MMG 2018

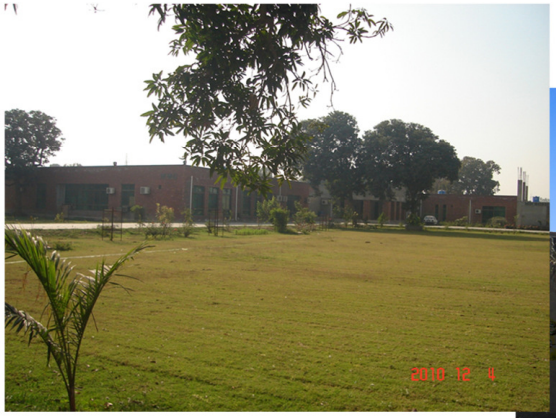
7-9 February 2018

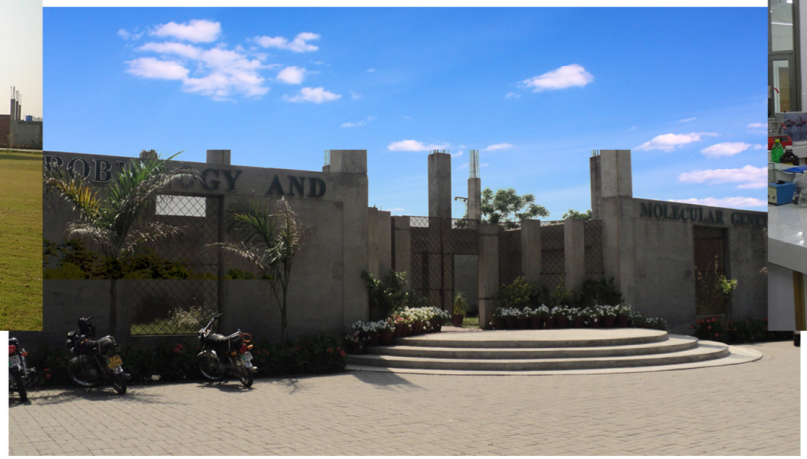
A Conference on
Microbiology & Molecular Genetics

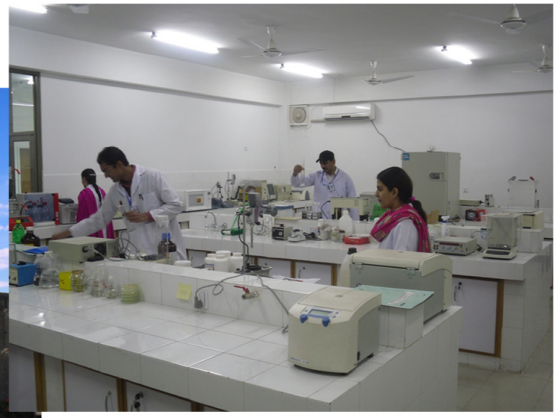
Organized by
Department of Microbiology & Molecular Genetics,
University of the Punjab
In Collaboration with
Department of Allied Health, Sciences, Superior University

Department of Microbiology & Molecular Genetics (MMG)

Department of Microbiology & Molecular Genetics (MMG) occupies a unique niche at the University of the Punjab, Lahore as its research and educational mission covers both Microbiology & Molecular Genetics, a combination which is not found in any other educational institute of Pakistan. The department was founded in 2002 and since then it is serving with great zeal and zest. MMG is highly committed to excellence in both research and education. Research is being carried out in a vast array of microbiology, biotechnology & molecular genetics. The department is home to undergraduate and postgraduate programs and students are passed through an activity based approach that makes them able to investigate and discover scientific concepts. The department is also recognized internationally by TWAS (Third World Academy of Science).

