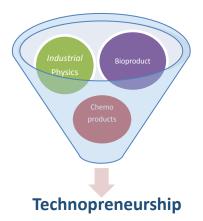
BIO-PHYSICOCHEMICAL BASIS FOR TECHNOPRENEURSHIP

2ND-3RD APRIL 2013

ABSTRACT BOOK





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Abstracts

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3rd April-Day2: BPC 46 -97

PL-01

Drug Discovery and Pharmaceutical

Prof. Dr. Muhammad Mukhtar
Vice Chancellor
Islamia University, Bahawalpur, Pakistan.

PL-02

"Building a Brand" & Management

Prof. Dr. Muhammad Ehsan Malik
Director, Institute of Business Administration,
University of the Punjab, Lahore, Pakistan.

Abstract:

Building and sustaining brand is an effective tool of the Managers for the long lasting progress of a company. The new products are the life blood of a company which, come into existence through its dedicated research & development (R&D). The successful launching of a new product mainly depends upon the efficient corporate branding. So, company is supposed to protect its brand because the brands are more than just names and symbols. A company must consider expense on brand development as an investment, not a cost because brand loyalty never just happen rather brand managers have to make it happen. Hence, the Brand may be built by market research, responsiveness to change, flexibility, vision and reaction to change.

Brand development requires effective management as effective managers are always ready to explore, discover and learn. They desire positive change, healthy growth and challenge the status quo.

PL-03

Managing Knowledge in the Corporate Sector

Dr. Muhammad Ramzan
Director Libraries, LUMS.

Abstract:

World economies are turning fast into knowledge economies. Making knowledge as their base for development; a physically small country like Singapore has better GDP then many of the highly populated and geographically large countries. Significant amount of resources are being used to tape the intellectual capital and embed organizational knowledge in organization's systems and processes to gain competitive edge over others and continuously increase the profits. With the application of information and communication technologies, we have seamlessly turned into global community. Companies and organizations have to compete internationally. The knowledge is being generated at a much faster pace and today's employees have greater opportunities to move to other local and international organizations. Hence, institutions and organizations are at high risk to lose their experienced knowledge workers. Resultantly they need to put in lots of time and resources in searching, training and then re-generation of knowledge, which they already possess. In other words they have to re-invent the wheel if they do not indentify, capture and use the existing knowledge; that is somewhere in the minds, undocumented ideas and in-accessible records in their organizations. The knowledge management lecture will help participants distinguish between information and knowledge, tacit and explicit knowledge, the possible sources of knowledge in organizations and appropriate systems for their capturing, organization, sharing and dissemination.

PL-04

Biological Research for Creating Wealth

Prof. Dr. Kauser Abdulla Malik

Department of Biological Sciences

Forman Christian College (A Chartered University)

Lahore, Pakistan.

Abstract:

It is generally accepted that this is the century of biology. Many fundamental discoveries in life sciences made during the last century have enabled us to develop biology into a technology which can be commercialized for the welfare of mankind. In view of its potential the Government of Pakistan has been investing liberally in propagating research and development in biological sciences at all public sector universities and R&D Institutions. Nucleus scientific human resource has been developed through an aggressive PhD program of HEC. Effort will be made in this presentation to give an overview of the activities in biological research in the country and the potential for commercialization. Some bottlenecks in way of achieving these objectives will also be discussed.

PL-05

Academic research from the alleys of the research centres to the market

Nuzhat Ahmed^{1,2}

¹Centre for Molecular Genetics, University of Karachi.

²National Institute of Blood Diseases and Bone Marrow Transplantation, Karachi.

Abstract:

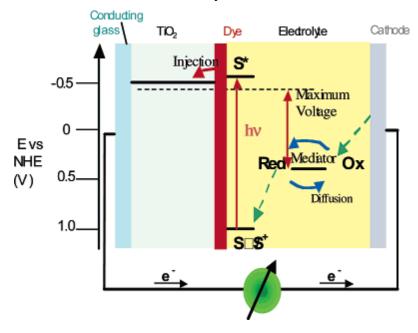
We, at Centre for Molecular Genetics University of Karachi, have been involved in developing environmentally safe technologies for sustainable development. These technologies have been developed by using indigenous bacteria isolated from the local environment. These technologies include metal removal from the waste water, biodegradation of toxic chemicals such as pesticides, crude oil and its individual components, production of antibiotics, biofertilizers and polymers such as biodegradable plastic and dextran. We are also involved in studying indigenous lactobacilli for the production of probiotics and for the treatment of hypercholesterolemia. Recently we got involved in the development of vaccine for cutaneous Leishmaniasis by using indigenous *Leishmania* isolates. Here at this forum the presentation shall be on the brief introduction of the development of these technologies at the centre. Focus shall be on commercial exploitation of these indigenously developed technologies as all of these hold good promises for developing bioindustries in Pakistan.

Keywords: Biofertilizer, Indigenous bacteria, Probiotics, Technologies, Vaccine

PL-06 Economic and Technological Feasibility of Dye Sensitized Solar Cells

Prof. Dr. Syed Arif Kazmi

HEJ Research Inst of Chem. University of Karachi, Karachi75270, Pakistan



Abstract:

The world in general and Pakistan in particular are running short of non-renewable traditional sources of energy. Our population is increasing and our lifestyle is becoming more energy dependent. If we were to rely only on traditional energy sources we will be facing even greater energy crisis than what we have now. Even if we had more fossil fuel (e.g., Thar coal) it will not be sufficient for too long besides the environmental damage which it will cause. Alternate energy is therefore of utmost importance. Solar power must be explored. There are several available technologies using solar radiation which could satisfy our energy needs, at least partially. Among these are Dye sensitized solar cells which use cheap materials, are easy to make and are light weight but their efficiency is not very good. On the other hand, solid-state junction devices, usually made from crystalline or amorphous silicon are used for making solar cells connected in series in solar panels require more sophisticated technology to manufacture and the panels are quite heavy. The cost of these devices is coming down and they are more efficient. Attempts are being made to use solar energy and photocatalysts to split water into hydrogen for use as fuel. Microbial fuel cells offer another possibility of alternate energy use. In this presentation principles of Dye Sensitized Solar Cells will be described and possible research to improve their quality and cost for feasible commercialization will be discussed.

PL-07

Modified Agrowastes: Ecofriendly Approach for Metal Removal from Aqueous Solutions

Prof. Dr. Muhammad Makshoof Athar

Director, Institute of Chemistry, University of the Punjab, Lahore, Pakistan

Email: atharmakshoof@gmail.com

Abstract:

Biosorption is emerging as a technique that offers the use of non-conventional biological materials for the removal of toxic substances (especially metal ions) from wastewaters. Functional groups like carboxyl, hydroxyl, sulphydryl and amido etc. present in these biomaterials, make it possible for them to attach metal ions from waters. Agricultural wastes and by-products are among various biological materials being sought as potential biosorbents. Every year, large amounts of straw from Triticum aestivum (wheat), Oryza sativa (rice) and Gossypium hirustum (cotton), major crops of Pakistan, are produced as by-products/waste materials. In our laboratories, we have studied the potential use of straws from these crops in their simple and modified form for the removal of various toxic metal ions. High efficiency, high biosorption capacity, cost-effectiveness and renewability are the important parameters making these materials as economical alternatives for metal removal and waste remediation. Applications of available adsorption and kinetic models as well as influences of change in temperature and pH of medium on metal are reviewed. The biosorption mechanism has been found to comprise a number of phenomena including adsorption, surface precipitation, ion- exchange and complexation. The biosorption capacities of modified biosorbent were found to increase significantly. Moreover, materials exhibited greater capacity than a number of other biosorbent materials like algae, fungi and bacteria etc. Modified materials exhibited comparatively better biosorption properties. The straw, an agricultural by-product and urea, both are easily available. The modification process is simple, cost- effective and 'green'. Hence, modified materials appear to be potentially better candidates for remediation purposes for various industrial effluents containing these metals.

Keywords: Agrowaste, Biosorption, Heavy metal, Industrial effluent

PL-08

Green Economy in Technopreneurship

Prof. Dr. Shahida Hasnain
Vice-chancellor, Women's University Multan

HEC National Distinguished Professor

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore.

Email: genetic@brain.net.pk

Abstract:

Microbes are present everywhere in each and every environment, and they effectively influence the environment in which they are growing. Microorganisms in their environment can be beneficial or harmful, but their beneficial potentials can be exploited to strengthen economy. The beneficial effects of microbes derive from their metabolic activities in the environment, their associations with plants and animals, and from their use in food production and biotechnological processes. Plant-microbe interaction is a well-known biotechnological method for improvements of plant growth and yield. PGPR (plants growth promoting rhizobacteria) are inhabitants of the rizosphere which may sense and respond to plant signals, exchange nutrients with plants cells, suffer damage due to plants defense responses and colonize or even invade root tissues creating symbioses. Salt tolerant bacteria from saline soils, rhizosphere, rhizoplane and histoplane of halophytes can promote yield and growth parameters in Wheat, Sunflower, and Mungbean under salinity stresses. Microbe's interaction with metals has been used to remediate the heavy metal contaminated environments. Similarly in the presence of plants, such microbes offer a potential alternative for detoxification / removal of heavy metals from variety of industrial effluents via various energy dependent or independent mechanisms. Microbial metabolites also have an industrial importance. Ability of microbes to secrete compounds similar to plastics opens new methods to develop biodegradable plastics that can decrease the burden of environmental pollution. Other important metabolites includes almost all kind of phytohormones i.e. Auxins, cytokinins, gibberellins etc. that have significant importance in plant biotechnology and tissue culture industry. Microbes, that are freely available in nature, with a variety of characteristics, and their proper exploitation in industry can generate usage of valuable income from relatively less resources.

PL-09

Technopreneurship: Challenges and Opportunities for Developing Diagnostics to
Control Infectious Diseases
Prof.Dr. Shahana Urooj Kazmi

Immunology and Infectious Disease Research Laboratory – Department of Microbiology ,University of Karachi- Karachi – 75270 – Pakistan.

Abstract:

Although, technological advancement has enabled us to tailor make new and novel microbes with tremendous economic potential, some of them which cause dreadful diseases still remain number one cause of death and morbidity. Providing microbiologically safe and nutritious food as well as rapid diagnosis, control and management of new and emerging infectious and chronic disease still remains a big challenge for the researchers and healthcare scientists.. Despite decades of epidemics that we have experienced, we still do not have adequate number of diagnostic tests. More than 200,000 die of typhoid, 1 million people die of malaria, 4.3 million of acute respiratory infections, 3 million from enteric infections, 5 million die of AIDS and tuberculosis every year, because diagnostics and protective vaccines are inaccessible to those who need them most. Manufacturing Companies are not interested in developing diagnostics for low resource countries who do not offer a big market. Diagnostics developed globally do not address the disease needs of developings and are not easily accessible,, they are ill-adapted for countries that lacked trained personnel, ifl available still cannot be used due to the lack of proper infrastructure/facilities or not affordable due to high cost. Therefore people do not get effective treatment .. In view of the world being a global village, of climate change, escalating fuel price and cost of living, what used to be diseases of the developing countries like TB /HIV/AIDS /Enteric diseases / Malaria etc., now are also affecting the developed countries. In the area of diagnostics, R&D need to be performed with the client in mind so that the product developed is relevant, Technology chosen should be innovative yet allow for a low cost of production, with commercial viability and relevance, Developed products should be used to enhance economy of the country (via commercialization and creation of spin-off companies) as well as enhance the quality of life of the people. In developing kits, R&D need to be performed with the client in mind so that the product developed is relevant to address the correct disease that is most needed by the target population. When designing diagnostics for low resource countries, it is imperative that the criteria for designing include the sustainable values such as availability, accessibility, affordability, quality and appropriate to the people who need them most. Methods developed should be Rapid, Specific, Sensitive, Easy to perform, Easy to interpret results, Cost effective, may be Transported without cold chain. Research and development in diagnostics especially for neglected diseases that are relevant only in the region or in the poor countries must be addressed if possible with the local researchers so as to provide relevant local solutions which could be used globally. There is an urgent need to design economically viable proposals to create diagnostics to help the poorest communities enjoy the basics taken for granted in the developed world. For this purpose we need to conduct classes on "Entrepreneurial Design for Extreme Affordability". Teach a new generation of young entrepreneurs to use their education, training, business and engineering skills to design and sell products – profitably- for the developing world, transforming higher education and research for sustainable tomorrow . We should develop disease relevant indigenous diagnostic kits making use of available modern technologies. Specially rapid protein or DNA - based diagnostics for diseases like Typhoid ,Cholera,Campylobacteriosis Tuberculosis, Dysentery, Dengue ,Paratyphoid,Nosocomial infections.Also consider developing point of care tests (Protein /DNA tests). Try to combine Diagnostics with green technology/alternative energy, Using solar powered energy, alternative energy, Non PCR-based diagnostics would be more suitable for low resource with No electricity, No cold chain facilities ,Lack of skilled workers to perform the test. Entrepreneurship can transform research results to products, patents, economically feasible industries. Technopreneurship is a solution to many economic problems like urbanization, poverty, unemployment and economic development. It helps in rural and urban development. But development of entrepreneurship in the areas of Health, Education and Agriculture requires special skills like human development, knowledge of global biomedical and agriculture products market.

PL-10

Prof. Dr. Mian Ahmad Hanan

Chairperson, Mass Communication
Forman Christian College (A Chartered University), Lahore, Pakistan.

PL-11

Ecopreneurship in Environment Services

Mr. Asif Farooki

Chief Eexecutive Officer, Waste Buster, Pakistan.

PL-12

Bio-Inspired Superhydrophobic Surfaces by Colloidal Routes

Dr. Muhammad Akram Raza
Centre of Excellence in Solid State Physics,
University of the Punjab, Quaid-e-Azam Campus, Lahore-54590, Pakistan

Abstract:

Superhydrophobicity is a hot topic of today's research owing to its emerging applications ranging from lab-on-chip devices to self-cleaning purposes for glasses, textiles and solar panels. Surface roughness and surface chemistry are the important factors that control surface wettability, but hierarchical roughness plays a vital role to achieve stable superhydrophobicity of the surfaces as depicted by biomaterials such as the lotus leaf.

In this work, a simple colloidal route is presented to fabricate substrateswith diverse surface roughness to achieve a range of surface wettability from superhydrophilicity to sticky and non-sticky superhydrophobicity. Single length scale roughness was obtained either by adsorption of silica spheres ofdifferent sizes (850nm, 440nm and 130nm diameter) or by deposition of AuNPs of 13nm, 25nmand 45nm. The hierarchical was achieved by bottom up technique employing silica sphere arrays as coarser structures, decorated with AuNPs as the finer structure. To lower the surface energy,the silica surfaces were hydrophobized by perfluorooctyltriethoxysilane and AuNPs bydodecanethiol. The surface morphology was examined by scanning electron microscopy, whilewettability measurements were performed by using the sessile drop method. It was concluded that wettability can be controlled by changing the surface chemistry and/or length scales of the structures. To achieve truly non-sticky superhydrophobic surfaces, hierarchical roughness plays a vibrant role.

PL-13

Biophysical and biochemical characterization of novel R7BP and Gaoa *in-vitro* interaction explains binding specificity towards RGS11

Yasar Saleem¹*, 2, 3</sup>, Key-Sun Kim^{2, 3}, Shaista Nawaz¹, Muafia Shafique¹, Khurram Shahzad¹ and Quratuain Syed¹

1. FBRC, PCSIR Labs Complex, Lahore, Pakistan. 2. CNS, Korea Institute of Science and Technology (KIST), Seoul, South Korea. 3. University of Science and Technology (UST), Daejeon, South Korea. *Email: ysaleem73@hotmail.com

Abstract: Regulator of G protein signaling 11 (RGS11) is the least characterized member of the R7 family of Gγ-like GGL domain-containing RGS proteins. All R7-RGS proteins of a variety of cell types are found in Gβ5-containing complexes that exhibit a number of unique functional properties. However, presence of Gβ5 reduced the affinity of R7-RGS7 for Gα subunits, also only RGS7 bound to Muscarinic M3-Receptor, but the Gβ5-RGS7 dimer did not, making it difficult to study differential interaction of R7-RGS proteins. Here, we report the successful purification of functionally intact, Gβ5-free recombinant RGS11 (rRGS11), obtained by expressing N- and C-terminally truncated form of RGS11 in Escherichia coli BL 21 (DE3), that differentially interact with R7BP and Gα_{oa}. rRGS11 was capable of interacting with Gα_{oa} and R7BP (RGS7 family binding protein) with equilibrium dissociation constants (KD) of 904 (±208) nM, and 308 (±97) nM, respectively. It also induced several-fold increase in the GTPase activity of Gα_{oa}. The binding of rRGS11 was differential with a binding preference for R7BP over Gα_{oa} implying extended roles of R7BP. In addition, we identified a novel interaction between Gα_{oa} and R7BP with a KD of 592 (±150) nM. The production of stable and functional rRGS11 would provide chances to discover more functions of RGS11 yet to be identified.

BPC-01

In vivo pathogenicity testing of Aspergillus species

Naureen Akhtar

Institute of Agricultural Sciences, University of the Punjab, Quaid-e-Azam Campus, Lahore-54590, Pakistan.

Abstract:

Thespecies of Aspergillus are ubiquitous in nature and have been involved in human affairs mainly due to their industrial applications and pathogenicity. In the recent past larvae of Galleria mellonella are emerged as effective infection model for both bacteria and fungi. In this present study different species of Aspergillus have been evaluated for their pathogenecityusing G. mellonella larvae. The main findings presented here are (i) a wild type prototrophic laboratory strain of A. nidulans used extensively in research work on nitrate assimilation and transport are safe to handle in the laboratory (ii) A. fumigatus and A. flavus are pathogenic as reported before in certain published research papers and (iii) previously unreported aspergilli; A. terreus, A oryzae and A. sojae have been observed to have quite nasty consequences for moth larvae and most likely constitute a health risk for humans.

Keywords: Aspergillus, Pathogenicity, Moth larvae

BPC-02

Taxonomic evaluation of family Moraceae on the basis of leaf epidermal anatomical characters

Zubaida Yousaf

Department of Botany, Lahore College for Women University, Lahore.

Abstract:

Family Moraceae is an economically and medicinally important angiosperm family. Important epidermal anatomical markers used for identification of shapes of epidermal cells, types of trichomes, stomatal type, developmental stages and stomata patterning. These markers could enhance our understanding about the systematics of fig family. Comparative lengths of epidermal cells, glandular and non-glandular trichomes are considered as an important taxonomic marker for species differentiation. Family Moraceae is characterized by eight different stomatal types (anisocytic, anomocytic, cyclocytic, encyclocytic, laterocytic, paracytic, paracytic predominant and staurocytic). However, staurocytic stomata were dominant in family Moraceae. Shape variation of silica bodies is ranged from irregular shape (Ficus macrophylla Roxb) to round (Ficus racemosa L., Ficus nerifolia J.E.Sm., and Ficus triangularis Roxb). Based on present studies leaf epidermal anatomy has been proved as an important tool for identification and characterization of family Moraceae, it can be effectively utilized for solving persistent taxonomic problems of family Moraceae.

Keywords: Leaf epidermal cells; stomatal types; stomatal development; stomatal patterning; trichomes.

BPC-03

Antimicrobial activity of *Allium sativum* on methicillin resistance *Staphylococcus aureus* (MDR) Strains

Saliha Hafiz and Saba Riaz

Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-e-Azam Campus, Lahore-54590, Pakistan.

Abstract:

MRSA is becoming a very serious health issue because of its resistance to a number of antimicrobial drugs. This research aims to introduce some natural therapy for treatment of such as infection. MRSA is of particular concern as a serious cause of deaths worldwide because it cause resistance to multiple drugs and as a result there are a very few treatment options available. Natural herbs are a major source of natural drugs and their use as an alternative medicine for various disease has been increased. MRSA Strains were isolated according to standard operating procedure recommended by Clinical laboratory institute. Antimicrobial activity of Garlic was checked by using Garlic extract and apply it on these strains by well diffusion method. The Strains were sensitive against the applied concentration of garlic extract and large zone of inhibition appear. Further evaluation of extract is under process. Preliminary findings suggests that *Allium sativum* can be used as potential therapeutic agent after further refining.

Keywords: MRSA, *Allium sativum*, well diffusion, Antimicrobial activity

BPC-04

Impact of bacterocin production against multidrug resistant *Pseudomonas Spps*.

Sumyyia Abrar and Saba Riaz

Department of Microbiology and Molecular Genetics, University of the Punjab Lahore, Pakistan. **Abstract:**

Bacteriocins are well known for their antibacterial activity against Multidrug resistant strains (MDRs). Lactobacilli are known friendly bacteria for their antibacterial activities against pathogens. The antibacterial activity of different strains of Lactobacilli was analyzed against MDR Pseudomonas sp. Well- it is necessary to purify the antibacterial molecule out of putative bacteriocin for further analysis. Diffusion assay was used for screening of putative bacteriocins produced by Lactobacillus strains against MDR Pseudomonas sp. Multidrug resistant strains were selected based on MAR (Multiple antibiotic resistance) index. Five bacteriocins were obtained from Lactobacillus strains isolated from commercial products. These bacteriocins showed a strong anti-bacterial activity against selected MDRs. Decrease in zone sizes was observed when putative bacteriocins were treated with heat, SDS and Protinase k. It was observed that Lactobacillus showed a significant antibacterial activity in-vitro in the presence of putative bacteriocins against selected MDRs and further experiments are underprocess. Putative Bacteriocins produced by Lactobacilli exhibit significant antibacterial activity against MDR Pseudomonas sp. The peptidal component of these bacteriocins can be used as an alternative therapy. Hence, it is necessary to purify the antibacterial molecule out of putative bacteriocin for further analysis.

Keywords: Lactobacilli sp., MAR, well-diffusion assay, putative Bacteriocins.

BPC-05

Genetic Study of Chlorpyrifos Degrading Bacteria

Adeela Irshad and Farkhanda Jabeen Department of Botany, University of the Punjab Quaid-i-Azam. Campus, Lahore-54590, Pakistan.

