic effect (P=0.003). On the biofilm state, Amikacin-phage combination showed biofilm eradication in 50% of the isolates (P=0.003). On the other hand, Meropenem-phage combination showed biofilm eradication in only 14.3% of the strains.

**Conclusion:** The combination of phage and antibiotics could have potentially more benefits on *P. aeruginosa* planktonic and biofilm states than just using phages or antibiotics alone.

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## The art of mathematical biology

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Mathematical biology is a fast-growing, well-recognized subject and one of the most exciting modern applications of mathematics. The increasing use of mathematics in biology is inevitable as biology becomes more quantitative. The complexity of the biological sciences promoted interdisciplinary involvement. For the mathematician, biology opens up new and exciting branches, while for the biologist, mathematical modeling offers another research tool commensurate with a new powerful laboratory technique but only if used appropriately and its limitations recognized. Currently, it seems that theoretical/mathematical biology offers lots of promising perspectives and possibilities for mathematicians and theoretically interested biologists. Mathematical biology has been successfully applied in many fields as for example; cancer detection, antiretroviral therapy including the integrase inhibitor Raltegravir in HIV-1 patients and molecular dynamics.

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## Effect of newly synthesized progesterone derivatives on apoptotic and metastatic pathway in MCF-7 breast cancer cells

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**Background:** Breast cancer is the second leading cause of mortality among women worldwide. Anticancer agents consisting of hybrid molecules are used to improve efficacy and reduce drug resistance. Alteration of different genes is involved in the development of cancer. Consequently, novel anticancer drugs with increased selectivity and specificity are required to overcome limitation of current drugs. A variety of synthetic steroid derivatives have been contrived, most these derivatives can interact with the steroid receptors because of a similarity of shape. Also, the investigation of modified steroid derivatives condensed with various heterocyclic rings has a great attention. Impaired apoptosis and metastasis are critical in cancer development and is a major barrier to effective treatment.

**Methods:** Several progesterone derivatives were synthesized. The structure of the newly derivatives was elucidated and confirmed using the analytical and spectral data. The newly synthesized progesterone derivatives, compounds 1, 2, 3, 4, 5, 6 and 7 were tested for their cytotoxic effects against human breast cancer cells (MCF-7) using neutral red uptake assay. Using QRT-PCR (Quantitative Real Time-Polymerase Chain Reaction), the expression levels of *P53, P21, Cdc2, Bcl-2, Survivin, CCND1, VEGF, HIF-1α, FGF-1, MMP-2, MMP-9, Ang-1 and Ang-2* genes were investigated.

**Results:** All tested compounds showed low IC50 values that were comparable to that of tamoxifen. The most active compounds against MCF-7 cancer cell line was in the descending order of 5>1>2>6>4>7>3. The study revealed that all newly synthesized compounds down-regulated the expression levels of *BCL-2, surviving, VEGF, Ang-2* and *MMp-9*. Compound 2-7 down-regulated CCND1 gene expression, nevertheless, this was only significant in case of compounds 2, 3 and 6. However, *P53* were up-regulated by compounds 3. Moreover, compound 1 significantly down-regulated *MMP-2* and compound 3 and 7 significantly down-regulated *FGF-1*.

**Conclusion:** This study introduced promising pro-apoptotic and anti-metastatic anticancer agents acting through the regulation of key regulators of apoptosis, cell cycle and metastasis related genes.

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## Biologics to enhance current orthopedic procedures

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In orthopaedic surgery a transition to improve current orthopedic procedures is underway. Surgical techniques, instruments and implants have been greatly refined and improvements in imaging accuracy, namely magnetic resonance (MR) can now clearly identify pathology. The final frontier is to improve biology and hopefully healing. The use of biological materials to foster improved outcomes has been highlighted by major scientific breakthroughs of which have defined the critical pathways of healing and regeneration. The list includes blood-derived preparations, growth factors, bone marrow preparations and expanded stem cells. These biologic adjuncts can provide effective treatments during surgery or during the postoperative period. It is likely that biologic treatments will actively enhance many areas of orthopedic surgery to improve the healing capabilities of currently performed surgical procedures today and in the future. This review will systematically assess the peer-reviewed evidence based literature highlighting advances in both pre-clinical studies and clinical trials involving biological substances in the treatment of meniscus, ligament and articular cartilage surgical repair.

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