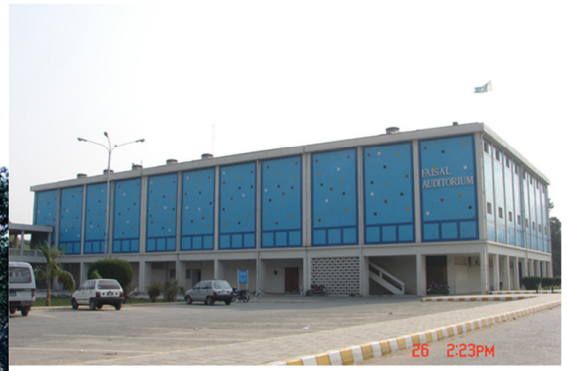






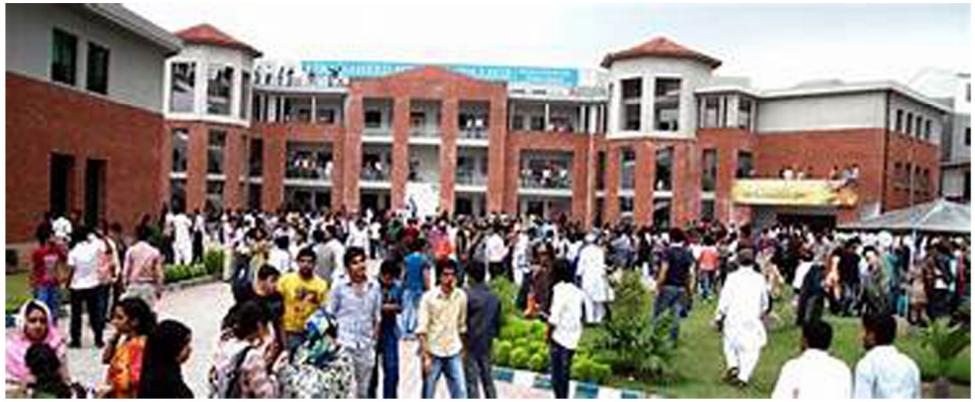
University of the Punjab, Lahore

The University of the Punjab is one of the oldest universities in South Asia and is in fact the mother of all other universities in Pakistan. It was founded in 1882, a campus now known as Allama Iqbal Campus. In order to cover all the new fields, a new campus (Quaid-e-Azam Campus) was founded with many new departments educating and researching in modern sciences and humanities. This is the campus housing the MMG department. And present a pleasant view with modern designed buildings, enormous plantation and canal.



Department of Allied Health Sciences (AHS)

Department of Allied Health Sciences presenting multiple disciplines i.e. Dietetics and Nutritional Sciences, Medical Lab Sciences, Medical Imaging Technology, Speech and language Pathology, Optometry, BS Biotechnology, BS Biochemistry and Biotechnology, BS Microbiology and Biotechnology and BS Molecular biology and Biotechnology The Allied Health Sciences department discipline is based on solid comprehensive curricula that emphasize quality, application to real-world situations, interdisciplinary learning, communication skills and team building.



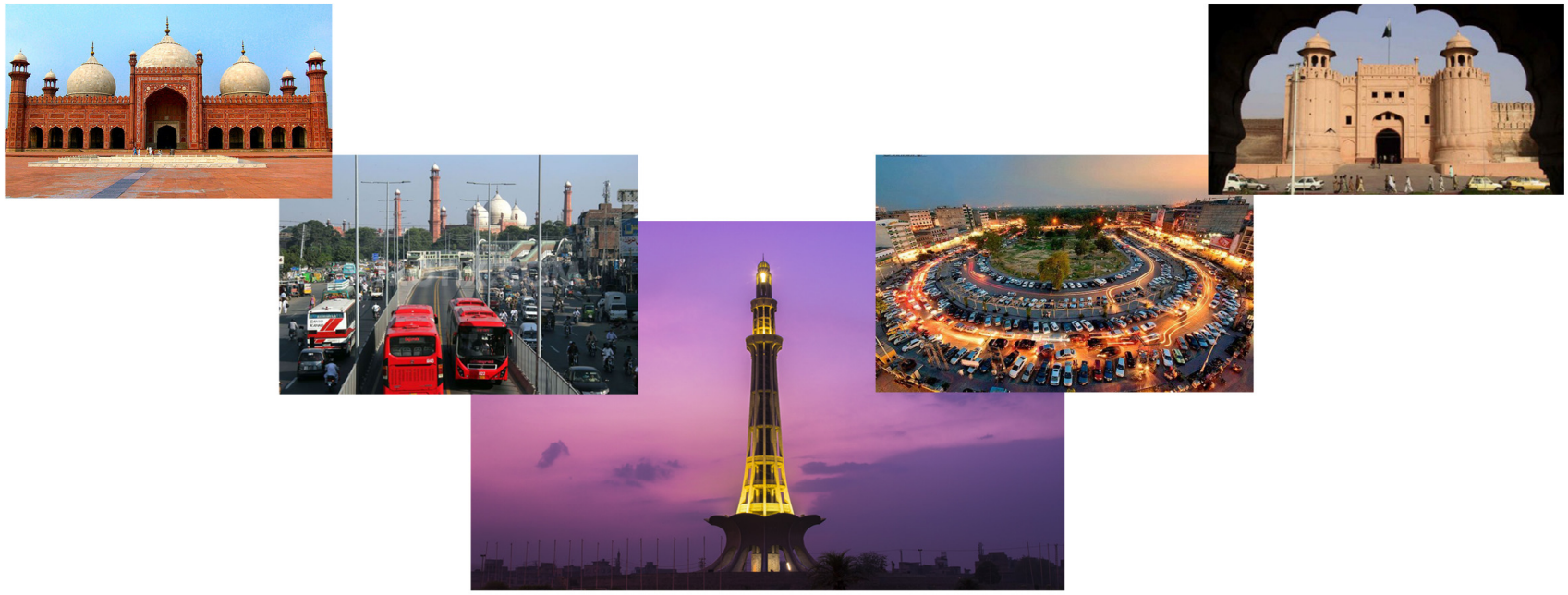
The Superior College, Lahore

Our history is overwhelmed with superior values which are depicted in every initiative we have taken. Facilitating superior human beings was the initiative behind the inception of Superior College Lahore. The dream of Prof. Dr. Chaudhry Abdul Rehman of an educated and energetic Pakistan drove him to establish this institute.