E-mail: rahimfarkhanda@hotmail.com

Abstract:

Eight Chlorpyrifos degrading Bacterial strains (S1a, S1-b, S2-a, S2-b, S3-a, S3-b, S4-a, S4-b) isolated from different soil samples of Nankana Sahib were selected for the Genetic study. Optimum growth response of the bacterial strains was observed with varying concentration of Chlorpyrifos (50ug/ml, 100ug/ml, 150ug/ml, 200ug/ml, and 250ug/ml) in minimal media. Different antibiotics were used to check the antibiotic resistant property of the strains. Alkaline lysis method was used for plasmid DNA isolation and detection. The bacterial strains S2-b and S3-b showed single plasmid in each. The approximate size of the plasmid was ranged between 3Kb and 2.7Kb respectively. To check whether the pesticide degrading gene was plasmid born or not some curing experiments were performed. Physical agents like high temperature and pH were used for plasmid curing. The bacterial strains S2-b and S3-b responded against high temperature 55° C and become cured. Analysis of cured strains was done with different parameters such as antibiotic sensitivity test, biodegradation potential which was confirmed by TLC and by the loss of growth in minimal media containing Chlorpyrifos. The cured strains (S2b_c and S3-b_c) showed no spot against 3, 5, 6- trichlo -2- pyridinol (TCP), which is the major degradation product of Chlorpyrifos so that the degradation property was lost as the confirmation of curing. Similarly antibiotic resistance against ampicillin, bacitracin and amoxicillin were totally lost in the cured strains.

Keywords: Chloropyrifos, Biodegradation, Plasmid DNA

BPC-06

Mutagenic strain improvement and lab scale production of the antitumor compound Actinomycin D from an indigenous isolate Streptomyces griseoincarnatus CTF15

Saliha Bashir and Imran Sajid

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus, Lahore-54590, Pakistan.

Abstract:

An indigenous actinomycete strain designated as Streptomycesgriseoincarnatus CTF15, investigated in a previous study was found to be the highly stable producer of the antitumor compounds, Resistomycin, Tetracenomycin D and Actinomycin D. In this study we have attempted the mutagenic strain improvement and lab scale production of the commercially important antitumor compound "Actinomycin D" from this strain. A total of 100 mutants were obtained by mutagenesis of the wild strain using chemical and physical mutagens. Ethidium bromide having concentrations of 10 ug/ml, 20 ug/ml, 50 ug/ml and 100 ug/ml was used as chemical mutagen, while the exposure of vegetative cells and spores to short UV (254 nm) for different time intervals (15 min, 30 min, 45 min and 60 min) was used as physical mutagen. A significant increase in the yield of the target compound "Actinomycin D" was observed in various mutants, while screened by TLC, HPLC-RI, comparison of the weight of crude extracts of mutants with that of the wild strain in a unit volume (50 ml) of culture broth and by

determining the level of cytotoxicity against *Artimia salina* in a microwell cytotoxicity assay. The selected two mutants with enhanced production were cultivated up to 5 liters in a lab fermenter, the culture broth and cellular mass was extracted by solvent extraction and Actinomycin D was partially purified from the crude extract by a series of chromatographic techniques.

Keywords: Actinomycin D, *Streptomyces griseoincarnatus* CTF15, Mutagenesis, Antitumor compound, Cytotoxicity

BPC-07

Desert Actinomycetes: Isolation, Identification, and Screening for Antimicrobial Potential against Methicillin Resistant *Staphylococcus aureus* (MRSA)

Adeela Fatima and Imran Sajid
Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore,
Pakistan.

Abstract:

The changing pattern of diseases and the emergence of resistant bacterial strains against currently used antibiotics continuously put demand for novel antibiotics. Actinomycetes is the most important group of bacteria for the production of medically valuable secondary metabolites specially antibiotics. The aim of this study was to screen the desert actinomycetes against methicillin resistant *Staphylococcus aureus* (MRSA). Thirty one strains of actinomycetes were isolated from the soil and sand samples of Cholistan desert. These isolated strains were characterized morphologically, biochemically, physiologically and genetically. Twenty five isolates exhibited promising antimicrobial activity against MRSA, however the isolate S₇19 was found to be the strongest inhibitor of almost all the tested MRSA strains while screened biologically in an agar diffusion assay. In chemical screening, the crude extracts were analyzed by thin layer chromatography (TLC) using different spraying reagents and by HPLC-UV/IR; the metabolic fingerprints of each of the extract demonstrated an impressive chemical diversity of bioactive secondary metabolites. The activity against MRSA and production of diverse metabolites reveal that the desert actinomycetes are the proliferant producers of useful antimicrobial agents, and should be screened further with respect to the novel drug discovery.

Keywords: Desert actinomycetes, Screening against MRSA, Biological screening, Chemical screening.

BPC-08

Estimation of toxic hexavalent reduction potential of *Bacillus pumilis*, *Exigubacterium* and *Cellulosimicrobium cellulans*

Fatima Rehman and Muhammad Faisal
Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore-54590, Pakistan.
Email: fatima_genius_@hotmail.com

Abstract:

Present study describes the three bacterial strains *Bacillus pumilis*, *Cellulosimicrobium cellulans* and *Exiguobacterium* which were grown in Luria-Bertani (LB) medium at 500 μ g/ml Cr (VI). The hexavalent chromium reduction potential was checked by growing the strains in Deleo and Erhlich medium at 200 and 400 μ g/ml K_2 CrO₄ using diphenylcarbazide methods. The optimal Cr

(VI) reduction potential in strains *Bacillus pumilis*, *Exigubacterium* and *Cellulosimicrobium cellulans* was 98, 72 and 87%, respectively, at an initial K₂CrO₄ concentration of 200 μg/ml in the presence of arsenic, cobalt and nickel salts at pH 3 temperature 37 °C. At 400 μg/ml Cr (VI) optimal reduction potential in strains *Bacillus pumilis*, *Exigubacterium* and *Cellulosimicrobium cellulans* was 57, 48 and 57%, respectively at pH 7 at 45°C after 72 hours. These three strains have significant Cr (VI) resistance and reduction potential and *Bacillus pumilis* is found to be prominent for having greater reduction potential.

Keywords: Chromium, bioremediation, heavy metals, *Bacillus pumilis*, *Exigubacterium* and *Cellulosimicrobium cellulans*

BPC-09

Screening of bioplastic producing bacteria from different environmental sources

Naseem Naeem and Nazia Jamil

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus, Lahore-54590, Pakistan.

Abstract:

Synthetic Plastics have resistance to biological breakdown that result in its accumulation in the environment. This emphasizes the need to search for biodegradable polymer, which is easily disposable and degradable. Bacteria synthesize and accumulate polyhydroxyalkanote (PHA) as reserve energy source under limiting conditions of mineral nutrients. For the synthesis of bioplastics, samples were isolated from different environmental sources (Root soil, sewage, polluted water, rotten potato and autowork shop soil).42 strains were isolated. Following Gram staining, out of 42 strains 20 were gram negative strains. These 20 gram negative strains were streaked on PHA media with Nile blue indicator for the screening of PHA producers. Five out of 20 strains showed fluorescence. These 5 strains were characterized by biochemical tests and the possible identified genera were Pseudomonas, Enterobacter and Klebsiella. PHA producer screening was further confirmed following Sudan B staining. Large black PHA granules were visible in the cells. Growth curves of these samples were plotted by using PHA Screening media with 2% glucose as a carbon source and Nutrient broth at different times (hours) intervals and their optical density was taken at 600nm. Most strains showed the best growth between the range of 48 to 81 hours. Maximum biomass was obtained at the range of 33 to 57 hours. PHA extraction was done by using Na hypochlorite and chloroform. DNA isolation was done by CTAB method. PHA extraction and PCR for the amplification purpose is under processing.

Keywords: Biodegradable plastics, Bacteria, Screening methodology

BPC-10

Effect of copper on adherence, hydrophobicity, aggregation and biofilm formation of salt tolerant bacterial strains

Iqra Jabbar and Anjum Nasim Sabri
Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore, Pakistan.

Abstract:

Copper is a toxic heavy metal and is well-known for its anti-bacterial properties. The presence of toxic heavy metals like copper, however, can induce various mechanisms that enable bacteria to

survive in the presence of heavy metals. The present study was aimed at investigating the influence of copper on various characteristics (i.e. motility, aggregation, hydrophobicity and biofilm formation) of two salt tolerant bacterial strains, Bacillus sp. HAa2 and Halomonas meridiana PAa6. Two Cu²⁺ concentrations 500 µg/ml and 1000 µg/ml were used for the subsequent tests. Motilities (swimming, swarming and twitching) were tested by growing the strains on swimming, swarming and twitching agar media. Co-aggregation and auto-aggregation assays were performed for testing the aggregation abilities of the strains. Soil aggregation abilities were also studied by growing the roots in-association with the strains. Hydrophobicity of the strains was determined by BATH (bacterial adhesion to hydrocarbons) and SAT (salt aggregation test). Biofilm formation on PVC micro titter plates was analyzed by CV staining and quantification, and on glass slides by acridine orange staining followed by fluorescent microscopy. Swimming, swarming and twitching motilities were greatly reduced in the presence of copper. However, the strains showed a varying response for co-aggregates and autoaggregates formation under copper stress, with an increase in aggregation at 1000µg/ml Cu²⁺ concentration. In addition, both strains showed a hydrophobic nature irrespective of copper treatment. In most of the cases, biofilm formation was significantly reduced in the presence of copper. It was henceforth concluded that the presence of copper affected the motility and biofilm formation of the strains, negatively.

Keywords: Copper, Biofilms, Motility, Aggregation, Hydrophobicity.

BPC-11

Ethnopharmacological investigation of a perennial shrub, *Rhynchosia pseudo cajan* Cambess, by antimicrobial, antioxidant and anthelmintic activities

Arusa Aftab

Department of Botany, Lahore College for women university, Lahore, Pakistan

Abstract:

The present study was under taken to evaluate the antimicrobial, antioxidant and anthelmintic activities of the plant *Rhynchosia pseudo cajan*Cambess. The crude extracts of powdered plant material were obtained in various polar and nonpolar solvents, viz: petroleum ether, chloroform, methanol and distilled water. Well defined zones of inhibition were recorded indicating that the plants were potent against pathogenic microbes, such as i.e. *Staphylococcus aureus*, *Staphylococcus saprophyticus*, *E.coli* and *Pseudomonas aeruginosa*, *Aspergillus parasiticus* and *Rhizopus oryzae*. The antioxidant activity of all the plant extracts was studied by DPPH Assay, Total Antioxidant Assay and Total phenolic Assay and the remarkable values comparable with the standard antioxidants were recorded. The in vitro anthelmintic activity of all the extracts was found much stronger than the standard medicine, Levamisole. Therefore, the plant can be declared as the possible source of antimicrobial, antioxidant and anthelmintic agents to treat the ailments of man and his domestic animals like sheep, goat etc. For this purpose additional studies including toxicity level of these plants are needed.

Keywords: Antimicrobial, Antioxidant, Minimum inhibitory concentration.

BPC-12

Role of MSX1 mutations in Pierre Robin Sequence and Non-Syndromic cleft lip/or cleft palate in Pakistani population

Saira Malik and Waqas Ahmad

Department of Microbiology and Molecular Genetics, University of the Punjab Lahore, Pakistan Email: saira.mmg@pu.edu.pk

Abstract:

Cleft lip and cleft palate (CLP) are genetic birth defects of the face. Cleft lip is manifested by the presence of a cleft in the upper lip and cleft palate involves a cleft in the roof of the mouth. Pierre Robin Sequence or "Pierre Robin Malformation Sequence" (PRS) is a congenital malformation and the majority of the patients present with a horseshoe-shaped cleft palate in addition to other facial abnormalities including small lower jaw, and posterior displacement of the tongue. *MSX1* gene (Muscle segment homeobox gene) encodes a member of the muscle segment homeobox gene family. The protein encoded by *MSX1* functions as a transcriptional repressor during embryo formation. Different mutations in *MSX1* are known to cause facial clefts in humans. We enrolled 17 patients with different cleft phenotypes from different areas of Lahore and Azad Kashmir. Mutational analysis of *MSX1* gene is in progress for 10 individuals with cleft palate only phenotype, 4 with cleft lip with or without cleft palate, and 3 individuals with Pierre Robin sequence. Currently, one patient has been sequenced for *MSX1* gene and no mutation was found. Sequencing is in progress for the other patients.

Keywords: Cleft palate, Pierre Robin sequence, MSX1.

BPC-13

Prospects of Cyanobacteria as next generation biofuel source

Sara Junaid and Mehboob Ahmed

Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-e-Azam Campus, Lahore-54590, Pakistan.

Email: sarajunaid7@yahoo.com

Abstarct:

Continuous depletion of fossil fuels resources results in their declined production is the main global problem and also in Pakistan. Researchers in the field of chemical and biotechnology are working on the alternates of fossil fuels. Globally, so far the main attention was on the production of first generation biofuels i.e. from edible crops. This was a better approach but with severe concerns about the decrease in available food from crops. Therefore, there was a shift to second generation biofuels i.e. non-edible but cellulosic biomass producers. Finally, now it's the age of third generation biofules, fuel from various algae. The third generation biofuel is considered one of the most efficient ways of generating bioenergy due to its major advantages over other methods. Some of the main features such as higher biomass yield per hectare as compared to plants, continuous harvesting and very less or negligible input as fertilizers. People so far have targeted macroalgae for biofuels due to their higher biomass and ignored cyanobacteria. But cyanobacteria, being prokaryotic in nature have higher growth rate and high oil to biomass ratio is an excellent candidate for biofuel production. They also have a limited nutritional requirement making them easy to grow on large scales. Their prokaryotic nature and

small genomic DNA further enhance their genetic manipulation. Thus Cyanobacteria can be an efficient source of various biofuels in future.

Keywords: Renewable energy, Biodiesel, Oil extraction

BPC-14

Biofilm formation by isolated strains and antibacterial activity of Acacia against isolated strains

Rabia Sultan and Anjum Nasim Sabri

Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore-54590, Pakistan.

Email: rabiammg@yahoo.com

Abstract:

Biofilm formation is an important phenomemon seen in nature by different microorganisms. Plants have certain phytochemicals that can inhibit boifilm formation by microorganisms. Aim of this study was to check biofilm formation by isolated strains and to check antibacterial activity of Acacia on these strains. Biofilm formations is basically adherence of microorganisms to surfaces in their exopolysaccharide matrix. Past studies have shown that many microorganisms are involved in biofilm formation .Studies have also shown that Acacia extract can inhibit this activity. Biofilm formation by isolated strains was checked by coaggregation and autoaggregation assays, slime production test, microtiter plate assay, biofilm formation on glass, bacterial adherence to salt assay, bacterial adherence to hydrocarbons assay. Further experiments like EPS extraction are ongoing. Methanolic extract of Acacia have shown antibacterial activity that was determined by standard procedures of agar well and disc diffusion assay. Minimal inhibitory concentrations of extract were also determined. Phytochemical screening for plant was done to know that which secondry metabolites of plants are causing this inhibition. Strains have shown ability of biofilm formation. They have also shown sensitivity against Acacia extract Preliminary findings suggest that isolated strains are involved in biofilm formation and Acacia. Extracts have ability to inhibit this. So Acacia is a strong candidate for preventing biofilm formation in future.

Keywords: Acacia, Microtiter plate assay, EPS extraction, Minimal inhibitory concentration.

<u>BPC-15</u>

Microbiologically Influenced Corrosion of stainless steel 304 (SS304) in the presence of *Bacillus subtilis* and *Pseudomonas aeruginosa*

Hafiz Zeshan Wadood¹, Aruliah Rajasekar², Yen-Peng Ting³ and Anjum Nasim Sabri ¹ Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore-54590, Pakistan.

²Department of Civil and Environmental Engineering, National University of Singapore, Singapore 117576.

³Department of Chemical and Biomolecular Engineering, National University of Singapore, Singapore 117576.

Abstract:

The microbial production of extracellular polymeric substances (EPS) forms a biofilm on metal surface that may either inhibit or accelerate corrosion of underlying metal surface. In the present

study, the biocorrosion behavior of stainless steel 304 (SS304) in the presence of aerobic bacteria *Bacillus subtilis* strain S1X and *Pseudomonas aeruginosa* strain ZK was investigated in minimal salt medium with 1.5% sodium chloride as electrolyte. Biocorrosion was evaluated using Tafel polarization, electrochemical impedance spectroscopy (EIS), scanning electron microscopy—energy dispersive spectrum analysis (SEM-EDAX), atomic force microscopy (AFM) and Fourier transform infrared spectroscopy (FTIR). The corrosion rate and corrosion current were lower in the presence of both bacteria when compared to control medium. Electrochemical data showed that both bacteria inhibit corrosion of SS304 through the formation of a passive layer on metal surface, scanning electron microscopy-energy dispersive spectrum analysis (SEM-EDAX) also supported this observation. Atomic force microscopy (AFM) and Fourier transform infrared spectroscopy (FTIR) showed the formation of thick biofilm on SS304 surface. The pH values of bacterial inoculated systems decreased with increasing incubation time which showed the production of some acidic metabolic products by bacteria.

Keywords: Microbiologically corrosion, tafel polarization, Electrochemical Impedance Spectroscopy (EIS), Scanning Electron Microscopy-Energy Dispersive Spectrum Analysis (SEM-EDAX).

BPC-16

Lab scale production of polyhydroxyalkanoates using mustard oil as carbon source

Hasnain Javed and Nazia Jamil

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore54590, Pakistan.

Email: hasnain_javed@hotmail.com

Abstract:

Mustard oil was considered relatively cheap, easily available, included in vegetable oil and don't having much volatile characteristics. Fermentation and PHA production was done in batch culture in 5 liter fermenter to optimize the PHAs production under various experimental conditions. Samples subjected to PHA production were checked by using inoculating on PHA detection agar and PHA broth. Total of 67 bacterial strains were isolated and purified from different industrial and domestic waste waters from various regions of the Pakistan. Strains were checked for PHA production by Sudan black and Nile blue staining. Quantitative analysis for biodegradable plastic produced by different bacterial species was performed by Modified surfactant hypochlorite method. High PHA production was detected in 35 strains belonging to different genera including Pseudomonas, Staphylococcus, Escherichia coli and Enterobacter as predominant genera. PHA production under different carbon sources, nitrogen concentration, pH and temperature was estimated. The isolated strains of Pseudomonas showed the ability to synthesize polyhydroxyalkanotes in batch culture. Synthesis of PHA was observed in exponential growth and it depended on carbon/nitrogen ratio in the culture. It can be concluded that the PHAs storage capacity was higher two to three times in aerobic compared to anoxic conditions. PHA production of Pseudomonas species were subsequently authenticated at molecular level by PCR analysis and the phaC gene (540 bp) encoding PHA synthase was amplified. After phaC gene sequencing and 16Sr RNA ribotyping, most of the phaC gene containing strains showed homology to Pseudomonas aeruginosa. Positive samples yield a specific 540-bp PCR product representing partial coding sequences of the phaC1/C2 genes. PHA polymerase 1 (PhaC1) and PHA polymerase 2 (PhaC2) from Pseudomonas aeruginosa were

screened, sequenced and submitted to gene bank with accession numbers HM598296, HM598297 and HM598298.

Keywords: Mustard oil, Polyhydroxyalkanoates, Fermentation, *pha*C2

BPC-17

Production of medium chain length poly-3-hydroxyalkanoates (mcl-PHAs) from unrelated carbon source by indigenous *Pseudomonas* sp.

Iftikhar Ali¹, Nazia Jamil¹, Bruce A Ramsay² and Juliana A Ramsay²

Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore-54590. Pakistan

Department of Chemical Engineering, Queen's University, Kingston, K7L 3N6, Canada.

Email: iftikhar ali iftikhar@yahoo.com

Abstract:

Poly-3-hydroxyalkanoates (PHAs) are the biodegradable biopolyesters that resemble in their properties with petroleum based non-biodegradable plastics. Bacteria can produce these PHAs using different carbon sources under unfavorable conditions. Medium chain length poly-3-hydroxyalkanoates (mcl-PHAs), a class of PHAs, production has been optimized by newly isolated *Pseudomonas* sp. These bacteria were grown on varying ratios of C/N under nitrogen or phosphate limitation in shake flask. Polymer was extracted from bacteria and purified before its characterization by GC-FID and/or GC-MS. This work resulted the production of mcl-PHAs from unrelated carbon source i.e., glucose, instead of short chain length (scl) PHAs that are characteristic when used glucose as carbon source. Gas chromatography confirmed the presence of mcl-PHAs i.e., 3-hydroxyoctanoate (3-HD), 3-hydroxydecanoate (3-HD) and 3-hydroxydodecanoate (3-HDD). The isolated bacteria have shown great interest in their biopolymer production at higher levels using unrelated carbon source, glucose. The limitation of phosphate or nitrogen to produce unfavorable conditions has been addressed in this work as well. **Keywords:** Biopolymer, GC-MS, *Pseudomonas* sp.

BPC-18

Risk assessment studies of transgenic Bt diet on rats

Sana Khalid

Department of Botany, Lahore College for Women University, Lahore, Pakistan. Email: Sanakhalid4@yahoo.com

Abstract:

Advances in genetic engineering in recent years have led to the development of plants that are resistant to some insects through incorporation and expression of genes encoding delta-endotoxins (δ - endotoxins) from the bacterium *Bacillus thuringiensis*. As transgenic crops have revolutioned the world agriculture on one side, there has been a great concern regarding the impact of these transgenic food on animals and human being which are the main consumers of transgenic feed. A 120 days study was carried out to evaluate the potential risk of transgenic *Bt* diet on experimental rats. Weight gained/loss, mortality/survival, insecticidal gene integration and expression, biochemical and histological studies were the main parameters undertaken to investigate effect of transgenic *Bt* diet in these animals. No lethal effects of transgenic *Bt* diet on both groups of animals were observed. Rats had very similar growth pattern when fed either *Bt* transgenic or non *Bt* diet. No significant differences in relative weights observed during the entire period of study. No any evidence of insecticidal gene integration and expression was

found. Histological and biochemical tests also suggested that transgenic diet did not make any significant morphological and physiological differences among experimental organisms.

Keywords: Risk assessment, Transgenic; *Bt* diet, Rats.

BPC-19

Anti-Proliferative activity of indigenous Actinomycetes isolated from different habitats of Pakistan: Prescreening and Ribotyping

Usman Aftab and Imran Sajid

Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore 54590, Pakistan.