21st Century is the competitive era of creativity and innovation where only those universities will excel who have the ability to transform human resource into skillful human capital. We aim to develop passionate individuals who are willing to add value to the lives of society as a whole. At Superior University we teach Information Technology, Mass communication, Management, Medical Sciences, Allied Health Sciences, Biotechnology Sciences and many other initiative courses aiming to enhance the IQ and EQ within the student. Azra Naheed Center for Research & Development, Ch. Muhammad Akram Center for Entrepreneurship Development and Center for Human Resource Development are working to inculcate values in the students.

The City of Lahore

Lahore is perhaps one of the most attractive cities of South East Asia due to its rich history, culture and food. There are a lot of places to visit, to shop and to dine. Historical places such as Minar-e-Pakistan, Badshahi Mosque, Shahi Fort, Shalamar Gradens are worth mentioning. Shopping places include old shopping spots such as Anarkali bazar as well as modern shopping malls such as Pace and Hyperstar. Lahore is also known for its delicious traditional foods and Food Street and many other places are worth visiting.



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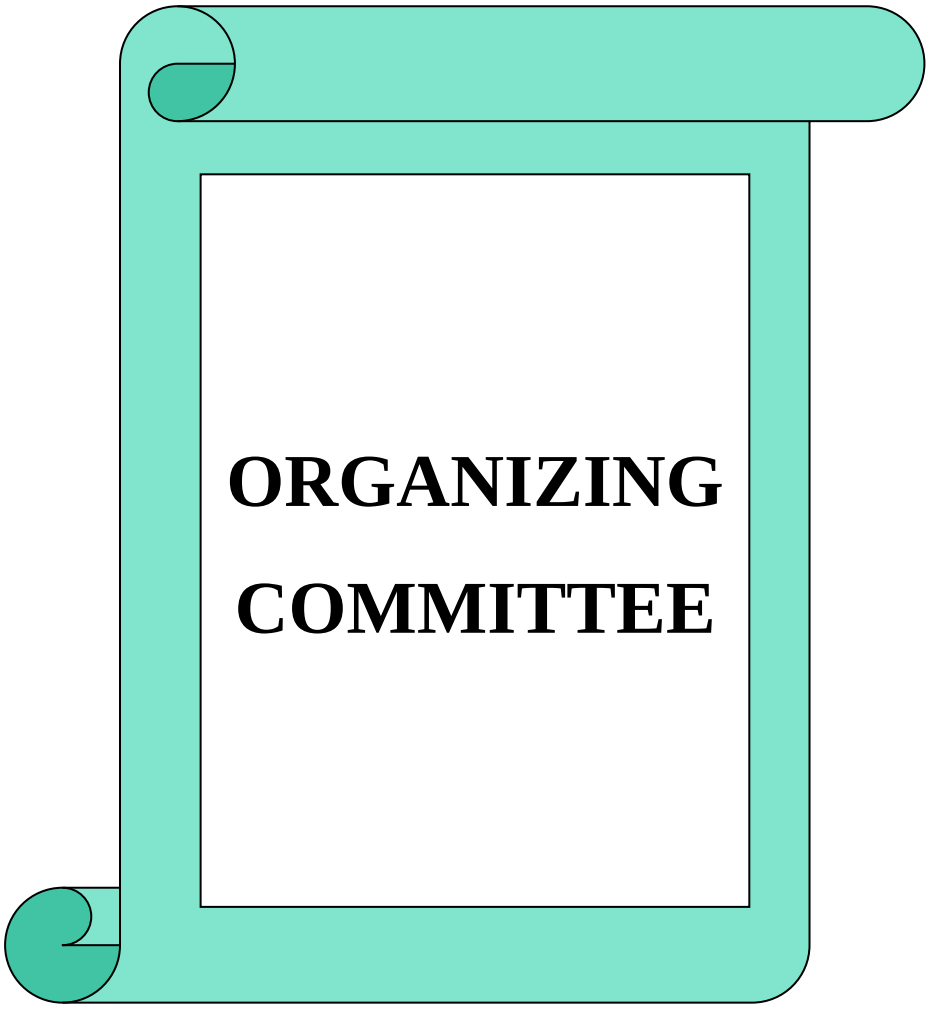

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INVITED SPEAKERS

MMG 2018
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Invited Lecture

IL001

Higher Education, Science, Technology and Innovation for Socio-economic Development

Prof. Dr. Atta-ur-Rahman, FRS, UNESCO Laureate, President NASIC
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We live in a world where truth has become far stranger than fiction. Each day brings thousands of new discoveries, many of which are transforming our lives in a multitude of ways. The blind can, amazingly, today see using their tongue elephant on strand of "graphene" that is 150 times thinner http://www.wicab.com/media/Wicab%20Press%20Release%203-19-2013.pdf. You can hang an than a human hair and the strand will not break since it is 200 times stronger than steel. The Harry Potter's disappearing cloak is now a reality by the discovery of metamaterials. Anything covered with metamaterials just disappears since they have the ability to bend light. Genes have been transferred from deep sea jelly fishes to orchids ---- the result are luminescent flowers that glow in the dark. Bullet proof paper has been developed through application of nanotechnology. Super-fast gene sequencing under development should allow the entire human genome to be sequenced in minutes! Objects can be moved by thought control and driverless cars are under development. Anti-ageing compounds have been discovered and when given to old mice, it made them younger! Stem cells promise to cure damaged organs and may change the manner in which medicine will be practiced tomorrow. Science today presents a myriad opportunity for research and exciting careers in many diverse fields. Some of these fascinating developments will be presented. Knowledge is now the single most important factor for socio-economic development and science & technology are great equalizers. Countries that have realized this and invested heavily in developing their human resources to the highest possible levels and then linked these resources to the manufacture of high technology industrial and agricultural products have leaped forward, leaving others far behind. The extent of darkness that prevails in the developing countries is apparent from the fact that while 90 Nobel Prizes have been awarded to faculty members of one University in UK, the University of Cambridge, and 32 Nobel Prizes have been awarded to faculty members of just one College of this University (Trinity College), not a single Nobel Prize has ever been won by a single scientist working in an Islamic country! We still live in the dark ages! Measures taken in Pakistan to improve science and higher education included a 6000 % increase in the development budget of science and a 3500 % increase in the development budget for higher education during the period 2002-2008. This led to spectacular progress. University enrolment has grown five-fold rising from only 276,000 in 2002 to about 1.4 million presently. The access to higher education grew from about 2.3% (of the age group17 to 23) in the year 2003 to about 10% of the same age group by the year 2016. A number of steps were taken to improve the quality of education. The most significant of these related to the programs to develop a strong faculty and

provide liberal research funding. Therefore about 11,000 scholarships were awarded to the brightest students of which some 5,000 scholarships were to obtain PhD degree sat top universities of the world. These and other such measures led to a sudden surge in university rankings. The research publications in journals with ISI impact factors went through an amazing increase from only about 800 per year in the year 2000 to about 10,000 per year by 2015, exceeding those from India on a per million population basis. They continue to rise by about 15% each year. Similarly the citations in the Science Citation Index increased by a 1000% in the same period. Thomson Reuters has published a glowing tribute to Pakistan about these developments in August 2016. Pakistan now needs to closely link these developments with the industrial growth planned in the China Pakistan Economic Corridor by establishing top level Centers of Excellence and Technology parks within the industrial clusters.

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Invited Lecture

IL002

Public health threat of food-borne bacterial pathogens: One health approach

Prof. Dr. Naim Deniz Ayaz
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Despite the improvement in consumer awareness, food hygiene and detection methods, foodborne diseases mediated by pathogenic bacteria or bacterial toxins still represent a significant threat to public health worldwide. Globally, WHO has estimated that; approximately 1.5 billion diarrhea episodes and more than 3 million deaths occurred in children under 5 years of age annually. A significant proportion of these results were caused from consumption of food, mainly food of animal origin contaminated with microbial pathogens and toxins. Approximately, 60% of the human pathogens are zoonotic and 75% of them are emerging zoonotic. Emerging foodborne pathogens are defined as those causing illnesses that have only recently appeared or been recognized in a population or that are well recognized but are rapidly increasing in incidence or geographic range. Emerging foodborne bacteria are reported as *Salmonella* (non typhoidal), *Campylobacter jejuni*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, *Staphylococcus aureus* (MRSA), *Vibrio vulnificus*, *Yersinia enterocolitica*, *Arcobacter* spp. and *Mycobacterium paratuberculosis*. *Salmonella* has emerged as a pathogen of significant public health concern in worldwide, and it is recognized as one of the major food-borne infection agent. According to the European Union (EU) and the United States (USA) data approximately 100.000 and 1.4 million cases occurs from *Salmonella* in each year, respectively. *L. monoctogenes* is a zoonotic food-borne bacteria that leads to a variety serious infections in humans such as encephalitis, meningitis, abortion and septicemia. Most *L. monocytogenes* strains can cause high morbidity and mortality depending on their virulence. *E. coli* O157:H7 is recognized as the major etiologic agent of hemorrhagic colitis (HC) and hemolytic uremic syndrome (HUS) in humans. Epidemiological studies indicated that the transmission of *E. coli* O157:H7 occurs through consumption of contaminated raw or undercooked meats of especially bovine origin such as minced meat and related products.

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Invited Lecture

IL003

Phage Therapy: Obstacles and Opportunities to Commercialization

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Phage therapy is often proposed as means of combatting the continued emergence of antibiotic resistant bacteria in clinical and community settings. The use of bacteriophages to treat bacterial infections, however, actually predates antibiotic therapy. The therapy is relatively low-tech, inexpensive, and, thus, potentially applicable to across economies and stages of development. To date, however, there are very few commercialized phage-based products available for use in human and/or veterinary medicine. Here, we will examine some of the barriers to commercialization of phage-based therapies, ranging from regulatory approval structures to efficacy and safety issues. We will propose some means to overcoming these barriers in effort to make phage therapy available to clinicians. Finally, we will discuss recent advances in phage therapy throughout the world and efforts to address remaining questions before phage therapy can develop into an effective, affordable, and accessible alternative to antibiotics in the treatment of bacterial infections.