E-mail: usmanaftab.mmg@gmail.com

Abstract:

Actinomycetes are a group of Gram positive bacteria with high G+C Content. This group of bacteria is capable of producing different types of novel secondary metabolites. Many of these metabolites possess different biological activities and have the potential to be developed as therapeutic agents. The aim of the present study was to screen the indigenous actinomycetes for cytotoxic and anti-proliferative compounds. In our screening, several water and soil samples were collected from various habitats of Pakistan. More than 500 isolates were isolated, 120 of which were selected for initial identification and cytotoxic profiling. The strains were characterized on the basis of their morphological, biochemical and physiological behavior. In a biological screening the crude extracts obtained from the culture broth of selected strains were analyzed for their cytotoxic activity through Brine shrimp microwell cytotoxicity assay. The isolates with high larvicidal activities were then tested for anti-tumor or anti-proliferative activity against various proliferative cell lines (Hela, MD-BK, Vero cell lines) through methyl thiazolyl tetrazolium (MTT) bioassay method. 20 isolates were selected with high inhibition rate against proliferative cell lines. These selected isolates were then genetically characterized through 16SrRNA sequencing and was found that majority of these strains were belonging to a well-known family of actinomycetes named as Streptomyces.

Keywords: Actinomycetes, Secondary metabolites, Cytotoxic profiling

<u>BPC-20</u>

Detoxification of Hexavalent Chromium by *Cellulosimicrobium cellulans* strain KCr16 and *Exiguobacterium* sp. strain KCr19

Samina Yasin and Muhammad Faisal
Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore-54590, Pakistan.
Email: samyyaseen@gmail.com

Abstract:

Hexavalentchromium is highly toxic, carcinogenic and teratogenic to man and other animals. Some bacterial species have the ability to reduce [Cr (VI)] to a stable speciation state of trivalent chromium [Cr (III)] which is insoluble and comparatively less toxic. Therefore reduction of Cr (VI) thus provides potential as a mean for environmental bioremediation of Cr (VI) pollution. In this study two chromium resistant bacterial strains *Cellulosimicrobium cellulans* strainKCr16 and *Exiguobacterium* sp.strain KCr19 were used. This study was conducted to describe the effects of some environmental factors such as pH, temperature, concentration and time on Cr(VI)

reduction. It was found that at the temperature optima of 37-45°C and pH7.0, the specific activity of Cr(VI) reduction was determined to be 48% and 67% for *Exiguobacterium* sp. strain KCr19 and *Cellulosimicrobium cellulans* strain KCr16 respectively. Addition of domestic wastewater causes an increase in reduction activity to 75% and 62.5% for *Cellulosimicrobium cellulans* strain KCr16 and *Exiguobacterium* sp. strain KCr19 respectively, while heavy metals such as Cu, Hg and Cd did not enhance nor reduce their Cr reduction potential effectively. Both of the strains completely reduced Cr (VI) in culture media at 400 µg/ml concentration within a period of 48–72 h. Assay with resting and permeabilized cells (treated with Triton X-100 and Tween 80) and cell-free assay demonstrated that the Cr (VI) reduction activity was mainly associated with the soluble fraction of cells. Considering the major amount of chromium being reduced within 24-48h, these fractions could have been released extracellularly also during their growth.

Keywords: Chromate reduction, Cr (VI), Heavy metals, *Exiguobacterium* sp. strain KCr19, *Cellulosimicrobium cellulans* strain KCr16.

BPC-21

Screening of chromium -resistant rhizobacteria and their role in promoting plant growth Fayqa Komal and Sikander Sultan

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus Lahore-54590, Pakistan.

Abstract:

Extensive use of chromium (Cr) compounds in various industries especially leather tanning industry in Pakistan has resulted in severe contamination of surrounding soils. Bacteria have been shown to play effective role in geochemical cycling of hexavalent chromium. So, bioremediation of chromium is of considerable interest as environment friendly approach. Fourteen chromium resistent and reducing rhizobacterial strains AP-1,AP-2,AP-3,AP-4,AP-5,AP-6,AP-7,SF-1,SF-2,SF-3,SF-4,SF-5,SF-6 and SF-7 were isolated from the rhizosphere of Alternanthera pungens and Suaeda fruticosa growing in tannery contaminated site in Kasur. They have the capability to tolerate a high level of chromium (3000 µg/ml) in nutrient agar medium.These strains were characterized morphologically, biochemically physiologically. The cells of most of these strains were gram +ve and aerobic rods. They showed phosphate solubalization in NBRIP medium and also produced indole acetic acid (IAA) both in the absence and presence of chromium. These strains also demonstrated substantial chromium reduction. The effect of different environmental factors such as temperature, pH, chromium concentration and other metallic ions on chromium reduction by these strains was also determined. These bacterial strains can be employed for the bioremediation of chromium contaminated soils as well as promoting plant growth in chromium contaminated soils.

Keywords: Chromium, Bioremediation, NBRIP medium

BPC-22

Fungal Bio-Pesticide: An Eco-friendly Solution of Crop Diseases

Fathia Mubeen

National Institute for Biotechnology and Genetic Engineering (NIBGE), Faisalabad, Pakistan E-mail: fmghafoor@hotmail.com

Abstract:

Newly emerging biocontrol approach is the replacement and/or reduction of the use of agrochemical through biological pesticides. It is a worldwide reported effective control strategy against phytopathogens causing various crop diseases. Biopesticides are based on the thought that nature always balanced itself, if there are some harmful organisms in form of pests to spoil the crop then same time there are organisms which are beneficiary and predator to the first one and thus maintaining the balance. In Australia "Fairway patch" disease of turfgrass was appeared in recent past several golf courses. The causal agent of this disease was a non-sporulating fungus with very dark brown to black mycelium. Symptomatically the disease appeared as the patches of unhealthy grass, exhibiting a brown discoloration of the leaves, stunting and eventual death, especially on the borders of patches but the centre of the patches remain green. Present study was planned to identify particular microorganisms that live naturally in the turfgrass root system and may responsible for controlling the pathogen and can be use to develop as a biopesticide against this disease. Various fungal strains were isolated from the healthy green centers of the diseased patches randomly collected and screened for their antagonistic efficiency against the possible causal agents of the disease. Efficient fungal biocontrol agents were identified and characterized as Trichoderma ssp.

Keywords: Trichoderma ssp., Biopesticides, Phytopathogens

BPC-23

Antibacterial Effect of *Aloe vera*on cell wall constituents and biofilm formation of bacteria isolated from dental unit water lines

Sumreen Hayat and Anjum Nasim Sabri

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-i-Azam Campus, Lahore 54590, Pakistan.

Astract:

The increasing resistance to traditional antibiotics exhibited by common pathogens has led to renewed interest in the discovery of novel anti-infective compounds. The present study was aimed at the screening for antibacterial activity of *Aloe vera* leaf gel. Agar well diffusion method indicated antibacterial effect of various extracts of plant in terms of diameter of zone of inhibition. Ethanol extract was more effective (diameter of zone of inhibition 22.0 mm for *B. cereus*) as compared to other extracts. Effect of heat treatment on the antibacterial potential of *Aloe vera* was also monitored. Peptidoglycan and teichoic acid content was reduced in cells treated with gel while diaminopimelic acid content was increased in gel treated cells. Microtitre plate assay had indicated the anti-biofilm effect of *Aloe vera* gel after 72 hrs and 120 hrs. Filtered gel was more effective as compared to heat treated gel.

Keywords: Aloe vera, Peptidoglycan, Teichoic acid, Biofilms

BPC-24

Bio-control effect of Eruca sativa Mill oil against the hazardousfood borne pathogens

Uzma Bashir, Amna Ali, and Muhammad Saleem Haider
Institute of Agricultural Sciences, University of the Punjab,
Quaid-i-Azam campus, Lahore- 54590 Pakistan
Email:uzmamppl@yahoo.com

Abstract:

Antimicrobial activity of *Eruca sativa* Mill oil was evaluated against phytopathogenic bacterial species (*Xenorhabdus luminescens, Acinetobacter sp., Bordetella pertussis, Ensifer adhaerens, Pseudomonas syringae, Acidovorax temperans, Xanthomonas axonopodis*) and fungal species (*Alternaria alternate, Dreschlera halodes, Aspergillus nidulans, Acremoniun kiliense, Fusarium oxysporum, Curvularia clavata, Rhizopus oryzae*). Antifungal activity was determined on MEA while antibacterial activity on NA media plates to measure the effects of oil. The antimicrobial activity was tested by well diffusion method *in vitro. E. sativa* oil was found to be highly active against all fungal isolates tested as compared to bacterial isolates. Results showed evidence of high antibacterial activity against *X. luminescens* with inhibition zone of 3.1 cm. *E. adhaerens* and *A. temperans* had least resistanceagainst oil with 1.4 cm and 1.7 cm zone of inhibition respectively. The oil showed high antifungal activity in the range of 6.0-6.8 inhibition zone against *D. halodes, C. clavata,R. oryzae* and *A. nidulans* whereas least active against *F. oxysporum* with 1.1 zone of inhibition. The antimicrobial components from this oil can be used as an alternative to develop novel pesticides by replacing some chemical commercial antifungal and antibacterial for the plant diseases.

Keywords: E. sativa oil, Antimicrobial activity, Phytopathogens

BPC-25

Prevalence of Methicillin and Vancomycin resistant *Staphylococcus aureus* among cancer patients of Lahore

Qandeel Fatima and Numan Javed

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-i-Azam Campus, Lahore-54590, Pakistan. Email: qandeel_mmg _@yahoo.com

Abstract:

Methicillin resistant *Staphylococcus aureus* (MRSA) is an aggressive Gram positive pathogen that is resistant to first-line antibiotics. MRSA is currently a leading cause of blood stream infections in patients with cancer. A complication rate of 33%, with mortality of 15%, was seen in a study of children with cancer. The focus of this study is to find out the prevalenceandassociated risk factors of MRSA and VRSAamongcancer patients from different hospitals of Lahore. In addition, we also reviewed choices of antibiotics used to treat it. Skin and nasal swabs from 47cancer patients, from various hospitals of Lahore i.e. Jinnah hospital and INMOL hospital were collectedduring the period of August 2012 to February 2013. The sampling was done with informed consent. Of the 47 samples (23 males and 24 females) tested, 27(57%) had MRSA and 3(6%) had VRSA. Among 23 males, 15(65%) were carrier of MRSA while of the 24 females, 12(50%) were positive forMRSA.Among 27 MRSA acquired patients, 20 patients were on chemotherapy and 23 hadsurgery. Out of 17 leukemic patient 11 had acquired MRSA infection.Our findings suggested an increasing trend in prevalence of MRSA and VRSA

amongcancer patients from different hospitals of Lahore. We observed that Leukemic, chemotherapeutical and surgical patients are more prone to carry MRSA. We also found that that MRSA and VRSA is equally prevalent among both genders.

Keywords: MRSA, VRSA, cancer, Chemotherapy, Leukemia

BPC-26

Biocontrol activity of *Pseudomonas aeruginosa* for the suppression of *Xanthomonas oryzae* pv. *Oryzae*: The causal agent ofrice bacterial blight

Sumera Yasmin

National Institute for Biotechnology and Genetic Engineering (NIBGE), Faisalabad, Pakistan. Email: sumeraimran2012@gmail.com

Abstract:

Xanthomonas oryzae pv. oryzae (Xoo) is the causal agent of bacterial blight of rice. The significant damage caused by this disease need the development of strategies to control it with eco-friendly means. Xoo strains isolated from diseased leaves were identified and characterized using light microscopy and biochemical tests. The virulence of Xoo strains was confirmed by in vivo pathogenecity tests. More than five hundred bacterial strains were screened in vitro for antibacterial activity using hole plate diffusion method. In vivo evaluation of selected bacterial strains antagonistic to Xoo was carried out at NIBGE Net House as well as at NIAB, Faisalabad in collaboration with "Plant Protection Division". Antagonistic strains were studied for different biocontrol and growth promoting mechanisms. Bacterial strain Pseudomonas aeruginosa BRp3 showed effective and consistent pathogen suppression in all the three pot experiments and in a field experiment at NIAB as well, conducted with different strains of pathogen. A field experiment was also carried out at NIBGE to test the potential agonistic bacteria BRp3 for improving growth and yield of rice variety Super Basmati. Immunofluorescence marker was employed in combination with confocal laser scanning microscopy for detection and colonization of BRp3 strain with rice plant. The present study resulted in the selection of Pseudomonas aeruginosa BRp3 as a potential biocontrol agent promoting rice growth with reduced disease incidence. This strain can also be used along with other strategies to achieve greater levels of protection and to sustain rice yields.

Keywords: Xanthomonas oryzae, Rice yields, Immunofluorescence marker

BPC-27

Antimicrobial activity of *Ocimum basilicum* against *Photobacterium* isolated from Indian Mackerel

Werda Zia, Anjum Nasim Sabri and Sumreen Hayat
Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, University of the
Punjab, Lahore-54590, Pakistan.

Abstract:

Medicinal plants constitute the base of health care systems in many societies. Different plants and their extracts are in use now-a-days as a source for treatment of various diseases due to presence of bioactive compounds. Basil is one of the oldest spices belonging to the *Ocimum* genus and to the Lamiaceae (Labiatae) family, which contains herbs and shrubs from the tropical and subtropical regions of Asia, Africa, and Central and South America. The antimicrobial efficiency of *Ocimum basilicum* (leaf and inflorescence extracts), was examined using methanol,

ethanol, and water, as solvents and tested against *Photobacterium leiognathi* subsp. *mandapamensis*, *Photobacterium leiognathi* and *Photobacterium leiognathi* subsp. *leiognathi* by using the agar well-diffusion method as well as minimum inhibitory concentration and minimum bactericidal concentration was determined. All extracts showed significant activity against the selected strains, but the methanolic extract showed the maximum zone of inhibition and minimum inhibitory concentration against all the three selected strains. The phytochemical analysis and Thin Layer Chromatography (TLC) was also performed, which confirms the presence of alkaloids and coumarins in extracts. The effect of crude plant extracts on protein leakage from bacterial cells showed that crude plant extracts had killing potential by inhibiting the growth as well as by disruption of cell integrity by protein leakage and resulted in loss of bioluminescence activity of *Photobacterium* sp. The crude plant extracts also showed antiangiogenic effect when growth and development of chick embryo was observed.

Keywords: Thin Layer Chromatography, Crude plant extracts, Well-diffusion method

BPC-28

Expression Analysis of Cellular Genes *HMGR* and *FAS* Involved in Steatosis in HCV Patients of Genotype 1a and 3a

Muhammad Aiman Shahzad and Saqib Mehmood
Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the Punjab, Lahore-54590, Pakistan.

Abstract:

Hepatitis C virus (HCV) has become a major threat for global health. HCV is genetically highly variable and exist as different genotypes and subtypes the severity of effect within subtypes of a major genotype was observed different. So it is necessary to find the role of HCV genes of different genotypes in HCV induced pathogenesis. HCV induced Steatosis, triglyceride accumulation in hepatocytes, is the most frequent cause of abnormal liver function. Cellular genes FAS and HMGR which are involved in the Fatty acids synthesis have been reported to be activated in HCV induced steatosis. To unfold the effect of HCV of different genotypes, we studied the expression level of cellular genes, mRNA expression of genes FAS and HMGR was increased in HCV 3a patients as compared to HCV 1a patients. RNA was extracted from the whole blood. cDNA was formed from the RNA by RT-PCR. Cellular gene expression analysis was done by using specific-primers of cellular genes on BIO-RAD iQTM5 Multicolor Real Time PCR Detection System. The enhanced expression of cellular genes FAS and HMGR in patients HCV is inducing steatosis. And these genes are expressing more in patients of HCV genotype 3a then the patients of HCV genotype 1a.

Keywords: Hepatitis C virus, Steatosis, Real Time PCR

BPC-29

Algal biomass production and its evaluation as biofertilizer – An improved cash crop production

Shazia Kanwal Malik and Hamad Ashraf
Department of Botany, Government College of science, Wahdat Road Lahore, Pakistan
Email: kanwal707@yahoo.com

Abstract:

The present study is concerned with the culturing of algae and its evaluation as biofertilizer in the improvement of *Lycopersicon esculentum* Mill.(tomato). Algae were collected from different localities of Lahore. The algae was identified and found to be *Hydrodictyon* and *Spirogyra*. In a series of experiments, *Hydrodictyon* was selected for the culturing in the laboratory. The algae were cultured in 1000ml beakers and then 1cubic foot aquarium. Different carbon and nitrogen sources were evaluated for the production of algal biomass. Of all the nitrogen and carbon sources tested, urea along with cellulose exhibited maximum production (9.5g/100ml) of biomass. The optimal biomass was incorporated in the soil as biofertilizer. Different plots were prepared and *Lycopersicon esculentum* was grown in the plots having control, urea, animal dung and algae. The significant growth of tomatoes was observed in the plots in which algae was used as a biofertilizer. Further experiments will be carried out to estimate the amount of tomato production (wt) in the respective fields.

Keywords: Biofertilizer, algae, Lycopersicon esculentum, Hydrodictyon, Spirogyra.

BPC-30

Molecular epidemiology of methicillin resistant *Staphylococcus aureus* based on *Spa* typing from local hospitals of Lahore

Anam Tariq and Numan Javed

Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the Punjab, Lahore-54590, Pakistan.

Abstract:

Staphlococcu saureus has become the commonest nosocomial infectious agent throughout the world, causing a wide range of hospital infections. Epidemiological studies suggest that hospitals of all sizes are facing the problem of the resistant form of Staphlococcus aureus that is Methicillin Resistant Staphlococcus aureus (MRSA). This problem seems to be increasing irrespective of hospital size and control measures for MRSA. Since there is less significant data available regarding the prevalence of MRSA in Pakistan so a thorough knowledge of the dissemination and the molecular epidemiology of MRSA strains are required to develop effective strategies to control MRSA. Protein A of Staphylococcus aureus is a virulence factor whose encoding gene, spa, shows a variation in length among different strains of MRSA. In this study the spa gene variation in methicillin resistant S. aureus was studied. Sixty strains of MRSA were collected from the local hospitals of Lahore. The identification and susceptibility testing of MRSA isolates was done by using DNase testing and Kirby Bar disk diffusion testing. The DNA isolation was done by CTAB method. Polymorphic X region of the spa gene was amplified using specific primers of spa gene through PCR. MRSA strains which showed a distinct band between 200 bp to 400 bp was considered as a distinct spa type. Then the sequencing of amplified X region of spa gene would be performed. Those sequences would be analyzed by the Spa Ridom

database and would be served as a rapid diagnostic tool for the identification of MRSA during epidemic.

Keywords: Staphlococcu saure, Kirby Bar disk diffusion testing, MRSA

BPC-31

Antifungal activity of *Calotropis procera* against phytopathogenic fungus (*Macrophomina phaseolina*)

Nidra Waheed, Khajista Jabeen and Sumera Iqbal
Department of Botany, Lahore College for Women University, Lahore, Pakistan
Email: khajista_1@hotmail.com

Abstract:

The antifungal activity of *Calotropis procera* (aak) was investigated against the phytopathogenic fungus Macrophamina phaseolina causes charcoal rot in various economically important crops. Different concentrations of leaf and stem methanol extracts viz. 1% 2% 3% 4% 5% was applied against Macrophamina phaseolina in vitro. Leaf extract was found more effective & showed significant antifungal activity as its 3% concentration maximum reduces the fungal growth diameter i.e, 16.5 %. Methanolic extracts of C. procera stem was promoting the growth of test fungus except 5% concentration. Calotropis procera leaf extract was effectively suppressing the growth of M. phaseolina in screening bioassays, so this was subjected for fractional guided bioassays. Methanolic extract of C. procera leaves was partitioned with n-hexane; chloroform (CHCl₃) followed by ethyl acetate (EtOAc) and n-butanol at room temperature by using separating funnel. Minimum inhibitory concentration (MIC) of these fractions and a commercial reference fungicide (Puslan 72 WP) was evaluated against Macrophomina phaseolina. Different concentrations from (700mg-1.36mg) were used for MIC bioassay, and data was recorded after 24 and 48 hrs. The tested fractions were showed variable antifungal activity when compared with control sets (Water & DMSO) .n-hexane was most effectual in retarding conidial germination with (1.36 mg) MIC and this fraction was also effective than commercial fungicide (Puslan 72 WP) as this fungicide. The other fractions were comparatively less antifungal.

Keyword: Macrophamina phaseolina, Calotropis, Antifungal

<u>BPC-32</u>

Analysis of antibacterial compounds produced by *Pseudomonas* sp. and *Bacillus* sp. under variable nutrient conditions against *S. aureus*

Rida Rashid and Nazia Jamil

Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the Punjab, Lahore-54590, Pakistan.

Abstract:

A wide range of bacteria produce different types of antimicrobial compounds including antibiotics and antimicrobial peptides i.e. Bacteriocins. In present study bacterial strains belonging to genus *Pseudomonas* and *Bacillus* were collected from MMG stock and screened for their bacteriocin production against *S. aureus* by culturing them in tryptic Soy Broth, Brain Heart Infusion and Nutrient-Broth.Maximum bacteriocin activity was observed in TSB medium in case of *Pseudomonas* while BHI was proved to be best medium for bacteriocin production in case of *Bacillus* 48 hours of incubation. *Pseudomonas* as well as *Bacillus* produced bacteriocins that have bacteriolytic effect on *S. aureus*. Ethyl acetate extract of *Bacillus* was sensitive to 121°C

temperature treatment, exposure to ultraviolet radiation and proteinase K treatment while in case of *Pseudomonas*, ethyl acetate extract retained its activity at all temperatures (60°C, 100°C, 121°C), ultraviolet radiation and proteinase K treatment. Antagonistic activity of all bands obtained from thin layer chromatography showed that one iodine active band, Rfvalue 0.96, in case of *Pseudomonas* produced 4 mm zone of inhibition while in case of *Bacillus*, iodine active band having Rf value 0.76 produced 2 mm zone of inhibition against *S. aureus*. Estimation of bacteriocin proteins by Bradford assay was also done. Qualitative analysis of bacteriocins by SDS-PAGE revealed that *Pseudomonas* produced 2 antibacterial peptides, 55kDa and 70kDa, while *Bacillus* produced 70kDabacteriocin. The results of 16Sr RNA sequencing revealed 99% homology of *Pseudomonas* with *Pseudomonas aeruginosa* while 97% homology of *Bacillus* with *Bacillus thuringiensis*.

Keywords: Antimicrobial compounds, *Pseudomonas*, Bacteriocin

BPC-33

Association of ACE I/D Polymorphism with Type 2 Diabetes mellitus

Mahwish Asif Ali and Nageen Hussain

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore-Pakistan Email: mahwishasifali@yahoo.com

Abstract:

Angiotensin Converting Enzyme levels differ in carriers with respect to their genotype; DD carriers have twice the levels of ACE than that found in II genotype individuals. The insertion/deletion (I/D) of a 287bp long *Alu* repetitive sequence in intron 16 is responsible for three genotypes, *DD* and *II* homozygotes and *ID* heterozygotes. However, individuals with I/D genotype show codominancy with intermediate levels of serum. The main purpose of this study is to investigate Angiotensin Converting Enzyme (ACE) gene (I/D) polymorphism in Pakistani type 2 diabetes mellitus patients primarily from Lahore. Hundred patients (type 2 diabetic) and fifty healthy controls were enrolled in this study. The ACE I/D polymorphism, located in intron 16, was analyzed by a triple primer method called nested-PCR. The frequency of ACE DD, ID and II genotypes among the patients with type 2 diabetes mellitus was found to be 77%, 9%, 14% whereas in control subjects, 38%, 4%, 8% respectively. Thus the frequency ACE DD genotype was found to be significantly higher in Type 2 diabetes mellitus patients as compared to controls.

Keywords: Angiotensin Converting Enzyme, Polymorphism, Nested-PCR, Genotypes

BPC-34

Cassia fistula as biocontrol agent against blight of chick pea (Cicer arietinum L.)

Bareera Khan, Khajista Jabeen and Sumera Iqbal
Department of Botany, Lahore College for Women University, Lahore, Pakistan
Email: khajista_1@hotmail.com

Abstract:

Cassia fistula L. was examined for its *in vitro* antifungal potential against Ascochyta rabiei (Pass) Lab., the cause of blight disease of chickpea (Cicer arietinum L.). Screening bioassay with methanolic leaf and bark extracts were performed *in vitro* against A.rabiei. This bioassay showed that leaf extract of Cassia fistula exhibited strong antifungal activity over the bark extracts, as 1% concentration retarded the A. rabiei diameter by 67.67%. This part of Cassia fistula was used

in fractional guided bioassays due to its high antifungal activity. Methanolic leaf extract was partitioned between n-hexane, chloroform, ethyl acetate and n-butanol. The minimum inhibitory concentration (MIC) of the four isolated fractions and a commercial synthetic fungicide Metataxyl + Mancozeb, 72 WP was investigated against *A. rabiei*. Different concentrations of isolated fractions and fungicide ranging from (500 mg - 0.976562 mg) were used in MIC bioassay, and the data was recorded after 24, 48 and 72 hrs. Results of these isolated fractions revealed the fact that chloroform fraction of the methanolic extract of *C. fistula* leaves was found highly effective against *A. rabiei* with MIC value of 0.976562 mg. Fungicide concentrations highly inhibited the fungal growth germination even after 72 hrs incubation period. Chloroform was found to be the more effective followed by ethyl acetate & n-hexane, while n-butanol was least effective.

Keywords: Cassia fistula, Cicer arietinum, Antifungal, MIC

BPC-35

Seroprevalence of dengue IgM antibodies in patients suspected of having dengue fever

Faiza Asghar and Nageen Hussain

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore-54590, Pakistan

Email: Nageen1704@hotmail.com

Abstract:

Dengue is a growing public health problem globally infecting about 50-100 million people annually. Dengue is now endemic in more than 100 countries, including Pakistan. This study was carried out to find out the seroprevalence of Dengue IgM antibodies in the patients suspected of having dengue fever in Lahore. This study was conducted in Jinnah hospital and Sir Ganga Ram hospital from September to November 2012. About 90 serum samples were tested from suspected dengue patients for IgM antibodies by using antibody capture ELISA (NovaLisaTM catalog # DENG 0120). Of the 90, 68(75.5%) were males and 22 (24.4%) were females. Mean age at the time of diagnosis is 35.2 years. In this study, 43 (47.78%) patients were confirmed as Dengue IgM positive and 47 (52.2%) patients as Dengue IgM seronegative. Fever with headache (81.1%), loss of appetite (91%), rashes (48.8%), nausea (54%), myalgia (72.2%), retro-orbital pain (88%) were the common symptoms found in suspected dengue virus infection cases. Laboratory investigations showed low platelet count (<150.0x10³) in 60%, leucopenia (<4.0 x10³) in 58.8% cases. The most affected age group was 15 to 25 years of age. It was also observed that dengue is not a gender biased infection.

Keywords: Dengue virus, Dengue IgM antibodies, Leucopenia, Myalgia, ELISA

BPC-36

Optimization of yogurt starter cultures: their preservation and bacteriocin producing potential

Fatima Ameer, Mehboob Ahmed and Shahida Hasnain
Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the
Punjab, Lahore-54590, Pakistan

Abstract:

Starter cultures are the backbone of dairy industry. A number of bacterial strains are already known that play crucial roles as starter cultures but still there is an ever increasing demand of

more strains that could produce better fermented products. Bacterial strains were isolated from curd (dahi) samples and commercially available yogurts. The purified isolates were screened for yogurt formation and bacteriocin production. The isolates were preserved by lyophilization and then they were used for yogurt production again, so as to determine their ability to survive freeze-drying conditions. Bacteriocin production was checked by stabbing and Agar-Well Diffusion method. The strains showed maximum bacteriocin activity at 96hours of incubation. The extracted supernatants of the bacterial isolates revealed bands at 310 nm that showed bacteriocin activity. Extracted DNA of all the isolates and was amplified for 16SrRNA gene and was sequenced. Using NCBI-BLAST, the resulting sequences were identified as Lactobacillus sp. SWM, Lactobacillus murinus, Lactobacillus murinus strain AU06, Bacillus tequilensi strain J8B-72, Lactobacillus sp., Bacillus subtilis strain EXWB4-09, Bacillus amyloliquefaciens subsp. plantarumstrain. They were checked for homology and the dendrograms were drawn which showed that isolated Lactobacillus strains were highly related to Lactobacillus species that are commonly involved in different fermentations. Whereas the resulting Bacillus strains were found to be more closely related to Bacillus cereus and Bacillus subtilus. The bacterial supernatants were also analyzed by Bradford method for protein estimation and maximum protein concentration was found in the supernatant of *Bacillus amyloliquefaciens* subsp. *plantarum* strain. The best yogurt was formed by a mixed culture of Lactobacillus murinus, Lactobacillus sp. and Bacillus subtilis strain containing about 6 X 10⁹ bacterial cells. The maximum bacteriocin activity was observed in case of Lactobacillus murinusand Bacillus amyloliquefaciens. These strains were capable of giving improved yogurt with less whey content and thus can be used as indigenous starter cultures in industries.

Keywords: Starter cultures, Bacteriocin production, Yogurt

BPC-37

$Seroprevalence\ of\ dengue\ IgG\ antibodies\ in\ patients\ suspected\ of\ having\ dengue\ fever$

Kanwal Ishfaq and Nageen Hussain

Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the Punjab, Lahore-54590, Pakistan

Email: nageen1704@hotmail.com

Abstract:

Dengue fever is a mosquito-transmitted viral disease caused by one of the four closely related dengue virus serotypes. This has increased dramatically in the recent year's affected about 2.5 million people over 40% of the world population. In the present study, a seroprevalence of Dengue IgG antibodies was measured in the patients suspected of having dengue fever. Dengue IgG antibodies were measured by using antibody capture ELISA (NovaLisaTM Cat # DENG 0120). A total of 90 patients admitted in especially established general public ward for dengue fever patients at combined military hospital (CMH) and Mayo hospital Lahore (September to November 2012) were included in this study. Of the 90, 45 (50%) were males and 45 (50%) were females. Mean age at the time of diagnosis is 32.8 years ± 15.1.In this study, 83 (92.99%) patients were confirmed as Dengue IgG positive and 7 (7.78%) patients as Dengue IgG seronegative. Fever with headache (100%), loss of appetite (91%), rashes (3.3%), nausea (54%), myalgia (87%), retro-orbital pain (88%) were the common symptoms found in suspected dengue virus infection cases. Frequencyof Dengue IgG seropositive (92.99%) cases indicated the presence of secondary infection, past dengue. Thus are at greater risk of Dengue Hemorrhagic

Fever (DHF) from a subsequent infection which is the most critical form. It was also observed that dengue is not a gender based infection.

Keywords: Dengue, myalgia, antibodies, ELISA, Dengue Hemorrhagic Fever

BPC-38

Isolation and characterization of lytic bacteriophages against pathogenic bacteria

Komal Amer and Shafiq ur Rehman

Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the Punjab, Lahore-54590, Pakistan.

Abstract:

The hospital acquired infections are becoming a major challenge for patient care due to emergence of resistance against commonly used antibiotics. Today, phage therapy can replace antibiotic treatment, which has been a cause of emergence and rapid spread of antibiotic resistance. Bacteriophages are the bacterial viruses that either can lyse (lytic phages) the bacterial cell or can integrate their genome in the bacterial genome (lysogenic phages) during their life cycle. The lytic phages and their gene products can easily be used as therapeutic agents against bacteria as they are host specific and show no side effects. In the present research work, bacteriophages of different plaque morphologies were isolated against Enterobactercloacae, Klebsiella pneumonae and Pseudomonas aeruginosa from the sewage water and hospital effluent samples. These bacteriophages showed lysis activity against the clinical isolates and exhibited narrow host range. The lytic activity of these bacteriophages at varying temperatures and pH establish their wide stability range while highest viability was observed at 37°C and pH 7.0. The Enterobacter phages TSE1, TSE2 and TSE3, the Klebsiella phages TSK1, TSK2 and TSK3, and the *Pseudomonas* phages JHP1 and JHP3 were viable at high temperatures till 60°C. While the Klebsiella phage TSK1 worked at an alkaline pH of 9.0. All the phages efficiently reduced bacterial growth in the bacterial reduction assay. The Pseudomonas aeruginosa phages predominently maintained their lytic phase of life cycle throughout the 24 hours. The Calcium ion majorly enhanced the adsorption rate of all the phages to their hosts except the Enterobacter phages TSE1, TSE2 and TSE3. The proper manipulation of these highly active phages, their extracted genome and protein analysis can be an ultimate key to their better application in phage therapy. As the initial low dose can eradicate the bacterial infection locally so, it signifies the underlying potential of the bacteriophage therapeutics.

Keywords: Antibiotic treatment, Bacteriophage therapeutics, Calcium ion

BPC-39

Leptin replacement therapy: A treatment of childhood obesity

Warda Fatima¹ and Saqib Mahmood²

¹ Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore.

Leptin is a hormone that is formed in the white fat tissues in humans and helps in regulation of the appetite. Leptin regulates food intake and energy expenditure by acting on the two populations of arcuate neurons in hypothalamus, these are POMC/CART neurons that are activated by action of leptin; and AgRP/NPY neurons that are inhibited by leptin. Congenital leptin deficiency is a rare human genetic condition clinically characterized by hyperphagia and

² Department of Human genetics and Molecular biology, University of Health Sciences, Lahore. **Abstract:**

acute weight gain usually during the first postnatal year. This condition arises when the leptin cannot be formed or secreted in blood to function in form of normal protein. Till now six pathogenic mutations have been reported in the *leptin* gene that render the protein inactive or nonfunctional. Childhood obesity due to leptin deficiency is more common in Pakistani population compared to rest of the world as most of children presented with congenital leptin deficiency are from Pakistani origin (75 % 21of 28 total reported cases). The condition of leptin deficiency is resolvable with the exogenous leptin replacement therapy. Trials of human recombinant leptin replacement are under process and so far yielded successful results in reducing weight, controlling appetite and enhancing immune and endocrine function. Nine children diagnosed with leptin deficiency in Pakistan have been selected to get this trial treatment in Germany.

Keywords: Congenital Leptin deficiency, Monogenic obesity, *Leptin* gene

BPC-40

Assessing the Phytotoxicity of Metal Contaminated Soil on Helianthus annuus Growth

Muhammad Yasin and Muhammad Faisal

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-i-Azam Campus, Lahore-54590, Pakistan

Email: yasin_mmg@yahoo.com

Abstract:

Here we reported the effect of four strains *Bacilluspumilus*-CrK08, *Cellulosimicrobiumcellulans*-CrK16, *Exiguobacterium*-CrK19and *Bacillus cereus*-CrK20 and tannery contaminated soil on *Helianthus annuus* L. var Hysun-33 growth parameters. Plants growing in tannery effluent contaminated soil have shown slowed leaf growth, reduced shoot length, burning of leaf margins and tips compared to plants growing in normal garden soil. The inoculated plants had shown overall increase in root length (14%), shoot length (30%) and fresh weight shoot (125%) compared to un-inoculated plants growing in stress conditions. Plants growing in tannery contaminated soil have shown increase in soluble proteins contents (9%), acid phosphatase activity (200%), peroxidase activity (203%) and decrease in chlorophyll a (39%), chlorophyll b (20%) and carotenoids contents (28%) compare to plants growing in normal control soil. Inoculated plants grown in contaminated soil have shown an increased in peroxidase activity, soluble proteins contents, acid phosphatase activity, chlorophyll a, b and carotenoid contents compare to respective un-inoculated plants. Heavy metal contents in soil and plants samples were determined by ICP-OES.

Keywords: Tannery, Chromium, Helianthus annuus, Bacteria

BPC-41

Efficacy of Rhizobacteria to control Fusarium wilt of tomato

Anam Imtiaz and Basharat Ali

Department of Microbiology and Molecular Genetics, University of the Punjab Quaid-e-Azam Campus, Lahore-54590, Pakistan

Email: ali.basharat@yahoo.com

Abstract:

A study was carried out to screen *Bacillus* strains for their ability to induce systemic resistance against *Fusarium* wilt of tomato. Ten *Bacillus* strains were used for initial screening under

greenhouse conditions. *B. megaterium* ZmR-4 was the most effective to control the severity of *Fusarium* wilt. Basis of induced resistance were elucidated by biochemical, histochemical and molecular analysis in an independent experiment. In biochemical studies, total phenolics and enzymes involved in phenylpropenoid pathway viz: Peroxidase (PO), Polyphenoloxidase (PPO) and Phenylalanine ammonia lyase (PAL) were quantified in a time course manner. Calorimetric assays proved significant higher levels of these defence related biochemicals in tomato plants. In histochemical analysis, intense localization of lignin and peroxidase were observed in tomato roots treated with this bacterial strain as compared to untreated control. During Semi-Quantitive RT-PCR analysis, higher expression levels of defence related genes were noted in tomato plants treated with *B. megaterium* ZMR-4.Our study clearly indicates the importance of *B. megaterium* ZMR-4 for suppression of *Fusarium* wilt and growth promotion in our agriculture system.

Keywords: Induced systemic resistance, *Bacillus metagerium*, *Fusarium oxysporum*, Phytostimulatory effects and Plant growth promoting rhizobacteria

BPC-42

Isolation and Characterization of chlorhexadine resistant bacteria from dental caries and different biofilm formation assays

Nabiha Mansur and Anjum Nasim Sabri
Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam
Campus, Lahore-54590, Pakistan

Abstract:

Dental plaque is the result of complex interaction of bacteria resulting in the biofilm formation, which may lead to serious infections such as caries. Predominant microorganisms found in the dental biofilm formation are mostly gram positive cocci primarily the streptococci species other bacteria such as Lactobacillus and Actinomycetes are also reported in the later stages of plaque formation. Chlorhexidine gluconate is an active ingredient of most of the mouthwashes used for the removal of dental plaque. Due to its long term use most of the bacterial species have adapted resistance to it therefore the present study aimed to isolate and characterize the bacteria involved in the dental plaque, to determine their resistance against chlorhexidine, to determine their biofilm forming ability. Efforts were also made to characterize EPS by spectroscopic methods. Dental plaque samples taken from the patients from the clinic were processed for the isolation of different bacteria. Later their resistance against chlorhexadine was checked at different concentrations. Biofilm formation was determined both quantitatively and qualitatively both in the presence and absence of chlorhexidine. EPS was also extracted and FTIR analysis was done. By biochemical and molecular characterization the bacterial isolates were characterized as Streptococcus salivarius, Neisseria flavescens, Erwinia pyliforiae and Pseudomonas stutzeri. All the strains isolated were resistant to chlorhexidine and they showed biofilm formation ability. The EPS characterization results showed that EPS matrix was made up of different biomolecules such as carbohydrates and proteins.

Keywords: Biofilm, Dental plaque, Chlorhexidine

BPC-43

Effect of carbon and nitrogen sources on biomass production of Spirogyra (L)

Orazi Javed¹, Firdous e Bareen² and Hamad Ashraf¹
¹Biotechnology Research Laboratory, Botany Department, Govt. College of Science,
Wahdat Road. Lahore.

²CEES, University of the Punjab. Lahore.

Abstract

The present study is concerned with optimization of different Carbon and nitrogen sources for biomass production of *Spirogyra (L)*. The Spirogyra was collected from different localities of Lahore. The experiment was carried out in 1 L glass beaker and then on 1 square feet aquarium. The different carbon sources (Lactose. Glucose. Cellulose and Starch) were evaluated for biomass production. Among all the sources tested cellulose was found to be best for optimal production of *Spirogyra* (5.81 g/100ml). The different nitrogen sources (Ammonium nitrate, Ammonium sulphate. Ammonium chloride. Potassium nitrate and urea) were evaluated for biomass production. Among all the sources tested urea was found to be best for optimal growth of Spirogyra (6.86g/100ml). Significant result (7.97g/100ml) was obtained when 0.1% Cellulose and 0.005% Urea was added in the culture medium. The maximum biomass (8.0g/100ml) was obtained after 20days of culturing period.

Keywords. Spirogyra. Carbon sources, Nitrogen sources, Biomass

BPC-44

Biofilm formation and characterization of bacterial isolates from Otitis media under stress conditions and molecular detection of adhesion genes

Saba¹, Anjum Nasim Sabri¹ and Shahbaz Mujtaba Ghauri²

¹Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus, Lahore-54590, Pakistan. Mayo Hospital/ King Edward Medical University, Lahore-54590, Pakistan

Abstract:

The present study was aimed to check the biofilm formation potential of the bacterial strains isolated from otitis media patients. In addition, to determine their cell surface hydrophobicity and the effect of different plant extracts and antibiotics on biofilms. Furthermore, to investigate the presence of intercellular adhesion (ica) genes which encodes production of adhesins that mediate adherence to biomaterials. Ear swab samples taken fromOtitis media patients at Mayo Hospital Lahore were processed to isolate bacterial strains. Antibiotic profiling of these strains was performed and their response towards natural plant extracts was also noted. Cell surface hydrophobicity and adhesive properties of the strains were evaluated. Biofilm formation was determined both qualitatively and quantitatively in the presence and absence of antibiotics and plant extracts. *ica* genes are going to be amplified from these strains by using polymerase chain reaction (PCR). Pseudomonas aeruginosa, Staphylococcus haemolyticus and Staphylococcus hominis were isolated. Staphylococcus sp. showed slime production and among various plant extracts tested, Acacia arabica gave significant antibacterial characteristics. In general, all the bacterial strains studied showed biofilm forming potential and cell surface hydrophobicity. Furthermore, ica genes will be targeted and amplified to study the association of biofilm formation with ica genes among these bacterial strains. Bacteria involved in middle ear infection showed strong biofilm formation potential that may lead to severe complications, thus making

them more difficult to be treated by antibiotics. Therefore, it is imperative to develop some advanced and effective treatments for these antibiotic resistant biofilm forming pathogens.

Keywords: Biofilms, Slime production, Adhesion, *ica* operon

BPC-45

Polyhydroxyalkanoates production from wastewater by mixed culture of *Pseudomonas* and *Stenotrophomonas*

Sajida Munir and Nazia Jamil

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore-54590, Pakistan.

Abstract:

Polyhydroxyalkanoates (PHA) are efficient, renewable and environment friendly polyesters. Various bacterial species accumulate intracellular polyhydroxyalkanoate (PHA) granules as energy and carbon reserves inside their cells. In this context, the present work describes the prospection and use of bacterial strains capable to bioconvert wastewater into polyhydroxyalkanoates (PHAs). In this work total of 30 bacterial strains were isolated from environmental samples. Screening for bioplastic production was done by Nile blue and Sudan black B staining. Biochemical analysis followed by ribotyping showed that PHA producing strains belong to *Pseudomonas*, *Bacillus* and *Stenotrophomonas* genera. On the basis of screening results two strains *Pseudomonas* and *Stenotrophomas* were selected for optimization and time profiling experiments with two different carbon sources; glucose and wastewater. Different percentages of wastewater were used at pH 6, 7 and 8 and incubation was given at 37 °C at 100 rpm. Wastewater used as a carbon source was analysed for different nutrients in it.

Keywords: Wastewater, Mixed culture, PHA

BPC-46

Effects of Bacillus subtilis and Halomonas aquamarina on growth promotion of Triticum aestivum and Brassica campestris

Zarwa Kamran and Anjum Nasim Sabri

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore-54590, Pakistan.

Abstract:

Plant growth promoting ability of the moderately salt tolerant strains: *Bacillus subtilis*, *Halomonas meridiana*, *Halomonas aquamarina* (1) and *Halomonas aquamarina* (2) were checked for IAA production, ammonia production, phosphate solubilization and HCN production. During field study, the seeds of *Triticum aestivum* and *Brassica campestris* were inoculated with bacterial suspensions and then allowed to grow till the plants matured and were then harvested. Biochemical tests (auxin content, protein content, peroxidase content, acid phosphatase content, chlorophyll and carotenoid content) were performed on six week old plants. The different growth parameters (root length, shoot length, spike length of *T. aestivum*, number of tillers of *T. aestivum*, number of pods per plant of *B. campestris*, number of seeds per pod/spike, weight of hundred seeds, leaf area of *B. campestris* and water content) of mature harvested plants were also checked. The strains were positive for IAA production, and ammonia production. The biochemical test showed better results for most of the monoculture and mixed culture as compared to control. It also was observed that both monoculture and mixed culture

showed better growth parameters as compared to control and hence enhanced growth and yield of both plant varieties.

Keywords: Salt tolerant strains, Biochemical tests, IAA production

BPC-47

Arsenic and Chromium Reduction in Co-Cultures of Bacteria Isolated from Industrial Sites in Pakistan

Yasir Rehman, Fariha Zakria Rizvi, Muhammad Faisal and Shahida Hasnain Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus, Lahore-54590, Pakistan.