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Invited Lecture

IL004

The virulence complex of the pathogen *Porphyromonas gingivalis*

Dr. Christine Ann Seers*, N Slakeski, JE Heath, PD Veith, LN Huq, SM Cleal and Eric C. Reynolds

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Chronic periodontitis, a complex inflammatory disease that causes loss of alveolar bone supporting the tooth, afflicts more than 10% of the adult population worldwide. *Porphyromonas gingivalis* is strongly associated with the aetiology of periodontitis. An important virulence factor of *P. gingivalis* is the cell surface protease-adhesin complexes (complex) comprised of tightly but non-covalently associated proteolytic cleavage products of RgpA-Kgp-HagA precursor proteins. The mechanism by which the protein fragments remain associated and are presented to the host has not been elucidated but is suggested to involve repeated sequences dubbed adhesion binding motifs ABM1 and ABM2.Methodology.We made a series of deletions and sequence rearrangements in Kgp ABM1 and ABM2 motifs and examined the effect on retention of the Kgp protease fragment (Kgp_{cat}) in the complex, using protease assay, SDS-PAGE and western blot. We modelled ABM1-ABM2 peptide structures and made structure-directed ABM point mutations. All ABM deletions and rearrangement mutations affected cell retention of Kgp_{cat} with Kgp_{cat} protease precursors released into culture fluid, supporting the contention that ABM1 and ABM2 are intra-complex binding motifs. Modelling indicated that ABM1 and ABM2 in separate protein fragments may combine in a novel non-covalent interaction to produce a module with a fibronectin type III-like structure. Each structure-directed ABM point mutation also affected cell retention of Kgp_{cat}, which validated the model. The *P. Gingivalis* RgpA-Kgp-HagA virulence complex may be held together by a novel structural mechanism which would facilitate presentation of protease and adhesin proteins to host tissue.

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Invited Lecture

IL005

Effect of *Salmonella* Specific Immunoglobulin Y extracted from Immunized *Lohmann Hens* and Probiotic bacteria against *Salmonella* Infection

Dr. Naheed Mojgani*[1], Amir Karimi Torshizi[2], Mehdi Babae[1] and Amir Karamzadeh Dehaghani[3]

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[2]*Department of Poultry Science, Faculty of Agriculture, Tarbiat Modaress University, Tehran*

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Salmonellosis is an important zoonotic disease caused by bacteria of the genus Salmonella that have a great economic interest worldwide especially in the poultry sector. Immunoglobulin Y (IgY) extracted from egg yolk is considered an ideal strategy for the diagnosis and therapy of Salmonellosis and can be used instead of polyclonal antibodies extracted from mammals (IgG). These antibodies are found to have better properties in terms of specificity and ease of large-scale production and as a low cost alternative that can be used in passive immunity in humans and animals. The objective of this study was to produce chicken IgY against *Salmonella enteriditis* and *S. typhimurium;* formulate themwith Probiotic bacteria *(L.plantarum, Enterococcus faecium)* and evaluatetheir anti-salmonella effect. Inactivated pathogens were used for immunization of laying hens. The immunized eggs were collected and yolk IgY extracted by PEG6000, dialyzed, filtered and analyzed on SDS-PAGE. Approximate molecular weight determined by Lowry method. The specificity of Anti-*Salmonella* IgY in chicken serum and yolk IgY were qualitatively measured by Rapid Slide Agglutination test and ELISA method. The extracted IgY were combined with $1x10^9$CFU/ml of the mentioned probiotic bacteria and their anti-salmonella effect evaluated in challenged Balb/c mice model.

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Invited Lecture

IL006

The Problem of *Listeria* in Food Industry

Dr. Kamran Shoukat
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While there are numerous pathogens that affect the food manufacturing industry, one of the most dangerous food-borne pathogens that exists today is *Listeria* – a bacterium that when present in foods, yields no difference in taste, smell or appearance. *Listeria* is the cause of the illness listeriosis. Nearly everyone infected with listeriosis is hospitalized, while one in five people or 20 percent will die from the disease. *L. monocytogenes* is widespread in the environment and can contaminate a wide range of foods. It is most commonly associated with chilled ready-to-eat foods such as cooked sliced meats, smoked fish, cooked shellfish, soft mould-ripened cheeses, pate and pre-prepared sandwiches that do not require further cooking or reheating. To help lower the risk for an outbreak, it is important to know that Listeria is a bacterium that can also grow at refrigeration temperatures. This means that if *Listeria* migrates onto food, it may continue to grow even if the food is held at refrigeration temperatures during shipping, storage, and display, thus increasing the potential for illness. A listeriosis outbreak is devastating, not only to the individuals that became ill but to the food processor that made that food, if the illness can be traced to their facility. Depending on the size of a facility and the size of the outbreak, a food processing plant could be forced to throw out all of their products that may be contaminated with *Listeria*. The processor may also have to issue a recall of contaminated or potentially contaminated food. The processor will also have to take steps to ensure that the *Listeria* is eliminated from the processing environment. This may require the processing facility to be shut down for hours or days while the facility is cleaned and processing equipment is disassembled as needed and deep cleaned. Plant closings can cost thousands to millions of dollars in lost time, decreased productivity and reputation damage. In addition to those losses there may be fines and litigation costs that can be even larger. Improved control measures starting in the 1990s have greatly reduced the prevalence *of L. monocytogenes* in many food categories, particularly in meats and meat products. However, the rate of Listeriosis has remained constant during the last decade and the more severe, systematic (invasive) form of listeriosis is now recognized as occurring more frequently in small outbreaks than previously recognized. In my lecture I will try to cover the *Listeria* issue in food industry, regulation to control *Listeria*, most recent *Listeria* outbreaks, the ways to control/spread of *Listeria* in food industry.