Email: yasir.mmg@pu.edu.pk

Abstract

Arsenic (As) and chromium (Cr) are toxic metals which are being released in the environment by many industries in Pakistan. Exposure and ingestion of these metals can cause severe damage and cancers. Microbial bioremediation of such toxic contaminants is a cost-effective and environment friendly solution to reclaim polluted sites. As(V) and Cr(VI)-resistant bacteria were isolated from the industrial city Kasur, Pakistan. The 16S rRNA gene sequencing revealed that the highly resistant bacteria KS2-1, KS2-2, MWM81, and KSKE41 were related to *Bacillus* sp, *Rhodococcus* sp, *Cellulosimicrobium* sp.,and *Exiguobacterium* sp., respectively. KS2-1 reduced As(V) up to 94% and MWM81 reduced Cr(VI) up to 45%. Co-cultures of KS2-1 and KS2-2 reduced As(V) up to 98%, whereas co-cultures of MWM81 and KSKE41 reduced Cr(VI) up to 55%. Bacteria living in same niches could work together to degrade contaminants which were common toxicants for them.

Keywords: Chromium, Arsenic, PICT, Bacillus, Rhodococcus, Cellulomicrobium, Exguobacterium.

BPC-48

Simultaneous quantitation and monitoring of Rosuvastatin with NSAIDs by liquid chromatography with UV-detection

Arman Tabassum¹, M. Saeed Arayne¹ and Najma Sultana²

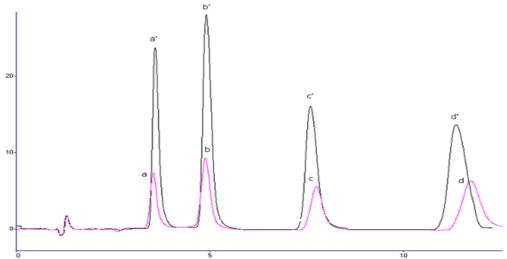
¹Department of Chemistry, University of Karachi, Karachi – 75270.

²Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Karachi, Karachi – 75270.

Abstract:

A simple, accurate, and sensitive high-performance liquid chromatography-ultraviolet detection method was developed for simultaneous determination of rosuvastatin with co-administered nonsteroidal anti-inflammatory drugs (meloxicam, ibuprofen, and mefenamic acid) in active pharmaceutical ingredient (API), pharmaceutical formulations and human serum. Isocratic separation was employed on prepacked Purospher Star C_{18} (5 µm, 25 × 0.46 cm) column at ambient temperature. The mobile phase consisted of methanol:water:acetonitrile (80:17.5:2.5 v/v), pH adjusted to 3.0 with *o*-phosphoric acid at 1 mL min⁻¹. The drugs in the eluant were monitored at isosbestic point of drugs at 230 nm. The method was compared by programming the detector adjusting the wavelength with time to match the individual analyte's chromophore which enhanced sensitivity with linear range. Linear behavior was observed between 0.1 and 2.5 µgmL⁻¹ for rosuvastatin, 0.4 and 10 µgmL⁻¹ for meloxicam, 0.25 and 6.25 µgmL⁻¹ for ibuprofen

and 0.15 and 3.75 μ gmL⁻¹for mefenamic acid, with $r^2 \ge 0.998$. The relative standard deviation for inter-day precision was 2 in API, formulations and human serum. Percent recovery for all drugs was 97.3%–100.89% in API and formulations and 99.3%–100.4% in human serum. Wavelength-programmed analysis made the method more sensitive, where 4 < limit of quantification (LOQ) < 11 and 1 < limit of detection (LOD) < 4 ngmL⁻¹for API; 6 < LOQ < 10 and 2 < LOD < 3 ngmL⁻¹for pharmaceutical formulations; and 3 < LOQ < 10 and 1 < LOD < 3 ngmL⁻¹in human serum, reduced from 9 < LOQs < 23 and 3 < LODs < 7 ngmL⁻¹for all drug analytes in API; and 4 < LOQs < 17 and 1 < LODs < 6 ngmL⁻¹in human serum recorded at isosbestic point for rosuvastatin, meloxicam, ibuprofen, and mefenamic acid respectively. Recovery of drugs was 99.998%–104.000% in all API, formulations and serum samples. The proposed method can be used for the quantitative analysis of these drugs in raw materials, in bulk drugs, dosage formulations and in human serum and for clinical studies even when the drug is present in low amounts.



Representative chromatograms of rosuvastatin (a & a'), meloxicam (b & b'), ibuprofen (c & c') and mefenamic acid (d & d') at isosbestic point and individual wavelength maxima respectively

Keywords: Meloxicam, ibuprofen, Mefenamic acid, Liquid chromatography, Quantitative analysis.

BPC-49

The study of pesticide biodegradation by using larvae of *Musca domestica* through bioassay *Michelle Maria and Farkhanda Jabeen*

Department of Botany, University of the Punjab, Quaid-i-Azam Campus, Lahore-54590. E-mail:rahimfarkhanda@hotmail.com

Abstract:

Chlorpyrifos IUPAC name: *O,O*-diethyl *O*-3,5,6-trichloropyridin-2-yl phosphorothioate, is a crystalline organophosphate insecticide. It acts on the nervous system of insects by inhibiting acetyl cholinesterase. Henceforth, this research was designed to study the effects of chlorpyrifos (pesticide) and its various biodegraded samples on the larvae of *Musca domestica* through bioassay. The production of larvae of *Musca domestica* is highly economical and simple, since

the ingredients used in this process are widely available. Medium for houseflies was prepared on Bunsen burner. Fifteen to twenty adult houseflies were introduced to each glass jar containing diet in form of slants which was at 45° covered with muslin cloth. After 3 to 4 days larvae were seen in each jar, which were used for the bioassay. Thin layer chromatography of samples was done to detect the biodegraded ability potential of the given samples. The samples with different time periods, zero day (T_0) 3 days, (T_1) 6 days (T_2) 9days (T_3) for incubation of samples1,2,3 and 4 of 500ul were introduced to medium /diet of 10 larvae separately. The mortality rate of the larvae was noted after 1 hour interval. The degraded samples had low relative front value and had insignificant effect on the larvae while the samples with higher relative front value had greater effect on the mortality rates because the highest value among these samples was of pure Chlorpyrifos.

Keywords: Pesticides, Larvae, TLC

BPC-50

In-silico hunt for efficient inhibitors of Dopamine Receptors (DRD2) to target Schizophrenia

Umara Shahzad, Mehboob Ahmed and Shahida Hasnain
Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-e-Azam Campus, Lahore-54590, Pakistan.
Email: mehboob.mmg@pu.edu.pk

Abstract:

Schizophrenia is a severe, chronic, debilitating mental disorder of central nervous system that affects about 1% of the population. It is associated with a dysregulation of subcortical dopamine system function. Current therapeutics is generally based on dopamine D2 receptor blockade. Continuous drug improvement is in process to improve affectivity and to minimize side effects. Computational (in silico) methods have been developed and widely applied to pharmacology hypothesis development and testing. Dopamine receptor (DRD2) amino acid sequences were downloaded from protein databases and their 3D structures were developed. Reported and potent inhibitors of these receptors were downloaded from standard compound databases. Ligandprotein docking was done by using commonly used docking softwares. A comparison was done between currently available FDA approved drugs for schizophereina and dopamine receptor inhibitors. Inhibitors belonging to family Trifluoperazine (CID_424885), Triflupromazine (CID_25010691), Spiperone (CID_3081175) showed significantly higher (43%, 38% and 36% respectively) docking score then currently available drugs. The drug design in this work may use for effective treatment of this disease with minimum side effects. In-silico pharmacology paradigm is ongoing and presents a rich array of opportunities that will assist in expatiating the discovery of new targets, and ultimately lead to compounds with predicted biological activity for these novel targets.

Keywords: Schizophrenia, Dopamine Receptors, DRD2, Protein-ligand docking, drug designing

BPC-51

Biochemical & Molecular characterization of bioplastic produced by Pseudomonas & Enterobacter using sugarcane bagasse

Muhammad Zaid and Nazia Jamil
Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-e-Azam Campus, Lahore-54590, Pakistan.
Email: jamil_nazi@yahoo.com

Abstract:

Polyhydroxyalkanoates (PHA) are efficient, renewable and environment friendly polyesters. Bagasse is a cheap carbon source which is industrial residue of sugar industries and can be used conviniently as carbon source in microbial media. Two PHA producing bacterial strains, Pseudomonas and Enterobacter wereused. Screening for bioplastic production was done by Sudan black B staining and PHA accmulation was observed as black granules inside the cells. On Nile blue agar plates strains showed flourescence under ultraviolet (U.V) radiations because of PHA prescence. The conditions were optimized for increased yield of PHA on PDA medium containing bagasse as carbon source. On the basis of screening results, optimization and time profiling experiments were performed with both the strains, with two different carbon sources; glucose and bagasse. Pseudomonas showed maximum yield (62% PHA) on PDA medium containing 2% bagasse as compared to Enterobacter (38% PHA). Quantitative analysis of biodegradable plastic was performed by Sodium hypochlorite method and brittle mass of PHA was observed. Extracted PHA was analyzed by gas chromatography coupled with mass spectrometry and presence of methyl ester was confirmed at 37.9th minute. Genomic DNA was isolated and a fragment of phaC gene (phaC) was amplified using 179L R and 179L F primers. The amplified products were purified and submitted for sequencing. The obtained sequences were identified by NCBI-BLAST, and their homolgy with phaC gene of Pseudomonas aeruginosa strains with accession numbers [EU781530.1], [EU781526.1], [JN969045.1], |AY596788.1|, and Bacterium TERI strains with accession numbers 13006|GU196137|, |GU196134.1| was determined. Multiple alignment was done by clustalw, and Phylogenetic tree was constructed using MEGA 5 software. phaC gene was translated to 140 aminoacids (AA) sequence using insilico.ehu. Query sequence of phaC gene was submitted to NCBI GenBank, under the GenBank ID grp3768567.

Keywords: Bioplastics, Polyhydroxyalkanoates, Cheap carbon source

BPC-52

Effect of chromium toxicity on physiological responses of two varieties of *Helianthus annuus* L. (FH-419 and FH-385)

Anis Fatima and Asma Zulfiqar
Department of Botany, University of the Punjab, Quaid-e-Azam Campus 54590, Lahore
Pakistan.

Email: asmazulfiqar08@yahoo.com

Abstract:

In recent years, chromium (Cr) contamination has become a major environmental concern of both soil and water. Efficiency of phytoremediation potential of two varieties (FH-419 and FH-385) of *Helianthus annuus* (sunflower) was investigated in the present study. Both the varieties of sunflower were compared on the basis of physiological attributes (Visual rating, Leaf

Electrolyte leakage (LEL), Chlorophyll content, Relative water content (RWC), Lipid peroxidation and chromium content) to assess their potential for extraction of Cr from soil. Plants were subjected to four different concentrations (50,100,250,500 ppm) of chromium supplied through soil. Relative water content of two varieties (FH-419 and FH-385) increased with gradual increase of chromium concentration in soil. Maximum leaf electrolyte leakage was observed at higher concentration of chromium indicating that toxicity of chromium was intensified at higher concentration in both varieties. Chlorophyll content declined with the increasing dose of chromium in both varieties. Lipid peroxidation increased with high concentration of Cr in both varieties reflecting the phytotoxic effects of Cr. A linear relationship in Cr uptake of shoots and root with increased Cr concentration was observed in both varieties.

Keywords: Chromium toxicity, Phytoremediation, Leaf electrolyte leakage

BPC-53

In-silico screening for disease-modifying agents for Alzheimer's disease: Potent inhibitors for Homo sapiens Acetylcholinesterase (hAChE).

Hafsa Amat-ur-Rasool and Mehboob Ahmed

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam

Campus, Lahore-54590, Pakistan.

Email: mehboob.mmg@pu.edu.pk

Abstract:

Alzheimer's disease (AD) is the most common form of dementia i.e. the severe loss of cognitive functioning in older people (In 2006, 26.6 million sufferers worldwide). It is an irreversible progressive brain disease that slowly destroys memory and even leads to death due to gradual loss of brain cells (neurons) and reduced levels of acetylcholine neurotransmitter. One aspect of its therapy is to inhibit the breakdown of acetylcholine, by blocking the enzyme responsible for its degradation. This can be done by a group of chemicals known as acetylcholinesterase (AChE) inhibitors. Drug discovery today has become immensely dependent on computational methods / approaches using in-silico techniques. In present study Protein-Ligand Docking Method was used to hunt for more efficient hAChE inhibitors. Amino acid sequences of hAChE were obtained from protein database and its 3D structure was generated. Structures in PDB format of 6 FDA approved drugs for treating AD (controls) and their 300 derivatives were obtained from standard online Ligand databases and were docked with the hAChE enzyme by using docking tools. Results for derivatives were compared with that of control on the basis of docking score. Top 10% more efficient inhibitors as compared to controls were subjected to Lipinski Rule of Five to determine their oral bioavailability. 13 inhibitors left after screening included two Donepezil (Aricept®) derivatives, one Tacrine (Cognex®) derivative, one galantamine (Reminyl®) derivative, hybrids such as Tacrine-Hupyridone, Tacrine-4-Aminoquinoline, Tacrine-Melatonin, Tacrine-Donpezil and three Tacrine heterobivalent compounds. Using in-silico drug designing method, bioactive compounds present in online chemical databases can be screened to develop more efficient drugs against cognitive symptoms of Alzheimer's disease.

Keywords: Alzheimer's disease, Acetylcholinesterase, Protein-ligand docking, Drug designing

BPC-54

Effect of surfactants on the dissolution profile of poorly water soluble acidic drugs: Flurbiprofen & Rosuvastatin

Urooj Haroon¹, M. Hashim Zuberi^{1, 2}, Nizamuddin¹ and Abdul Karim¹

Department of Chemistry, Federal Urdu University for Arts, Science and Technology, Karachi.

Department of Environmental Studies, Sindh Madressatul Islam University, Karachi.

Abstract:

We report the combined effect of pH and surfactant on the dissolution profile of two poorly water soluble acidic drugs, flurbiprofen and rosuvastatin, using dissolution bath and UV spectroscopy. Water-insoluble or sparingly water-soluble drug products are most likely to solubilize in the presence of the naturally occurring surfactant and micellar media of the GI tract. Therefore in the present study synthetic surfactants are used as dissolution media because of its mechanistic similarities to in vivo dissolution. Synthetic surfactants investigated were nonionic polysorbate 80 (PS-80), polyoxyethylene (20) cetyl ether and cationic cetyltrimethylammonium bromide (CTAB). Various pharmacopoeias suggest that dissolution characteristics of oral formulations should be evaluated in physiologic pH range of 1.2-7.5 to simulate GI environment, therefore dissolution was performed in three pH mediums; 1, 4 and 7.4.In vitro availability of the selected drugs was calculated alone and in presence of non-ionic and cationic surfactants. Results suggest that in absence of surfactants the availability of both flurbiprofen and rosuvastatin was very low between 10-15% however addition of surfactant brought significant enhancement in the availability of the drugs. The increase was more profound in the presence of cationic surfactant at pH 1 followed by pH 4 and 7.4 owing to electrostatic interaction leading to enhanced drug solubility. Our results show influence of surfactant and pH on the dissolution profile of selected drugs. The cationic surfactant was found to be most efficient in enhancing the dissolution rate of flurbiprofen and rosuvastatin among the surfactants tested. These results would help pharmaceutical researchers involved with the development of new dissolution media in selecting suitable surfactants for water insoluble acidic drugs.

Keywords: Dissolution, UV spectroscopy, Surfactants, Flurbiprofen, Rosuvastatin

BPC-55

Screening of Bacteria from Sindh for Novel Bioactive Compounds useful for Bio-control

Nadia Jamil and Nuzhat Ahmed

Centre for Molecular Genetics, University of Karachi, Karachi-75270.

Abstract:

Pakistan is an agricultural country and agriculture plays an important role in income of large part of the population. The aim of this work was to screen bacteria from Sindh for novel bioactive compounds that can be useful for bio-control. Nine isolates were screened for production of novel bioactive compounds. Three properties were chosen for screening i.e. antibacterial activity, antifungal activity and solubilization of inorganic insoluble phosphate salts. Out of nine isolates two isolates CMGN122 and CMGN370 showed strong positive results. These isolates were studied in detail. CMGN122 was found to produce three bioactive phenazines and CMGN370 was found to produce an isoflavonoid and a novel compound (N-(5-Acetylamino-pentyl)-acetamide).

Keywords: Bioactive compounds, Phenazines, Solubilization

BPC-56

In-Silico designing a fusion protein to combat obesity via inhibition of SOCS3

Komal Yasmin Malik, Mehboob Ahmed and Shahida Hasnain
Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-i-Azam Campus, Lahore-54590, Pakistan.
Email: mehboob.mmg@pu.edu.pk

Abstract:

Obesity has achieved the status of an epidemic and is a major concern, causing vulnerability to disease conditions such as hypertension and diabetes mellitus type II. Obesity can be traced to disturbance in satiety signal which itself is a joint venture of various stomach and neurons derived proteins. Satiety signal of Leptin, an adipocyte hormone, is inhibited by SOCS3 leading to a phenomenon "leptin resistance". SOCS3 is therefore major target for anti-obesity therapy. The objective of this project is to design a drug that blocks JAK2 and gp130 attachment sites on SOCS3, crucial for its inhibitory functions. The drug also constitutes a SOCS box motif which shall compete SOCS3 for elongin BC, required for its stability from degradation. For this purpose all protein motifs resembling human JAK2 and gp130 were searched for in uniprot database (using motif search tool) and docked with SOCS3 using PatchDock. Motifs depicting highest affinity for the respective sites were picked and the amino acids involved in intimate bond were mimicked in sequences coding human JAK2 and gp130. The modifications intended to improve the affinity of native proteins for SOCS3.A fusion protein comprised of these 3 domains ligated together via loops will be the final drug. Previous studies meant to halt SOCS3, employed DNA expression based approaches i.e. methylation and knock out studies. This drug presents one of a kind protein based model, sparing complications. It may prove a prospective drug for diabetes and hepatitis C as SOCS3 upregulation is accused in these cases as well, but of course, only clinical trials shall unravel the feasibility of this idea.

Keywords: Obesity, SOCS3, gp130, Jak2, Leptin, Protein-ligand docking, Drug designing, Fusion protein

BPC-57

Structural, surface and biocompatibility analysis of dental composites

Abdul Samad Khan¹, MariaTahir Azam¹, Saadat Anwar Siddiqi¹ and Ihtesham Ur Rehman²

¹Interdisciplinary Research Centre in Biomedical Materials, COMSATS Institute of Information Technology, Lahore, Pakistan.

²Department of Materials Science and Engineering, The Kroto Research Institute, The University of Sheffield, Sheffield, United Kingdom.

Abstract:

Bacterial adhesion on the surface of dental restorative composites is an important parameter in the etiology of secondary caries formation. Newly developed dental materials have to be tested for their susceptibility to adhere bacteria. The objective was to establish an *in-vitro* evaluation of adhesion of oral bacteria on the grounds of their chemical composition and surface characteristics of dental composites. Three newly developed commercial dental composites [FiltekTM Z350 XT, FiltekTM P90 (3M ESPE, Germany) and SpectrumTM TPH[®] (Dentsply, Germany)] were structurally (Fourier Transform Infrared Spectroscopy) analyzed with periodic time internals and the surface roughness and wettablility behavior was evaluated with Atomic Force Microscopy and Contact Angle Measurement respectively. The experimental materials and

control group (tissue-culture plate) were tested with three bacterial strains [Staphylococcus aureus (ATCC 25923), Pseudomonas aeruginosa (ATCC 27853) and Escherichia coli (ATCC 25922)] for microbial attachment at 0, 8, 24, 48 and 72 hours. Forty five sample discs of size 8x2mm were prepared (n=15 each) and evaluated with Scanning Electron Microscopy along with Colony Forming Unit Count and Optical Density measurement of the media. The data were statistically analyzed by using ANOVA. It was found from spectroscopic analysis that all composite materials showed degree of conversion with time. Median roughness values of these experimental composites were ranged between 20-40nm. The median contact angle was found between 65-92°. It was observed that among composites FiltekTM Z350 XT showed least adhesion with all strains and it is suggested that Z350 has a better chance of evasion of biofilm formation as compared to P90 and TPH. The trend in difference in number of bacterial attachment can be related to particle size, surface morphology, chemical composition and surface free energy of the dental composites.

Keywords: Dental composites, Secondary caries, Biofilm

BPC-58

Antimycobacterial compounds from indigenous Streptomycetes

Muhammad Abbas¹, Atiqa Ambreen² and Imran Sajid¹
Department of Microbiology and Molecular Genetics, Quid-i-Azam campus, university of the Punjab, Lahore-54590, Pakistan

²Gulab Devi Chest Hospital, Ferozpur road, Lahore, Pakistan

Abstract:

Tuberculosis is a problem of global significance, estimated to cause about 8 million new cases of disease and about 3 million deaths each year, more than half of which are in Asia. The emergence of multi-drug resistant tuberculosis (MDR-TB) and extensively drug resistant tuberculosis (XDR-TB) are of public health concern. Actinomycetes are best known for their ability to produce antibiotics and are gram positive bacteria. Among actinomycetes, the streptomycetes are the main source of clinically important antibiotics. Most of commercial anti TB drugs and other antibiotics, including streptomycin, erythromycin derivatives, macroloides, vancomycin and teicoplanin, tetracyclines, and daptomycin, have been isolated from various members of the genus Streptomyces. A collection of 44 indigenous streptomycetes strains originally isolated from saline soil were screened biologically and chemically to investigate their potential for the production of antimycobacterial compounds. In biological screening the crude extracts obtained from the culture broth of selected strains were analyzed for their activity against MDR and XDR Mycobaterium tuberculosis strains by proportional method. In chemical screening each of the crude extracts was analyzed by thin layer chromatography (TLC) using various staining reagents and by HPLC-RI. The results depicted an impressive antimycobaterial activity and chemical diversity of crude extracts produced by these indigenous streptomycetes strains.

Keywords: Indigenous streptomycetes, MDR-TB, XDR-TB, Thin layer chromatography, HPLC- RI

BPC-59

Effect of biofilm formation of dental plaque isolates on the surface of artificial teeth

Hafiz, Ghulam Murtaza Saleem and Anjum Nasim Sabri

Department of Microbiology and Molcular Genetics University of the Punjab, Quaid-e-azam Campus, Lahore-54590 Pakistan.

Email: g_murtazasa@yahoo.com

Abstract:

Four biocides (Benzidamine hydrochloride 0.15%, Chlorohexidine 0.2%, Sodium floride 0.05% and potassium chloride 0.05%, and Benzidamine hydrochloride 0.15% and Chlorohexidine 0.2%) were used by culture dependant method to monitor the efficacy of bioocides and resistance pattern of dental plaque isolates. Resistant profile from three dental plaque isolates e.g. Acenitobacter sp. (accession no. JF837190), Moraxella sp.(accession no. JF837191) and Bacillus sp. (accession no. JF837192) showed maximum resistance against all the biocides up to the concentration of 1000 µgml⁻¹. Artificial teeth of acrylic resin were constructed and used for bacterial inoculation and biofilm formation. Acrylic resin teeth are naturally more compatible with the denture base than porcelain teeth. After thirty days artificial teeth were drawn aseptically from 12 test (25ml media+ artificial tooth+ 25ml biocide) and 2 control (50ml media+ artificial tooth) flasks and subjected to differentiately procedures of surface microscopy, tooth hardness, and color parameters to find out the effect of bacterial biofilm. Artificial teeth were subjected for the microscopic (5×10 magnification)examination that reveals significant surface deterioration of artificial teeth from test flask compared with the surface of artificial teeth from control flask. Microscopic view depicted clear difference on the surface (before and after bacterial inoculation) of artificial teeth. Hardness of artificial teeth was measured (before and after bacterial inoculation)that result in the hardness reduction. Significant changes were observed in the color, brightness, and gloss of artificial teeth due to the impact of bacterial biofilm.

Keywords: Biocides, Artificial teeth, Biofilm

BPC-60

Effect of Nickel on adherence, hydrophobicity, aggregation and biofilm formation of salt tolerant bacterial strains

Maryum Fakhar and Anjum Nasim Sabri Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-i-Azam Campus, Lahore, Pakistan. Email: anjumsabre1@yahoo.com

Abstract:

The survival of bacteria exposed to toxic heavy metals is a multifactorial phenomenon, which involves various mechanisms of resistance and tolerance. Biofilm formation is one of the resistance mechanisms of bacteria for their survival. The main objective of this study was to assess the motility, hydrophobicity, aggregation and biofilm formation of two salt tolerant strains (Bacillus subtilis and Halomonas sp.) in the presence of toxic metals such as nickel. Two concentrations of nickel were used i.e. 500ug/ml and 1000ug/ml for subsequent tests. The results revealed that motility (swimming, swarming and twiching) was greatly reduced in the presence of nickel whereas hydrophobic nature was not significantly affected by the nickel stress as investigated by BATH (Bacterial Adherence to Hydrocarbons) and SAT (Salt Aggregation test).

In case of auto aggregation and co- aggregation, varying response was observed in the presence of nickel. Furthermore, the effect of bacterial strains on the soil aggregates formation in the presence and absence of nickel stress was also analyzed, which showed that soil aggregation was enhanced at the low nickel concentration (i.e.500ug/ml). In most of the cases, biofilm formation was markedly reduced in the presence of nickel stress on plastic surface (microtiter plate assay) and on glass surface as indicated by fluorescent microscopy after staining with acridine orange. In short, the toxic effect of nickel was observed for the salt tolerant isolates with few exceptions. **Keywords:** Nickel, Biofilm, Aggregation, BATH, SAT, Soil aggregates

BPC-61

DNA interaction studies of 3-Formyl chromone derived sulfonamide enamines and their transition metal complexes using gel electrophoresis

Mehwish Iftikhar¹, Faiza Zarif¹, Salma Mukhtar² and Mariya-al-Rashida¹
Department of Chemistry, Forman Christian College (A Chartered University), Ferozepur Road-54600, Lahore.

²Department of Biological Sciences, Forman Christian College (A Chartered University), Ferozepur Road-54600, Lahore.

Abstract:

3-formyl chromone derived sulfonamide enamines (L1-L6) were synthesized. Compounds L1-L3 were synthesized by reacting 3-formylchromone, 6-ethyl-3-formylchromone and 6,8-dibromo-3-formylchromone with 4-aminobenzenesulfonamide respectively. Similarly compounds L4-L6 were prepared by reacting 6-ethyl-3-formylchromone, 6, 8-dibromo-3-formylchromone and 6-fluoro-3-formylchromone with 3-aminobenzenesulfonamide respectively. All reactions were carried out in ethanol which is sufficiently nucleophilic to add into C2-C3 double bond of chromone ring to form enamines. Transition metal complexes of L1-L6 were prepared by refluxing equimolar amounts of ligands with Cr (III), Co (II), Ni (II), Cu (II) and Zn (II) metal ions. Compounds were characterized using standard analytical techniques such as UV, FTIR, ¹H-NMR, ¹³C-NMR, and MS, TGA, and Conductance studies. Single crystal X-ray diffraction study for L1 has already been reported by us. DNA interaction studies of L1-L6 and their metal complexes (at 100, 50 and 10 μM conc.) were studied using gel electrophoresis against prokaryotic DNA isolated from *Micrococcus luteus* (a gram positive bacterium). Compounds 24 (L4), 27 (L5) and 28 (Cu[L5]₂) exhibited relatively least binding with prokaryotic DNA at 10μM conc., closely followed by compounds 29-32, which showed similar binding patterns.

$$R^{1}$$
 R^{2}
 R^{1}
 R^{2}
 R^{2

Where, L1;
$$R^1 = H$$
, $R^2 = H$; $p\text{-SO}_2NH_2$
L2; $R^1 = Et$, $R^2 = H$; $p\text{-SO}_2NH_2$
L3; $R^1 = Br$, $R^2 = Br$; $p\text{-SO}_2NH_2$

L4;
$$R^1 = \text{Et}$$
, $R^2 = \text{H}$; m-SO₂NH₂
L5; $R^1 = \text{Br}$, $R^2 = \text{Br}$; m-SO₂NH₂
L6; $R^1 = \text{F}$, $R^2 = \text{H}$; m-SO₂NH₂

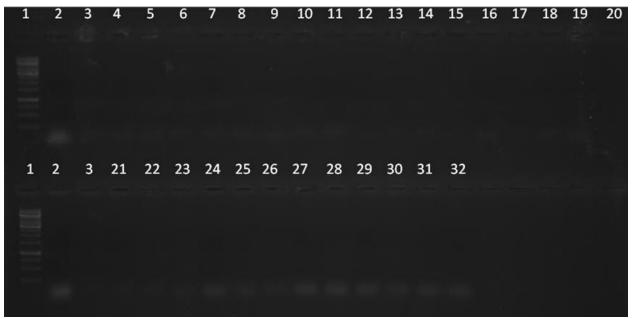


Fig 1. DNA interaction studies using gel electrophoresis of ligands and their metal complexes (at 10 μ M conc.); 1 = ladder, 2 = DNA (*Micrococcusluteus*), 3 = blank, 4 = CrCl₃, 5 = Cr(OAc)₃, 6 = Co(OAc)₂, 7 = Ni(OAc)₂, 8 = CuCl₂, 9 = Cu(OAc)₂, 10 = ZnCl₂, 11 = L1, 12 = Cr[L1]₂, 13 = Cr[L1]₂.2H₂O, 14 = Co[L1]₂, 15 = Ni[L1]₂, 16 = Cu[L1]₂, 17 = Zn[L1]₂, 18 = L2, 19 = Cu[L2]₂, 20 = Zn[L2]₂, 21 = L3, 22 = Co[L3]₂, 23 = Zn[L3]₂, 24 = L4, 25 = Cu[L4]₂, 26 = Zn[L4]₂, 27 = L5, 28 = Cu[L5]₂, 29 = Zn[L5]₂, 30 = L6, 31 = Cu[L6]₂, 32 = Zn[L6]₂.

Keywords: Transition metal, DNA, Ligands

BPC-62

DNA interaction studies of sulfonamide transition metal complexes using gel electrophoresis

Rashid Masih¹, Umer Khan¹, Mehwish Iftikhar¹, Faiza Zarif¹, Salma Mukhtar² and Mariya-al-Rashida¹

¹Department of Chemistry, Forman Christian College (A Chartered University), Ferozepur Road-54600, Lahore

²Department of Biological Sciences, Forman Christian College (A Chartered University), Ferozepur Road-54600, Lahore

Abstract:

Sulfonamide Schiff bases 4-[(E)-(2-hydroxybenzylidene) amino]benzenesulfonamide (L1) and 4-{2-[(Z)-(2-hydroxybenzylidene)amino]ethyl}benzene sulfonamide (L2) were synthesized by reacting 2-hydroxybenzaldehyde with 4-aminobenzenesulfonamide and 4-(2-aminoethyl) benzene sulfonamide respectively. Transition metal complexes of L1 and L2 were prepared by refluxing equimolar amounts of ligands with Cr (III), Co (II), Ni (II), Cu (II) and Zn (II) metal ions. Compounds were characterized using standard analytical techniques such as UV, FTIR, TGA, AAS and Conductance studies. Single crystal X-ray diffraction studies were carried out for L2. DNA interaction studies of L2 and its metal complexes (at 100 µM conc.) were studied using gel electrophoresis against prokaryotic DNA isolated from *Pseudomon asputida* (a gram

negative bacterium). Ni (II), Cr (II) and Cu (II) complex exhibited relatively least binding with prokaryotic DNA.

Fig 1. DNA interaction studies using gel electrophoresis of ligands and their metal complexes (at 100 μ M conc.); a = Ladder, b = prokaryotic DNA, c = Cr(OAc)₃, d = ZnCl₂, e = Ni(OAc)₂, f = CoCl₂, g = CrL₂, h = CuL₂, i = ZnL₂, j = NiL₂, k = L1, l = L2

Keywords: Transition metal, Sulfonamide, Prokaryotic DNA

BPC-63

Appraisal of volatile constituents; Antioxidant and antimicrobial Activity of *Ocimum basilicum* essential oil

Muafia Shafique, Shaista Jabeen Khan and Nuzhat Habib Khan
Biotechnology and Food Research Centre, PCSIR laboratories Complex
Lahore-54600, Pakistan.
Email: aine4me@yahoo.com

Abstract:

The aim of present study was to characterize the physical and chemical properties as well as to explore the antioxidant and antimicrobial scope of *Ocimum basilicum* essential oil. The essential oil was obtained from aerial parts of the plant, extracted through hyrodistillation. The % yield of *O. basilicum* oil was found 0.74 %. Physical properties i.e, odor, color, specific gravity, solubility, refractive index and acid value were recorded according to the standard procedures. Chemical composition of *Ocimum basilicum* essential oil was analyzed by Gas Chromatography-

Mass Spectrometry (GC-MS). It was found that the essential oil contained both terpenoids and phenylpropanoids among which 1, 6-octadiene-3-ol, 3, 7-dimethyl (24.43%) was most abundant component. Present investigation depicted that *O. basilicum* essential oil has significantly high antioxidant activity at all concentrations studied i.e. 20%-100% showing percent inhibition of DPPH ranging from 90.04% to 96.16%. Butylated hydroxytoluene (BHT) was used as positive control. The antioxidant activity of sweet basil essential oil at 100% concentration was recorded to be 12.33 % higher than the corresponding level of BHT. The results of antimicrobial assay showed that *O.basilicum* essential oil was active against all Gram positive and Gram negative microbial strains tested. It is evident from this study that *Ocimum basilicum* essential oil is more potent against tested organisms as compared to the standard antibiotics used as positive control. **Keywords:** Volatile constituents, Antioxidant activity, DPPH assay, *Ocimum basilicum*, Essential oil, Antimicrobial activity.

BPC-64

Determination of shear bond strength of nanocomposite to porcelain and metal alloy Zenab Sarfraz Sheikh

Interdisciplinary Research Centre in Biomedical Materials, COMSATS Institute of Information Technology Lahore, Pakistan.

Abstract:

Fracture of porcelain and metal alloy crown and bridges is a common ordeal all dentists go through. Although fracture of prosthesis does not necessarily mean failure, the replacement process is expensive, prolonged and may lead to weakening, pulpal damage and tooth fracture. This study was done to evaluate the shear bond strength of porcelain, porcelain fused to metal and metal alloy to Nano composite and the currently used conventional composite at compositeporcelain/metal interface and results compared. A total of 120 cylindrical discs of 10mm diameter and 4 mm thickness would be prepared from Porcelain, Porcelain fused to metal and metal substrate and divided accordingly into 1) Control group comprising of 60 cylinders and 2) Experimental group comprising of 60 cylinders. These cylinders would be further divided into 3 subgroups A) Porcelain B) Porcelain Fused to metal C) Metal. Control group(A)cylinders would be bonded with Conventional dental composite whereas Experimental group (B) with Nano composite, both in 3.5mm diameter and 2mm thickness and polymerized according to manufacturer's instructions. All specimens would be thermo cycled for a dwell time of 30sec between 5C to 50C for 200cycles. Before testing they would be stored in Distilled water at 37C for 7 days. To test Shear bond strength, Universal Testing machine with 0.5cm/min crosshead speed would be used. The quantitative data would be analyzed by two independent t-test. This study would pave way for the usage of Nano composite as Porcelain Repair Material due to superior mechanical as well as aesthetic properties with improved durability and longevity of fractured/ damaged porcelain and metallic dental prosthesis and hence serve as a cost effective, compliant and time saving procedure.

Keyword: Porcelain, Metal, Nano composite, Shear bond strength.

BPC-65

Targeting the mevalonate pathway for cancer therapy

Hafiz Muhammad Jafar Hussain, RimshaMunir, and Nousheen Zehra Zaidi Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-i-Azam Campus, Lahore 54590, Pakistan.

Abstract:

The mevalonate pathway is a complex biochemical pathway required for the generation of several key metabolic end-products, including cholesterol, isoprenoids, dolichol, ubiquinone, and isopentenyladenine. The significance of altered metabolism in the context of tumor development and progression has received renewed attention in the last decade. Though, several metabolic pathways have been linked to cancer progression and etiology, it was only recently that the mevalonate pathway was shown to have a distinctive role in cellular transformation. Hence, the specific hypothesis deriving the proposed research project is that targeting different enzymes involved in the mevalonate pathway may offer therapeutic opportunities for metabolically treating and preventing cancers. By examining the effect of silencing different genes involved in mevalonate synthesis in cancer cells we aim to elucidate the importance of these genes in tumor cell transformation, growth and survival. Our long term goal is to evaluate the therapeutic potential of tumor-associated metabolic markers and to apply this knowledge in developing remedial strategies for treatment of cancer.

Keywords: Mevaolnate Pathway, Tumor Metabolism, Cancer

BPC-66

Diagnostic and prognostic relevance of blood lipid profiles with cancer: An issue of ambiguity

Rimsha Munir, Hafiz Muhammad Jafar Hussain and NousheenZehra Zaidi Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus, Lahore 54590, Pakistan.

Email: rimsha.munir08@gmail.com

Abstract:

Cancer cells are categorized by high proliferation rates and exhibit increased demands for energy and macromolecules. Lipids constituteone of the major groups of macromolecules that are abundantlyrequired for membrane biogenesis and protein modifications in rapidly dividing cancer cells. Also, the cancer cells with lower proliferation rates require increased amounts of lipids for enhanced signaling and resistance against apoptosis. Lipids including cholesterol, phospholipids and sphingolipids are essential components of cell cycle and they also act as signaling messengers between cells. On the other hand triacylglycerols are the major reservoir of body fats. Numbers of epidemiological studies have indicated an inverse relationship between total serum cholesterol level and incidents of cancer. Plasma lipid profiles also haveassociationwith cancer progression. Steroid hormone depending cancers are more likely to be co-related with level of blood lipoproteins. Atypical blood lipid profiles are also associated with advanced stage of oral, colorectal, endometrial, gall bladder; breast and prostate cancer. Lowest total cholesterol and LDL-C levels have been reported in the case of hematopoietic malignancies. However, it is still obscure whether this aberrant blood lipid profile supports cancer progression or alternatively, the aggressive stage of cancer results in altered

blood lipid levels. In the present study we are investigating the relationships between serum lipid profile with cancer type, stage and grade.

Keywords: Blood lipid profiles, Cholesterol, Cancer

BPC-67

Study of serum lipid profile in patients with cardiovascular diseases in Lahore

Saleem Ullah Shahid and Abdul Rehman

Microbiology and molecular Genetics, Quaid-e-Azam campus, University of the Punjab, Lahore-54590 Pakistan.

Email: saleem_shhd@yahoo.com

Abstract:

Current study is conducted to find out possible role of serum lipid profile parameters in development and pathogenesis of cardiovascular diseases. For this purpose, 111 male and 81 female patients suffering from various cardiovascular diseases, diagnosed clinically and on the basis of electrocardiogram, echocardiography and angiography were taken from regional hospitals of Lahore. Hypertension and diabetes mellitus were present in 44.94% and 66.02% cases respectively. Ischemic cardiomyopathies were 47.6% where as others were 52.4% in selected population. Plasma lipids including total cholesterol, HDL Cholesterol, LDL Cholesterol, triacylglycerols and total lipids were measured by spectrophotometeric kits. A significant decrease in HDL (p=<.05) and increase in LDL (p<.05) was observed. There was a negative Pearsons correlation of -.123 between HDL and LDL. No significant difference was present in total cholesterol, triacyglcerols and total lipids concentration. Serum electrolytes (Na⁺, K⁺) were also unchanged. In conclusion, low HDL and high LDL cholesterols have major contribution in cardiovascular anomalies in the population under study.

Keywords: Lipid profile, Hypertension, Cardiomyopathies

BPC-68

Optical properties of Electrodeposited alumina thin films by using Spectroscopic Ellipsometer

Farzana Majid, Saira Riaz and Shahzad Naseem
Centre of Excellence in Solid State Physics, University of the Punjab, Lahore, Pakistan.
Email: farzanamajid@yahoo.com

Abstract:

Aluminum oxide thin films are prepared for their potential application as barrier coatings. Electrodeposition method is used for depositing alumina films. All the depositions are carried out at room temperature in the voltage range of 1-5 volts. Reaction time is also varied to observe the changes in the structure and morphology of thin films. Dense films with spherical nanoparticles ~ 200 nm are observed at 3 volts. JA Wollam spectroscopic ellipsometer is used to study the optical properties of thin films. Thickness of around 1 micrometer is observed for all the samples prepared at 3 volts. However, surface roughness was increased from 5 -12 nm by increasing the reaction time from 15 to 60 minutes. Energy band gap of ~ 3.9 eV is observed which is consistent with our previously reported alumina thin films by using sol- gel method. Refractive index of ~ 1.77 is observed at 300 nm. Low Porosity ~ 2.5% was observed for the films prepared at 3 volts for a reaction time of 60 minutes.

Keywords: Electrodeposition, Spectroscopic ellipsometer, Refractive index

BPC-69

Effects of microbiologically influenced corrosion on the mechanical properties of Al-Cu alloy

Muhammad Yousaf¹, Hafiz Zeshan Wadood², Ijaz Mujtaba Ghauri¹, Anjum Nasim Sabri², Saira Riaz¹ and Shahzad Naseem¹

¹Center of Excellence in Solid State Physics, University of the Punjab 54590, Lahore, Pakistan.

Abstract:

The present research work is aimed at the study of the effects of Microbiologically Influenced Corrosion on the mechanical properties of Al-Cu alloy in the presence of *Bacillus megaterium* isolated from industrial effluent. Bicorrosion behaviour and mechanical properties of Al-Cu alloy were evaluated using electrochemical impedance spectroscopy (EIS), scanning electron microscopy (SEM), Microstructure analysis, tensile testing and hardness measurements respectively. Electrochemical and SEM data showed more corrosion of Al-Cu alloy in bacterial inoculated medium compared to control medium. Mechanical properties such as yield stress (YS), ultimate tensile stress (UTS), percentage elongation and hardness of Al-Cu alloy were found to be decreasing in the presence of bacterial inoculated medium compared to control medium.

Keywords: Microbiologically influenced corrosion (MIC), Yield stress (YS), Ultimate tensile stress (UTS), Electrochemical impedance spectroscopy (EIS)

BPC-70

Phytostimulatory impact of *Kushneria* sp., *Arthrobacter* sp.and *Halomonas* sp. on the growth of *Triticumaestivum*Sadia Naseem¹, Ambreen Ahmed¹ and Shahida Hasnain²

Sadia Naseem¹, Ambreen Ahmed¹ and Shahida Hasnain²

¹Department of Botany, University of the Punjab, Lahore 54590, Pakistan.

²Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore 54590, Pakistan.

Abstract:

Plant growth promoting rhizobacteria (PGPR) have the growth promoting ability which can be utilized for plant growth enhancement in a cost effective manner. Healthy seeds of *Triticum aestivum* were inoculated with *Kushneria* sp., *Arthrobacter* sp. and *Halomonas* sp. isolated from the indigenous environment and after germination, the seeds were grown under various stresses of chromium salts (CrCl₃, K₂CrO₄ and K₂Cr₂O₇). Various growth and biochemical parameters of inoculated and non-inoculated seedlings were recorded. Results of these experiments revealed that bacterial inoculation has a stimulatory impact on germination, growth parameters and enzyme activity but caused a decrease in uptake of Chromium by seedlings thereby reducing the adverse effects of chromium accumulation in plants on their growth. Thus these strains can be utilized as biofertilizers for better growth and development of plants under chromium stress.

Keywords: Rhizobacteria, Chromium salts, Biochemical parameters

² Department of Microbiology and Molecular Genetics, University of the Punjab 54590, Lahore, Pakistan.

BPC-71

Variation of phyllospheric bacteria at different developmental stages of *Helianthus annuus* and role of metabolites in microbial selection

Saher Mahmood and Shahida Hasnain
Microbiology and Molecular Genetics, University of the Punjab, Quiad-E-Azam Campus,
Lahore-54590

Email: genetic@brain.net.pk

Abstract:

Phyllosphere is an unreceptive habitat for microbes due to swift and recurring alterations in surrounding environment. However, plants passively arrange metabolites to support the growth of microbial flora on leaves. The nature of the metabolites released depends on plant species, its developmental stage and abiotic factors. Phyllospheric bacteria participate actively in nutrient cycling especially by nitrogen fixation. Less is known about the mechanisms that operate between plant's excreted metabolites and phyllospheric commensal bacteria. To exploit the potential of these bacteria a better understanding of plant-bacteria association and bacterial role in plant growth and development is necessary. We are working on isolation and characterization of bacteria from phyllosphere of *Helianthus annuus* at different life spans. Both surface and endophytic bacteria have been isolated using three different types of media as well as media supplemented with leaf extracts. This could provide insights of physiological and metabolic mechanisms that operate on the leaves of the plants and will help to understand the roles of these bacteria play in plant growth promotion.