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Invited Lecture

IL007

Bacterial Production of Poly-3-hydroxyalkanoates using Sustainable Raw Materials for Biodegradable Plastics

Salaam Temitope O.* [1,2], Jamil, Nazia[2] and Lawal, Adekunle K. [1]
[1] *Biotechnology Department, Federal Institute of Industrial Research, Oshodi, Lagos, Nigeria.*
[2] *Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore, Pakistan.*

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Poly-3-hydroxyalkanoates (PHAs) are bio-polymers accumulated by several bacteria under unbalanced growth conditions. Their physical and chemical properties are analogous with synthetic polymers and are thus candidate substitutes in plastic production. This work report findings from the optimization of sustainable carbon sources using four novel bacteria strains in PHAs production. Poly-3-hydroxyalkanoates producing bacterial (PPB) were isolated from soil collected from cassava dumpsite and sugarcane farm. The organisms were screened for PHAs production in a carbon rich medium using the viable colony (0.5 µg/ml of Nile Red and Nile BlueA) and Sudan Black B (0.3 %) staining methods. Six selected PPB were identified by 16SrRNA sequencing. PHAs were extracted from lyophilized bacterial biomass by sodium hypochlorite/chloroform method. Extracted PHAs were analyzed by FT-IR for the detection of functional groups. The PHA synthase genes, *PhaC* & *PhaR* of the organisms were also partially amplified and sequenced. The organisms were identified as *Bacillus aryabhattai* C48, *Bacillus megaterium* SF4, *Bacillus cereus* C113, *Acinetobacter oleivorans SD12, Acinetobacter pitti*SF6 and *Enterobacter cloacae* C312. They produced orange, blue, and yellow fluorescence with Nile Red and Nile Blue A for all carbon sources used indicating the presence of PHAs. Blue black intracellular bodies of PHAs were also detected with Sudan Black B. Growth curves revealed greatest biomass accumulation in 2% starch medium for *B. aryabhattai* C48, *B. megaterium* SF4 and *B. cereus* C113 while *A. oleivorans SD12* showed greatest biomass accumulation in 2% sugarcane molasses medium. *B. megaterium* SF4 achieved 26.5% PHA production in starch at 24 hours while *B. cereus* C113 produced 33% PHA in glycerol at 24 hours. *B. aryabhattai* C48 and *A. oleivorans SD12* achieved 10% PHA in sugarcane molasses at 48 and 24 hours respectively. *A. oleivorans SD12*alsoachieved 18.5% PHA production in glucose at 48 hours. FT-IR spectra showed peaks indicating the presence of P3HB & P3HB3HV polymers. The detection of peaks at points ranging from 1721-1723 cm^{-1} & 1500 - 800 cm^{-1} further reveal conformational changes of mcl-PHA & scl-mcl PHA in crystalline and amorphous phases. Nucleotide sequences of 16SrRNA, *PhaC* and *PhaR* have been deposited in the NCBI GenBank repository. The results show that the four newly isolated bacteria have exciting potential for sustainable PHAs production.

MMG 2018
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Invited Lecture

IL008

Osteogenic Potential of Herb, *Cissus quadrangularis*: Effect of Organic Solvent Extracts on Differentiation of Mouse pre-Osteoblasts Cell Line MC3T3-E1