Keywords: Epiphytes, Endogenous bacteria, Plant growth promotion, Culture-dependent Method, Diversity of bacteria

BPC-72

Biodegradation of Chlorpyrifos by bacterial consortium enriched from agriculture soil

Shamsa Akbar¹, Sikander Sultan¹ and Michael Kertesz²

¹Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore, Pakistan.

²Faculty of Agriculture and Environment, University of Sydney, Sydney, Australia.

Abstract:

Chlorpyrifos is chloride radical containing organophosphate, widely used for protection of cotton, paddy fields and vegetable crops. The widespread use of chlorpyrifos is hazardous to the environment and also toxic to mammals therefore exploration of chlorpyrifos degrading microbes to clean-up the pollutant is of immense importance. This study aims at enrichment, isolation and screening of bacteria capable of metabolizing chlorpyrifos and 16s rRNA molecular fingerprinting of soils. From chlorpyrifos contaminated soil morphologically different strains were selected and potential for chlorpyrifos utilization was estimated. Robust strains were characterized and identified as *Acinetobacter calcoaceticus (MCp-231)*, *Stenotrophomonas maltophilia (MCp-232)*, *Pseudomonas aeruginosa (YCp-233)* and *Pseudomonas mendocina (MCp-234)*. Chlorpyrifos degradation efficiency of separate strains and as a consortium was estimated in broth and in soil, HPLC analysis showed 50-70% chlorpyrifos degradation at 200mgL⁻¹ concentration with separate strains in 7 days while consortium was able to degrade it completely. Survival and proliferation of chlorpyrifos degrading bacteria in soil incubated with known concentration of chlorpyrifos for several weeks and in enrichment cultures was monitored

by molecular fingerprinting of bacterial 16S rRNA genes by PCR-Denaturing gradient gel electrophoresis (DGGE). 16S DNA sequence analysis of continuously appearing bands showed homology to group Rhizobiales (subgroup Rhizobium, Xanthobacter, Aminobacter and Ochrobactrum), Pseudomonas, Pseudoxanthomonas and Stenotrophomonas. The results of 16s rRNA analysis of DGGE bands matched with gene analysis of isolates indicating that these organisms are involved in the degradation of chlorpyrifos in soil therefore these organisms could prove valuable for in-situ bioremediation of contaminated soils and waters.

Keywords: Chlorpyrifos, HPLC, Molecular fingerprinting

BPC-73

Molecular basis of antifungal resistance in tomato varieties

Shazia Shafique, Aqeel Ahmad and Sobiya Shafique Institute of Agricultural Sciences, University of the Punjab, Quaid-e-Azam Campus, Lahore, Pakistan.

Email: shazia.iags@pu.edu.pk

Abstract:

Tomato has a significant share in daily human food consumption and its yield should necessarily be enhanced against a number of its fungal pathogens. Use of plant innate resistance against pathogens is an effective and cheapest tool of crop protection and analysis of proteomics and transcriptome of plant explore the qualitative and quantitative basis of this resistance which is helpful to design future agriculture plans. So, constitutive antifungal resistance of tomato varieties commonly cultivated in Pakistan has been evaluated on the basis of their transcriptome and protein profile analysis. Study disclosed the fact that six pathogenesis related genes belonging to families PR1, PR2, PR3, PR7 and MT2bL exhibited higher transcriptional rate in comparison with the Chitinase 3 acidic, which showed lower expression in resistant tomato variety. According to the semi quantitative RT-PRCR results Osmotin-like PR5 didn't regulate constitutive antifungal resistance and showed equal expression with the varying resistance of plants. But, resistant tomato plants had four additional protein species ranging in size from 40-52 kDa, in their cellular contents and those proteins might be resistance ensuring factors of tomato as they were absent in susceptible plant protein profile. This study demonstrates the molecular basis of tomato resistance against fungal pathogens and will be helpful for researchers to improve such resistance in tomato varieties under development.

Keywords: Antifungal resistance, Pathogenesis related genes, Gene expression profile, Proteomics studies.

BPC-74

Eco friendly response of alternaria tomato leaf spot by citrus peel

Sobiya Shafique, Shazia Shafique and Ageel Ahmad Institute of Agricultural Sciences, University of the Punjab, Quaid-e-Azam Campus, Lahore, Pakistan.

Email: sobiya.iags@pu.edu.pk

Abstract:

Citrus is an abundant agricultural produce of Pakistan. All of its uses including food and beverage industries result into the waste of its peel which may be a remarkable reservoir of plant defensive biochemicals including phenolics; and can be used to control various plant pathogens

e.g. *Alternaria alternata* causing leaf spots of tomato. Present study concentrates upon percentage phenolics recovery and evaluation of antifungal potential of phenolics isolated from peels of various citrus types i.e. Lemon, Kinno, Malta, Musammi and Fruiter. Maximum phenolics recovery was carried out from Musammi (0.21%) with the decreasing trend towards Malta (0.19%) > fruiter (0.14%) > Lemon (0.13%) > kinno (0.06%). Moreover, Musammi contributed non-significantly better control of *A. alternata* in comparison with the Malta. But growth inhibition was significantly less in case of Lemon, Kinno and Fruiter. A spray of phenolics prior to infection provided protection from fungal pathogen with the maximum disease control in case of 1% Musammi. While a decreased curative potential of phenolic acids, was prevailed in order with Musammi > Malta > Fruite r > Kinno > and Lemon with the maximum individual percentage control of 73.4 > 59.7 > 54.7 > 45.5 > and15.3%; respectively. Such high antifungal potential of phenolics makes citrus peel an important source of pesticide.

Keywords: Citrus peel, Leaf spots, Curative potential

BPC-75

Prevalence of enteric bacteria in fresh vegetable salads and occurrence of integrons among drug resistant isolates

Atique ur Rehman and Sikander Sultan

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam

Campus, Lahore-54590

Abstract:

The objective of this study was to examine the bacterial load and the occurrence of some disease causing enteric bacteria in fresh vegetable salads to evaluate whether fresh vegetable salads contain a possible reservoir of antibiotic resistance. Total 46 samples of fresh vegetable salads were collected from hotels and street points of Lahore. Total viable count of these samples ranged from 10 x 10⁴ to 1000 x 10⁶ and coliform count from 8 x 10⁴ to 300 x 10⁴. From 46 samples, 76 isolates were isolated including 16 *Salmonella*, 11 *Proteus*, 9 *Shigella*, 12 *E.coli*, and 28 *Kalebsiella* strains. These isolates were examined for their susceptibility to 9 different antibiotics like amoxicillin, ciprofloxacin, amikacin, tetracycline, cefuroxine, nalidixic acid. The carriage of integrons in the isolates resistant to two or more antibiotics was determined. Genomic DNA was isolated from antibiotic resistant strains and PCR for integron DNA amplification was also conducted.

Keywords: Enteric bacteria, Antibiotics, Integron DNA

BPC-76

Comparative assessment of heavy metal biosorption by wood rotting fungi

Amna Shoaib

Institute of Agricultural Sciences, University of the Punjab, Quaid-e-Azam Campus 54590-Lahore, Pakistan.

Email: aamnaa29@yahoo.mail.com

Abstract:

Biosorption by macromycetes (fungi) has recently been projected for the remediation of wastewater containing toxic heavy metal ions as an alternative to expensive and cautionary chemical based procedures opted as purification treatment for industrial effluents. In the present study the biosorption potential of three species of Basidiomycetous fungi namely *Schizophyllum*

commune Fries, Ganoderma lucidum (Curt. Fr.) P. Karst and Pleurotus ostreatus (Jacq.) Quéletwas evaluated for removing Cr(III) ions from wastewater of tannery treatment plant (TTP) To predict the biosorption performance of the test fungi with actual water of TTP preliminary laboratory assays were conducted with synthetic pure metal-bearing solutions in a batch fashion. Experiments were performed as a function of pH, biomass dose, equilibrium time and initial metal ion concentrations. Results showed, S. commune exhibited the highest removal of Cr(III) followed by P. ostreatus and G. lucidum. Solution pH evaluated between a range of 2.0-6.0, seems to be an important parameter affecting biosorption performance of the test fungi, being negligible uptake of metal at pH < 3.5 and adsorption efficiency reached plateau at around pH 4.5-5.0. Varying amounts of adsorbent doses i.e. 0.03-0.8 g, revealed removal capacity declined while efficiency was substantially improved with increase in biosorbent dose. Equilibrium was obtained in contact time of 1 hour for both S. commune and P. ostreatus and in 3 hours by G. lucidum. The regenerated fungal biomass, reemployed in at least four cycles of biosorption, showed no significant loss in heavy metal removing capacity with high desorption capacity of P. ostreatus (> 89%) in comparison to rest of two fungi (> 87%). The distribution of metallic ions between liquid and solid phase was adequately described by Langmuir and Freundlich isotherms model. With actual water of tannery treatment plant, P. ostreatus exhibited 55% sorption efficiency followed by 43% efficiency by both S. commune and G. lucidum for Cr(III) ions. To make this technique economically feasible, the study was further extended by mass cultivated this fungus on wheat straw and assessment of biosorption potency of colonized wheat straw for Cr(III) ions. Wheat straw colonized with either of three fungi mycelia possessed tremendous ability to remove Cr(III) (removal efficiency: 70-90%) from aqueous solution thus could be utilized as an excellent biosorbent for Cr(III) adsorption at low concentration of the metal (4-20 $mg L^{-1}$).

Keywords: Biosorbent, Tannery treatment plant, Macromycetes

BPC-77

Cr(VI) removal by indigenous bacteria in combination with *Chlorella*

Saima Akram and Rida Batool

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus Lahore-54590, Pakistan.

Email: ridazaidi_1@yahoo.com

Abstract:

Chromium (Cr) is the 17th most abundant toxic element in the Earth's mantle that is widely used in many industrial practices. Due to the wide anthropogenic use of Cr, the subsequent environmental contamination increased and has become an increasing concern in the last years. The hexavalent form of the metal; Cr(VI) is more toxic species than the relatively less mobile Cr(III) form. The 7 bacterial strains were isolated from soil contaminated with high stress of oil, 3 of them named as S1, S6 and S7 exhibited significant resistance against Cr(VI) these were characterized as *pseudomonas* sp. biochemically, morphologically and physiologically. The reduction potential of Cr(VI) of S1, S6 and S7 was observed 95%, 96% and 98% respectively by diphenylcarbazide method, after the 24 hours incubation with 500μg/ml concentration of K₂CrO₄. Cr(VI) removal capacity of hydrophyte *Chlorella* is evaluated with nutritional conditions by incubating it with the three different concentrations (0, 350, 500 μg ml⁻¹) of Cr(VI) in natural environment for two weeks; considerable uptake of Cr(VI) was examined

which showed capability of *Chlorella* to remediate metal ions through phytoremediation. Later on experiment was done by using the *Chlorella* in the combination of isolated bacterial strains S1, S6 and S7. The uptake of Cr(VI) for *Chlorella* and *Chlorella* in combination with bacterial strains was checked by wet digestion method.

Keywords: Cr(VI) removal, Phytoremediation, *Chlorella*,

BPC-78

Effect of chromium-resistant bacteria on the growth of wheat

Rabia Faryad Khan and Rida Batool

Department of Microbiology and Molecular Genetics, University of the Punjab,
Quaid-e-Azam campus 54590, Lahore, Pakistan.

Email: ridazaidi 1@yahoo.com

Abstract:

Cr(VI) is a highly toxic metal for all the living organisms. Due to its extensive industrial use, it has become a serious pollutant in diverse environmental settings. The presence of Cr(VI) in the environment has selected various microbes and plants, to tolerate high levels of Cr compounds. The purpose of this study was to isolate the chromium resistant bacteria and to check the inoculation effect of chromium resistant bacteria on growth characteristics of wheat plants. For this purpose, total twelve bacterial strains were isolated from the Garden soil of MMG. Out of twelve, seven bacterial strains were selected on the basis of their resistance to tolerate high concentration of chromium (500 mgl⁻¹). Morphological, biochemical and physiological studies revealed that the selected bacterial strains belong to genus Bacillus. Cr(VI) reduction potential studies showed that the selected strains exhibited excellent chromium reducing capability(15-75%). Growth curve experiment exhibited that bacterial isolates showed best growth at 37° C and pH 7. The selected strains showed multiple resistance to various heavy metals (Cu, Ni, Co, Hg Zn) and antibiotics (Erthromycin and Gentamycin). The isolated strains showed positive results for certain plant growth promoting characters like auxin production, siderophore production, phosphate solubulization, and hydrogen cyanide test. Plant microbe interaction studies of monocultures and mixed cultures (both in labs as well as in fields) revealed that inoculation of plants with mixed cultures exhibited much better growth as compared to the monocultures that might be due to symbiotic relationship between various bacterial strains. Cr(VI) had exerted adverse toxic effects on plant microbe interaction.

Keywords: Chromium toxicity, Chromium reduction, Chromium resistance, Wheat plant

BPC-79

In a refinery at reformer

Umar Siddique

Department of Chemical Engineering & Technology University of Engineering & Technology Lahore KSK Campus.

Abstract:

The term "Reform" means "Re-arrange". The Reforming Process is the re-arrangement of molecular structure of hydro-carbons by converting long chain n-paraffins first to branch chain iso-paraffins and from branched chain iso-paraffins to the cyclic form of paraffins, usually called as "Naphthenes" or "Cyclo-Paraffins". Aromatics are obtained catalytically due to their high octane ratings under a couple of reactions like naphthenes dehydrogenation, isomerization of naphthenes and paraffins, dehydro-cyclization of paraffins, hydro-cracking, de-methylation and

de-alkylation. The catalytic reforming process has been a mainstay in most refineries throughout the world for many years. The original function of this process is to upgrade low octane number straight-run naphtha to higher octane motor fuel blending components by catalytically promoting specific groups of chemical reactions. Naphtha boiling range products from other processes (thermal cracking, coking, etc.) are soon being included in the charge to catalytic reforming units for octane improvement. The reforming application is logically and-rather quickly expanded to include the production of specific aromatic hydrocarbons. High-purity benzene, toluene, and mixed xylenes are made available to the chemicals industry from petroleum fractions by the combination of reforming, aromatics extraction, and fractionation. Hydrogen, the "by-product" from the aromatic producing reactions, is found to be useful in supporting the operation of reformer feed preparation units as well as other hydro treating units. The light hydrocarbon gases, by-products of the cracking reactions, are generally added to refinery fuel gas systems. Butanes, other cracking by-products, are commonly used in adjusting vapor pressures of gasoline pools.

Keywords: Reforming, Naphtha, Aromatics, High Octane, Catalysts

BPC-80

Effectiveness of Social Networking Sites in creating brand image among youth in Pakistan Nadeem Iqbal, Pirzada Sami Ullah and Zahra Amjad

Business Administration, Superior University Lahore, Raiwind Road, Lahore, Pakistan.

Abstract:

Massive popularity of social media with 1.43 billion social network users round the globe in 2012, representing a 19.2 percent increase over 2011 figureswas predicted. Considering this huge usage, Social Networking Sites (SNSs) have been markedly adopted by marketing professionals for creating, modifying, and enhancing their brand image amongst their target customers. This very use of the SNSs sets the argument for conducting a study to investigate the role of SNS and measure the level of its effectiveness in creating brand image in the customers specifically among youth. That's why this research was aimed at measuring the Effect of SNSs and determining their impact on the political mind-set of youth in Pakistan.A social media user survey was conducted to cope with the objectives of the study. Primary data were collected from 300 SNS users, ageing between 15 to 30 years, belonging to three major cities of Pakistan. Marketing Blogs and enterprise pages on SNSs were thoroughly explored to locate and select the appropriate sample for data collection. Later on close ended questionnaires were distributed among Social Media Users (SMUs) online (173) whereas 127 were visited personally. Data was analyzed through SPSS software to draw results. The interest of youth in social media was found very high as 83% of respondents revealed that they spend 5 to 10 hours on social media either through computers or through mobiles. 53% of the respondents shared that they prefer to get knowledge about products/brands through social media. Out of which 14% develops a brand image based on SNS information where as 52 % uses this information to modify their existing brand images whereas the rest of the respondents take this information just for their awareness of the products, brand, and market trends. 47% of the respondents believed the social media as factual and truthful whereas 38% considered that this information is not trustworthy while rest of the respondents didn't argue. 78% respondents considered it a best source of information regarding market and new products. 63% respondents perceived social media as positive tool for marketing and branding believes where as 16% consider it not. The rest of the respondents take it

both positive as well as negative. The correlation statistics show strong relationship between SNS and brand image (r = .673*) whereas regression show that social media causes 37% change in the brand image of the youth. The findings reveal that SNSs are a strong and growing technology with very high potentials and implications regarding their role in marketing and branding of products and services. It is the cheapest way to establish a strong brand image among target market. These findings are high value able for all sort of marketing and branding professionals. Besides this, the study, being unique in its theme, reveals the scope of modern technologies in marketing and branding fields. The study will set grounds for further researches in the area, helping scholars reveal hidden benefits, uses and abuses of social media to make its role and implication more value able.

Keywords: Social Media, Social Networking sites, Branding, Brand image, youth

BPC-81

Impact of entrepreneurship education on intention and desire for venture creation (An empirical study of entrepreneurs and non-entrepreneurs graduates)

Pirzada Sami Ullah, Muhammad Rafique and Nadeem Iqbal Business Administration, Superior University Lahore, Raiwind Road, Lahore, Pakistan.

Abstract:

The scope of entrepreneurship education is increasing round the globe butin Pakistan, Entrepreneurship education is being neglected because of unawareness of its paybacks. Realizing the significance of entrepreneurship education, this research aims at investigating the scope of entrepreneurial education in developing and designing creative ideas and implementing them innovatively. Furthermore it is also aimed at determining the impact of entrepreneurship education on intention and desire for venture creation among university students in Pakistan.

This study uses positivism paradigm. A surveywas conducted from 160 suitable respondents from private and public sector educational institutes by using close ended questionnaires. For explanation, this study used a five point Likert scale questionnaire. The students, who have taken entrepreneurship education and who have not, were the respondent's category to explain the results. Statistical Package of Social Sciences (SPSS) software wasused to analyse the results. Results and analyses were derived byindependent sample t-tests with the help of SPSS software to measure the significance and difference of two groups. This study is focused only on the students studying in Lahore and Faisalabad with the sample size of 160 participants. The sample size is narrow which can hinder the generalised results of study and results cannot be applied for general study of Pakistan because of cultural differences. Financial implication on new creation is missing which may change the effect of study. Further studies can accommodate these limitations for more effective results. The study proposes that education has strong impact on venture creation. The candidates who hold an entrepreneurship degree grow exponentially by utilising opportunity, ability and situation (O-A-S). Knowledge of venture creation and confidence to venture have more impact on the establishment and growth of the venture.

Literature recognises that entrepreneurship education and research on its importance is minute. So, this study will suggest avant-garde techniques for graduates to start new ventures. The results of this study will help graduates to make economy stronger and independent by adopting new businesses. This study will open gate for new researchers to invent more creative techniques.

Keywords: Entrepreneurship, Education, Team Building, Intention, Desire, Trust, Venture Creation

BPC-82

Extraction and purification of Polyhydroxyalkanoates from gram positive and gram negative bacterial strains

Nasir Javaid and Nazia Jamil
Department of Microbiology and Molecular Genetics, University of the Punjab,
Lahore 54590, Pakistan.

Abstract:

Various bacterial species accumulate intracellular polyhydroxyalkanoate (PHA) granules as energy and carbon reserves inside their cells. This work was done to find out the ability of various bacterial strains to produce PHA using different carbon sources. All strains were identified by basic microbiological and biochemical tests. After that their lipase activity was checked by using Tributyrin agar followed by PHA detection by using PHA detection media. PHA positive strains were further checked out for the quantity of PHA produced by them in PHA screening media with glucose and canola oil as carbon sources. Strains 1,2 and 5 showed lipase activity while strains 3 and 4 did not show. In nitrogen limiting conditions, Strain 1 showed maximum growth at 72 hrs in both carbon and canola sources. Strains 2, 4 and 5 also showed same trend like strain1, while strain 3 showed maximum growth in ,case of, glucose at 48 hrs and canola at 72 hrs.All Strains either lipase positive or lipase negative can produce PHA in diffrent environmental contions i.e nitrogen limitation. Strains 1, 2, 3, 4 and 5 showed maximum PHA 66.7%, 77.8%, 75%, 76.9% and 66.7% respectively.

Keywords: Polyhydroxyalkanoates, Renewable, Lipase, Canola oil

BPC-83

Association between ACE I/D gene polymorphism and hypertension in smokers

Hateem Zafar Kayani, Sana Riaz and Nageen Hussain
Department of Microbiology and Molecular Genetics, Quaid-e-Azam Campus, University of the
Punjab, Lahore, Pakistan

Abstract:

Hypertension is a major and common health problem which is associated with many risk factors. It usually results in the rise of the blood pressure due to arterial stiffness and deficiency of oxygen. The precise factors responsible for hypertension are largely unknown. Smoking is a common risk factor associated with hypertension and cardiovascular diseases. The main aim is to study the association of ACE I/D polymorphism with healthy smokers. A total of 49 smokers and 49 healthy controls were enrolled in this study. The general population of Rawalpindi and Lahore was selected as an area of study. Intron 16 of the ACE gene was amplified by using nested PCR. The possible banding patterns were as follows; DD, 210 bp fragments; II, 264bp and 498 bp fragments; ID, 210 bp, 264 bp, and 498 bp fragments. The subjects comprised of males only. Among 49 subjects the frequency of ACE DD genotype was 75.5% (37/49), II 24.5% (12/49), while no ID genotype was found in the study. All controls showed DD genotype. The association of ACE I/D polymorphism with different levels of blood pressure were found to be significant.

Keywords: ACEI/D Polymorphism, Hypertension, Smokers

BPC-84

Callus induction and plant regeneration in three local cultivars of Indica rice (Oryza sativa.L)

Abdul Rehman, Khurram Liaqat and Tahira Malik
Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-i-Azam
Campus, Lahore 54590, Pakistan.

Email: malitah@yahoo.com

Abstract:

Improvement of rice production is mainly dependent on the suitable regeneration protocol for the locally preferred rice cultivars. In this study we tried to establish a process of rice callus induction in three local rice cultivars. Seeds of three rice varieties (super basmati, MG and 1121) were evaluated for in vitro callus induction using Murashige and Skoog medium (MS) medium supplemented with different concentrations (2mg/l, 2.5mg/l, 3mg/l and 3.5mg/l) of 2,4-Dichlorophenoxy acetic acid (2, 4-D). The influence of two carbon sources i.e. maltose and sucrose on callus induction at different concentrations (3%, 4% and 5%) were also investigated. Mature seeds of all varieties were used as starting material for callus production. Different concentrations of sodium hypo-chlorite were used to optimize the sterilization of seeds and 90% sodium hypo-chlorite was proved to be most effective. Cultured seeds were incubated at 25±2°C for 2 weeks in dark. Primary cultures were transferred to fresh media after every 3 weeks interval. Maximum callus induction was observed at 3mg/l concentration of 2, 4-D in all rice varieties. At 3mg/l concentration of 2, 4-D, the percentage of callus induction was maximum for super basmati (85%) followed by MG rice (82%) and 1121 rice (75%). Calli obtained at 3mg/l concentration of 2, 4-D were large, creamy, nodular and embryogenic as compared to calli obtained at other concentrations of 2, 4-D. Among different sugar sources, maltose was most effective, resulting in highest frequencies of embryogenic callus formation. At concentration of 4% maltose, 76% callus induction was observed. An efficient regeneration is pre-requisite for successful genetic transformation in addition to produce soma-clonal variants.

Keywords: Oryza sativa. L, MS: *Murashige and Skoog medium*, 2, 4-D (2, 4-Dichlorophenoxy acetic acid), Callogenesis, embryogenic callus.

BPC-85

Rice husk as an efficient biowaste ingredient for the removal of Cr (VI)

Aniqa Naeem and RidaBatool

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus Lahore-54590, Pakistan.

Email: ridazaidi_1@yahoo.com

Abstract:

Cr(VI) is a renowned toxic and carcinogenic heavy metal to biological system. The aim of study is the removal of Cr(VI) from aqueous solution by usingrice husk, chromium resistant bacteria and rice husk coated with bacterial strains. Previously isolated chromium resistant bacterial strains belonging to *Exiguobacterium*; E1 and E4 were checked for their EPS production ability. It was observed that stress conditions and presence of glucose enhanced the EPS production ability of both bacterial strains. Among chemically modified rice husk, citric acid treated rice husk showed significant removal of Cr(VI). Rice husk treated with different chemicals (HCl, H₂SO₄, formaldehyde, citric acid and phosphate), raw rice husk, boiled rice husk and both

bacterial strainsE1 and E4 exhibited \leq 50% of Cr(VI) removal from aqueous solution after 24 hours of incubation. Citric acid treated rice husk was coated with E1 and E4 separately and checked the Cr(VI) removal after optimized the physiological conditions. More than 99% removal of Cr(VI) was noted at 7 pH, 37°C temperature, 150 rpm shaking speed by using1.5g of rice husk from the 100ml solution having the conc. of $1000\mu g/ml$ of K_2CrO_4 . 100% Cr (VI) removal was observed in case of column experiments where solution of Cr(VI) was passed through bacterial coated rice husk filled glass column providing all the optimized conditions within 24 hours. Uptake of Cr(VI) was also studied to confirm the biosorption of Cr(VI) by rice husk. FTIR analysis indicated the contribution of various functional groups in the binding of Cr(VI) on the surface of rice husk.

Key words: Cr (VI) removal, Exiguobacterium sp., Rice husk, FTIR spectroscopy.

BPC-86

Change in *Arabidopsis* root architecture by *Pseudomonas fluorescens* WCS355 and PFO-1 in MS media and system

Atia Iqbal and Shahida Hasnain
Department of Microbiology and Molecular Genetics,
University of the Punjab, Lahore-54590, Pakistan.
Email: pisces_dream2000@yahoo.com

Abstract:

In the present study, The *Arabidopsis thaliana* was used to characterize the change in root morphogenesis caused by inoculated *Pseudomonas* strains. When the auxin production ability of these two strains was evaluated, they were found to produce auxin ranging from 4.6 to 84.6 μg/ml without and with the amendment of L-tryptophan by the Salkowski's method that was further confirmed by high performance liquid chromatography method. The strains were further studied for their potential to change the root architecture of model plant. *Arabidopsis Thaliana* (*Col-0*) showed contrasting responsiveness in root development when inoculated with these two *Pseudomonas* strains in MS media and sand system in *invitro* condition by increasing primary root length and lateral root densitysignificantly (P<0.05). *Arabidopsis thaliana* reporter plants (DR5: GUS) showed an increased in the endogenous auxin in vasculature of primary and lateral roots when compared to the non-inoculated plants. In the present study the sand system is a model system close to the natural environment to investigate root architecture of the plants without damaging delicate roots as compared to MS media or natural soil. Furthermore, the selected bacterial strains were found to be an efficient candidate to be used as an agriculture input.

Keywords: *Pseudomonas*, Rhizobacteria, *Arabidopsis thaliana*, Auxin

BPC-87

Effect of FTO gene variants on familial obesity in local population

Shabana and Shahida Hasnain
Microbiology and Molecular Genetics, Quaid-e-Azam Campus
University of the Punjab, Lahore-54590 Pakistan.

Abstract:

Obesity is an oligogenic disease whose development can be modulated by various polygenic (modifier) genes and by environmental influences. The prevalence of obesity has dramatically

increased in the last thirty years and need has arisen to understand genetic basis of obesity. Success has only been achieved in the identification of monogenic forms of obesity. Five human monogenic obesity genes involved in food intake regulation pathway have been determined named leptin, leptin receptor, proopiomelanocortin, melanocortin 4 receptor and proconvertase 1 genes. Genome wide association scans showed association of FTO gene variants with obesity. We used tetra amplification refractory mutation system (ARMS) PCR to detect the frequency and association of SNPs rs9939609, rs8050136 and rs1121980 with familial obesity. Seven obese families were selected for this purpose. rs9939609 was genotyped using standard PCR as well followed by restriction analysis. Results showed the presence of rs9939609 in 2% of the population with allele frequencies of 0.99 and 0.01 for T and A allele, respectively and genotype frequencies of 0.98, 0.19 and 0.01 for TT, TA, and AA, respectively. For rs8050136, allele frequencies for C, and A allele were 0.994, and 0.006, respectively while genotype frequencies for CC, CA, and AA were 0.988, 0.119, and 0.003, respectively. For rs1121980, allele frequencies of C, and T alleles were 0.987 and 0.013, respectively while genotype frequencies were 0.974, 0.256 and 0.01 for CC, CT, and TT, respectively. Pedigree analysis showed that these SNPs are present at random, appear spontaneously and are not inherited in the families therefore; these are not associated with familial obesity.

Keywords: Obesity, Monogenic, Leptin, FTO (fat mass and obesity associated), Genotype, Familial

BPC-88

Effect of magnetic nanoparticles on cell wall constituents of bacteria isolated from dental unit water lines

Sumreen Hayat and Anjum Nasim Sabri
Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam
Campus, Lahore 54590, Pakistan

Abstract:

The present study was designed to study the effect of magnetic nanoparticles on cell wall of bacteria isolated from dental unit water lines. The antibacterial activity of nanoparticles was determined by agar well diffusion method. The results showed that magnetic nanoparticles exhibited their antibacterial effect in terms of zone of inhibition. Furthermore, there was a decrease in teichoic acid content in cells treated with magnetic nanoparticles while peptidoglycan (PG) content was found tobe increased in cells treated with nanoparticles. In short, magnetic nanoparticles manifested their effect on growth and cell wall constituents of bacteria isolated from dental unit water lines.

Keywords: Nanoparticles, Peptidoglycan, Teichoic acid.

BPC-89

Microbial Evaluation of Unprocessed And Processed Meat At Low Temperatures

M. Aurangzeb¹, Ayesha Ilyas² and Quratul- Ain-Syed³

Govt. College Township, Lahore.

²F. C. College, Lahore.

PCSIR, Laboratories Complex, Lahore.

Abstract:

The microbiology of meat and its products is often investigated in order to determine the safety and keeping quality. The characteristic microbial population that develops in meat and its products reflects the prevailing environmental conditions, to which it is exposed. The resent research work was undertaken to investigate the microbial levels of different types of meat at refrigerated and frozen conditions. Five samples of each three different types of raw minced meat i.e. Chicken, Mutton and Beef at chilled and frozen conditions were analyzed. Four processed meat samples, five each of meat balls, chicken samossas, drumsticks and beef shame kebab were also evaluated for their microbiological examination, under frozen conditions. The parameter included Psychrophiles, Mesophiles, and Anaerobes, Total coliforms, fecal coliforms, *E. coli, staphylococcus aureus* and *Salmonella spp*. During chilled and frozen conditions, Psychrophiles were the most commonly growing microorganisms. The growth rate of mesophiles and anaerobes was less in the above mentioned conditions. The growth of pathogenic microorganisms, like E.coli, Staphylococcus and salmonella was less common in the chilled and frozen samples.

Keywords: Meat, Pathogenic microorganisms, Psychrophiles.

BPC-90

Molecular characterization of *bio plastic* producing *pseudomonas spp.* isolated from industrial and domestic waste water

Saiqa Tufail, Hasnain Javed and Nazia Jamil
Department of Microbiology and Molecular Genetics,
University of The Punjab, Quaid-e-Azam Campus. Lahore, Pakistan.

Abstract:

Polyhydroxyalkanoates (PHAs) are raw materials for production of biodegradable plastics, generated by a range of microbes, cultured under different nutrients and experimental conditions. The aim of this study was the molecular characterization of bioplastic producing bacteria especially Pseudomonas species isolated from minimal environmental conditions (domestic and industrial soil and waste water)and to develop a biological process to produce PHAs from carbon source other than glucose. PHA was produced by the cultivation of bacteria with Mustard oil in lab scale fermenter. No previous work has been reported using mustard oil as carbon source using this strain. Fermentation and PHA production was done in batch culture in 5 liter fermenter to optimize the PHA production under various experimental conditions. Total of 67 bacterial strains were isolated and purified from industrial and domestic waste water and sewage sludge different regions of Pakistan. Isolates were checked for PHA production by Sudan black and Nile blue staining. Quantitative analysis for biodegradable plastic produced by different bacterial species was performed by Modified surfactant hypochlorite. High PHA production was detected in 35 strains belonging to different genera including Pseudomonas and others as predominant genera. From 35 strains best strain was selected for further work. PHA production under different carbon sources, pH and temperature was estimated. PHA production of Pseudomonas species

were subsequently authenticated at molecular level by PCR analysis and the phaC gene (540 bp) encoding PHA synthase was amplified. After phaC gene sequencing and 16SrRNA ribotyping, most of the phaC gene containing strains showed homology to *Pseudomonas aeruginosa*. Positive samples yield a specific 540-bp PCR product representing partial coding sequences of the phaC1/C2 genes. PhaC gene operon was amplified. PHA polymerase 1 (PhaC1) and PHA polymerase 2 (PhaC2) from *Pseudomonas aeruginosa* were screened and sequenced and submitted to gene bank with accession number bankit1367455, bankit1367459 and bankit1367463 with PCR product size of 1300 bp and 1700 bp respectively.

Keywords: Polyhydroxyalkanoates, Biodegradable plastic, *PhaC* gene

BPC-91

Microbial Studies of Fruits and Vegetables Stored At Low Temperature

Maliha Islam, Shazia Kanwal Malik and Hamad Ashraf Biotechnology Lab, Department of Botany, Govt. College of Science Wahdat Road Lahore. email:kanwal707@yahoo.com

Abstract

The present study is based on the microbial examination of fruits and vegetables stored in Refrigerator. Different samples of fruits and vegetables i.e. tomato, lemon, apple, pomegranate collected from refrigerator stored at home. The samples were examined for fungi and bacteria and these samples were than cultured on the agar plates. The agar plates were incubated at low temperature (4-6°C) as well as optimal temperature. The specimens were found to be highly contaminated with fungus as compared to bacteria. The fungus were identified and found to be i.e. Yeast, Aspergillus niger, Aspergillus oryzae, Mucor, Halmintosporium, Pl and Alterneria alternate. Bacterial colonies were observed and the colour of colonies was pink, yellow and white. The gram staining was performed and many cocci, bacilli and spore forming bacteria were observed. From the present study it is investigated that the storage of vegetables and fruits at low temperature (2-10°C) is not sufficient to prevent microbial growth. So, it is recommended that the fruits and vegetables will not be stored for more than 3 days in the refrigerator

Key words: Aspergillus niger, Aspergillus oryzae, Mucor, Alterneria alternate, Halmintosporium

BPC-92

Antibacterial activity of *Nigella sativa* (Kalvanji) against different strains of *Salmonella* characterized by microsatellite fingerprinting and 16s rRNA sequencig

Arslan Sarwar and Zakia Latif

Department of Microbiology and Molecular Genetics, University of the Punjab Lahore, Pakistan. Email: arslan.microbiologist@gmail.com

Abstract:

Salmonella is a food borne pathogen and causative agent of human Salmonellosis and Typhoid fever causing many deaths in developing countries. Unnecessary use of antibiotics have stimulated the bacterial resistance which has adverse effects associated with the significantly increase in the occurrence of infectious disease. Therefore, it is a need to develop alternative antibacterial drugs from medicinal plants and other natural extracts for the treatment of

infectious diseases. The present study was planned to investigate the antibacterial effects of Nigella Sativa (Kalvanji) against different strains of Salmonella. Sixty bacterial strains were isolated and purified from different environmental sources including drinking water, waste water, sewerage water, poultry, beef, eggs, fruits, vegetables and clinical samples. After biochemical analysis, twelve bacterial strains were confirmed as Salmonella. Antibiotic susceptibility test was done by well diffusion assay against different concentrations of Ceftriaxone and Ciprofloxacin. The behaviour of both antibiotics was different against Salmonella strains. Six Salmonella strains resistant to both antibiotics were analysed for the antibacterial activity of natural extracts of Nigella Sativa(Kalvanji). Partitioning of Kalvanji powder was performed with Ethyl acetate. Nigella sativa oil extract was found to be more effective against Salmonella species for which even Ceftriaxone and Ciprofloxacin were ineffective. The genetic diversity of the selected Salmonella strains were analysed by microsatellite fingerprinting further molecular characterization of the selected strains was done by 16s rRNA sequencing.

Keywords: Antibiotic susceptibility, Salmonellosis, microsatellite fingerprinting

BPC-93

Bioremediation of Mercury Compounds by Using Immobilized Nitrogen-Fixing Bacteria

Aysha Tariq and Zakia Latif

Department of Microbiology and Molecular Genetics, University of the Punjab, Quaid-e-Azam Campus, Lahore

Abstract

The mercury contamination is of great concern because of its toxicity and ubiquity. The environmental level of mercury is rising day by day because of anthropogenic activities. Bearing in mind the toxicity of mercury, in this study, we have isolated various strains of nitrogen-fixing bacteria (NFB) from nodules of different plants on YEM medium. The preliminary screening of NFB strains to resist mercury was done by well plate assay. Characterization of the selected strains for the production of hydrogen sulfide (H₂S) was done by growing them on LA medium. The hydrogen sulfide producing NFB strains co-precipitated the mercury in the form of HgS resulting in transformation from toxic (Hg²⁺) to non-toxic (Hg⁰) form. Further screening was done by quantitative assay by using Dithizone method. Mercury resistant and hydrogen sulfide producing NFB strains, characterized by biochemical tests as *Enterobacter, Cronobacter and Pseudomonas*, gave the most promising results in the detoxification of mercury. Finally these strains were immobilized in sodium alginate (synthetic beads) and their ability to detoxify mercury containing industrial effluents was determined as compared to free cells by Dithizone method. It was concluded that immobilized NFB bacteria detoxified more concentration of mercury as compared to free cell cultures.

BPC-94

Nano-gold particles mediated detection of NS1; an early diagnostic marker of Dengue virus infection.

Abid Hussain and Zaigham Abbas

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore, Pakistan.Email: abihqureshi@gmail.com

Abstract:

Dengue is one of the emerging arbo-viral infections, globally approximately 2.5 billion people are in peril. Dengue virus is prevailed in many areas around the globe including the tropical and sub-tropical areas of the Asia. 50-100 million cases being reported each year. Different techniques are used for the diagnosis of dengue virus infection like virus isolation, RNA detection, Immuno histochemistry, Ig M, Ig G detection and NS1 detection by ELISA but these techniques have the limitation of high cost, expertise and late diagnosis. Immuno chromatographic based lateral flow device using colloidal gold particles as detectors labeled with anti NS1 antibody for the diagnosis of NS1 antigen (an early diagnostic marker) of dengue virus developed in this study. Device comprises of sample pad, conjugation pad, nitrocellulose membrane and absorbent pad. Nitrocellulose membrane contains test(anti Ns1 antibody) and control(anti mouse IgG) region. Assay device was evaluated by purified NS1 and prior ELISA confirmed NS1 positive and negative samples, The results ensured rapid, early, cost effective, easy to use and specific diagnosis of the Dengue viral infection.

Keywords: Dengue Virus, ImmunoChromatographic Technique (ICT), Colloidal Gold Particles, Rapid Diagnosis, NS1 protein.

BPC-95

Screening and physiochemical characterization of bacteriocin produced by Rhizopheric bacteria

MisbahAslam and NaziaJamil

Department of Microbiology and Molecular Genetics, New campus, University of the Punjab, Lahore. Email: misbahmmg@gmail.com

Abstract:

The present study was carried out for screening and molecular characterization of bacteriocin produced by bacteria isolated from rhizosphericsoil. Rhizosphericsoil samples were collected from the indigenous areas of Punjab University to isolate bacterial strains that exhibit the antibacterial activity. Extraction and purification of the bacteriocin was done by TLC and SDS-PAGE. Purifiedextracts were treated with high temperature 121°C, Proteinase K and UV radiation to check the stability of the bacteriocin. Five out of ten bacterial strains (AZS, A2, P, U and LB) showed high bacteriocin activity against the target organism. The stability of the crude extract of bacteriocin to high temperature 121°C, Proteinase K and UV radiation indicated that these compounds could be a better option for utilization as a probiotics. The SDS-PAGE analysis indicated that bacteriocin were low molecular weight peptide i.e. molecular weight of *Brevundimonas* and *Bacillus* (A2, AZS & LB) was approximately in the range of 10 to 25 kDa whereas the molecular weight of *Pseudomonas* and *Alcaligenes*(P & U) was approximately 40 kDa.It was concluded that bacteriocinsare low molecular weight compounds with diverse stability which indicated that these compounds could be a better alternative to antibiotics.

Keywords: Transformation, bacteriocins, plasmid, SDS-PAGE.

BPC-96

Impact of Single and Mixed Species Inoculations on the Growth Promotion of Economically Important Crops

Saiga Razi

Department of Biology, Lahore Garrison University, DHA Phase VI, Sector C, Lahore.

Corresponding author: saiqarazi@gmail.com Keywords: Industrial pollution, Bioremediation, plant-microbial interaction

ABSTRACT

Pakistan is an agricultural country, where industrialization is taking place in a gradually increasing phase. Agricultural irrigation with industrial wastewater is a common practice in arid and semiarid regions and it is used as a readily available and inexpensive option to fresh water. Excess of heavy metals in water results in low crop yields, and soil deterioration occurs as well. The inoculation of pollutant-degrading bacteria on plant seed can be an important additive to improve the efficiency of phytoremediation (Kuiper *et al.*, 2003).

AIM: The aim of this study was to investigate the plant-microbial interactions between pollutants-resistant bacteria and the economically important crops. The strains used in this study were isolated from industrial wastewater of a local tannery. Three strains *Bacillus thuringiensis*, Enterobacter sp. and Pseudomonas fluorescnes were used in this study.

The seeds of all crops were inoculated with single species as well as mixed culture inoculations were also tested. The plants were grown in pots and plates and the growth parameters were determined after appropriate time interval. *Bacillus thuringiensis* was tested for its ability to colonize roots and to promote the growth of *Triticum aestivum*, *Helianthus annuus*, and *Cicer arietinum* alone or in combination with Enterobacter sp. and Pseudomonas fluorescnes. All three strains were able to colonize the root system of crops but only B. *thuringiensis* and P. fluorescens promoted plant growth in single strain inoculation tests. Mixed strain inocula were less effective than single ones. Moreover, in one case, the dual strain inoculum (B. *thuringiensis* and P. fluorescens) did not have any significant effect on plant growth in contrast to the separate inoculation of both strains. Bacterial inoculations improved the growth of all crops. However establishment of large populations of bacterial inoculants on roots did not appear to be essential for plant growth promotion.

BPC-97

Impact of *Synechocystis* AHZ-HB-MK and its derivatives on the growth and biochemical parameters of *Triticum aestivum* under chromium stress

Saiqa Razi and*Shahida Hasnain

Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore 54590, Pakistan

ABSTRACT:

Application of hexavalent chromate adversely affects the growth and anatomical characters of *Triticum aestivum*. *Synechocystis sp.* AHZ-HB-MK (DQ381960) isolated from a local chromium contaminated industrial site and was able to tolerate up to 200 g/ml Cr (VI). The ability of *Synechocystis* to reduce toxic chromate was increased after exposure to different doses of gamma rays (0.5, 1, 2, 5, 10 and 20 Gy) generated by a ⁶⁰Co source at different stages of growth (i.e. after 5, 15, and 30 days of incubation) (Razi and Hasnain, 2006). Gamma radiation enhanced the

chromium reduction potential at all stages of growth. A maximum increase was obtained at low-irradiation doses. In this study selected mutants with increased reduction potentials were used to analyze their impact on the growth and biochemical parameters of *T. aestivum* grown under Cr (VI) stress. *T. aestivum* seeds were inoculated with either irradiated or non-irradiated *Synechocystis* AHZ-HB-MK and grown in pots in quadruplicated under controlled conditions. Non-inoculated treatment was considered as control. Chromium salts (300 g/g) were added to the soil. After 10 days, the seedlings were harvested and different growth and biochemical parameters were studied and compared with non-inoculated and non-irradiated controls. All of the selected mutants enhanced the growth. Mutants obtained at low-irradiation and early growth stage gave a maximum increase. These mutants can be used in future for the bioremediation of chromium contaminated soils and wastewater and to enhance the growth of economically important crops at these sites.