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Cissus quadrangularis is one of the medicinally important herbs which is reported in Ayurvedic literature as well as scientific literature for its possible role in management of osteoporosis and healing of fractures. This multi-step study examines the effectiveness of *Cissus quadrangularis* in promoting osteoblast differentiation of murine pre-osteoblast cell line, MC3T3-E1. The ethanolic extract of *Cissus quadrangularis* (CQ-E) was used to determine its effect on growth parameters of cells. CQ-E affected the growth kinetics of cells in dose dependent manner; lower concentrations being non-toxic. CQ-E treatment did not have detrimental effect on metabolic activity however, mitogenic effect of CQ-E at 0.1 and 1µg/ml was observed on proliferation of MC3T3-E1 cells. Osteogenic effect of CQ-E was analyzed by differentiating MC3T3-E1 in mineralization medium supplemented with different concentrations of CQ-E for three weeks. At 0.1 and 1µg/ml the extracellular matrix mineralized more heavily as manifested by bone specific staining. These concentrations augmented alkaline phosphatase activity of differentiating MC3T3-E1. Expression of early phase osteoblast related genes *RUNX-2* and *COL1A1*coincided with findings of histochemical staining. Their expression was upregulated in cultures treated with 0.1 and 1µg/ml CQ-E. During later stages of differentiation, the transcript level of *BGLAP-2* was up-regulated in the presence of CQ-E, compared to the positive control. None of CQ-E treatments affected expression of *IBSP* gene. This study was further extended on further purified fractions of CQ-E *i.e.* n-Hexane (CQ n-Hex), Dichloromethane (CQ-DCM), Ethyl Acetate (CQ-EthAc) and n-Butanol (CQ-n But). MC3T3-E1 cells were grown in various concentrations (0.01 - 200µg/ml) of each of these fractions and their effect determined on growth parameters of the cells. At 1 and 10µg/ml of CQ-EthAc and CQ-nBut fractions, significant increase in number of cells was observed as compared to control and other concentrations tested. All concentrations of these two fractions showed no detrimental effect on metabolic activity of the cells. The fractions CQ-n Hex and CQ- DCM affected growth of cells in dose dependent manner, the lower concentrations being less toxic. Same results were obtained for metabolic activity of the cells. Further lower concentrations (0.005 – 0.0001µg/ml) are still to be tested to find out suitable non-toxic concentrations for the cells. Osteogenic effect of non-toxic concentrations of all the four fractions will be studied by differentiating MC3T3-E1 cells in mineralization medium. Presentlyosteogenic effect of CQ-EthAc at 1µg/ml is being investigated. Expression of early and late phase osteoblast related genes will also be checked. Taken together the findings of the study so far, *Cissus quadrangularis* can be a potential

medicine to treat osteoporosis and bone related problems. Moreover, long term plan also includes investigating the effect of CQ on osteoblast differentiation at molecular level, its role in lineage commitment of osteoblast differentiation of mesenchymal stem cells, as well as its role in osteoclastogenesis and adipogenesis.

MMG 2018
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Invited Lecture

IL009

Microbes: Foe or Friends

(Carlos J. Finlay UNESCO Prize for Microbiology Awardee-Lecture)

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Microorganisms exist everywhere, all around us as well as within us. They have major useful roles to play such as in environmental biogeochemical cycles and in digestion within our body, etc. Sometimes, some microbes can also turn against us causing lots of problems manifested as infections of various types. This lecture will highlight the significant roles of microbes based on research of my microbiology career.

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Invited Lecture

IL009a

Metagenomics of hypersaline environment and microbiome of halophytes as a source of osmoregulatory genes

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The distribution of saline soils on more than half a billion hectare worldwide, warrants efficient, economical and environmentally acceptable management practices. In addition, scarcity of water compounded by climate change resulting in long drought spells has warranted development of crops with tolerance to both salinity and aridity. In this context several efforts are being made to understand osmoregulation in living systems. In this regard, studies on halophytic plants and halophilic bacteria can provide useful information. More recently it has been shown that plant bodies harbor a large number of microbes which are more than the number of plant cells. Similar findings have been made for human body. Plant microbiome has thus been reported to contribute significantly to plant performance and can provide information regarding complex ecological processes involved in osmoregulation of halophytes. Metagenomic based molecular retrieval of 16S rRNA gene (rDNA) sequences have become the most important tools for exploration of microbial diversity. The basic aim of this study is to investigate the microbiomes associated with aboveground (phyllosphere), below-ground (rhizosphere) and internal (endosphere) tissues of halophytes. Culturable bacteria were characterized morphologically, physiologically, biochemically and identified by PCR amplification of specific 16S rRNA gene sequences. Sixty two strains were selected after screening of salt tolerance. It has been earlier reported that some of the osmoregulatory genes are present on the plasmid. In order to demonstrate it, plasmid curing of isolates was done by heat shock method, using SDS (3%) and sodium benzoate ($130gl^{-1}$) to study the effect of plasmid conferring salt tolerance. These plasmids were isolated and transformed into *E. coli* and growth response of original strains and transformed *E. coli* was found to develop high salt tolerance at 2-4M NaCl concentration. Thus indicating some of the osmoregulatory genes to be located on the plasmid. The sequencing of the plasmid is underway which will reveal the array of genes responsible. These genes can then be used with appropriate promoters to transform some of the economic crops.

MMG 2018
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Invited Lecture

IL010

Eco-Friendly Biological Agents; Current and Future Perspective of Food Security

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As the human population is increasing globally and it is becoming difficult to meet the needs of human being. Hence, microorganisms play significantly a supplementary role in meeting food requirements and combating with various diseases. To exploit the beneficial role of microbes, bacterial strains were isolated from rhizosphere and endosphere of various field crops and selected on the basis of best N-fixation, P-solubilization, Silicate solubilization, Zn solubilization, Indole acetic acid production, and *in vivo* bio-control activity under field conditions. These effective PGPR belonged to different genera such as *Acetobacter, Pseudomonas, Azospirillum, Rhizobium, Azotobacter, Bacillus, Enterobacter, Aeromonas, Burkholderia* and *Serratia* as identified by a number of molecular markers such as *16S rRNA, 16-23S rRNA, gyrase A, B, acdS, nif* and *PqqC* genes. Bio-control activity of the antagonistic strains was found to be associated with the production of siderophores, antibiotics such as, surfactin, pyoluteorin and hydrolytic enzymes like protease, glucanase and chitinase. For preservation of food, bacteriocin "Nisin" produced by lactic acid bacteria and a commercial variant was embedded in monolaurin nanoparticles (MNPs) which showed greater stability with loaded nanoparticles. The FTIR analysis had confirmed that the encapsulation of nisin into MNPs is based on electrostatic attraction only. Nano based bio-formulation was developed by coating the seeds with bio-functionalized electrospunnanofibers which presented an excellent alternative in order to protect bio-agent from dehydration, direct effects of toxic chemicals present in the seed tegument and have itself role in seedling emergence. This novel coating and encapsulation system played a realistic role in sustainable crop production and environmental promotion. A great efficacy was observed in the probiotic cells encapsulated in electrospunnanofibers which showed slow releasing fight against pathogens and aided in healing the inflammatory mice wounds. In addition, bio-fortification of grain crop is an important feature of PGPR based technologies. The potent strains were preserved at Pakistan Collection of Microbial Cells (PCMC). Some efficient strains have been commercialized as Humiphos™ and Biophos™ under Academia-Industry linkage. Improvement in the developed phosphatic bio-inoculant as well as development of new Zn based bio-fertilization is in progress.

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Invited Lecture

IL011

Industrial Enzymes Diversity from Single Bio-factory

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There are many industrial setups that use microbial enzymes for particular applications: in the textile industry enzymes are able to improve the quality of the fabrics; in the pulp and paper industry they are involved in biomechanical pulping and bleaching; in the food industry they are used in the fermentation processes for the production of bread and drinks such as wine and beer, also they participate in the extraction of substances, such as carotenoids and olive oil; in the detergent industry they have superior cleaning properties, increasing the brightness and removing dirt; they are also used in cosmetics, animal feed, and agricultural industries, among others. Single specie able to produce multiple enzymes is of special interest, as multiple enzymes can be harvest from the same unit. Extreme environments merit special attention and significance because of the possible existence of thermophilic microorganisms in such ecological niches. Keeping this in mind indigenous stove ash samples were explored for extremophilic bacteria in term of their biodiversity. A *Bacillus* strain was isolated from stove ash sample. The isolate was found able to grow up to 70°C. Isolate was then tested for its ability to produce industrially important hydrolytic enzymes including cellulase, lipase, esterase, xylanase, pectinase, amylase, dextranase, keratinase, caseinase and β-galactosidase by plate screening methods. Results showed that the isolate was able to produce all the tested enzymes except pectinase. The isolate was then identified up to specie level by 16S rDNA sequencing as *B. licheniformis.* This is the pilot study setup the platform for simultaneous production of different industrial enzyme by single species. Further study will be carried out for optimizing different fermentation conditions for highest co production yield.

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Invited Lecture

IL012

Antimicrobial Activities of *Trichoderma Harzianum* Against the Plant Pathogens

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*Trichoderma harzianum*is a fungus and used as an effective bio-control agent for various soil born and plant diseases caused by variety of fungi. It is commonly found in the rhizosphere and highly interactive in root. It has been successfully used in field trials in controlling different crop disease causing pathogens, where its use is as a bio-control agent for eradication of soil borne diseases. It has diverse potential of being used as bio-control and performs mycoparasitism, competition, hyphal interactions, enzyme secretion, and antibiosis which arrest the growth. *Trichoderma* have been reported to suppress the growth of the pathogenic microorganisms associated with the roots of plants in the rhizosphere resulting in eradication of plant diseases. Various different antimicrobial and tonic chemicals such as trichothecin and a sesquiterpene, Trichodermin, are produced by *Trichoderma* that have antagonistic effects on other microorganisms dwelling in roots and causing plant infections. In order to exhibit the antagonistic effects, *Trichoderma* hyphae grows itself and coil it around the host hyphae followed by secretion of various lytic enzymes including pectinase glucanase and chitinase. These enzymes actually enable the *Trichoderma* to start the mycoparasitism process. *T. harzianum* utilizes the same process of interaction against a wide range of hosts such as *F. roseum*, *Fusarium oxyporum*, *Phytophtharacolocaciae*, *Sclerotium rolfsii* and *F. solani*. *Trichoderma* besides this interaction may also enhance yield, boost germination rate and the quality of production. Increase in shoot & Root length. It also has various other beneficial characteristics as they fix nitrogen and solubilize various insoluble forms of phosphates. Keeping in view the all good qualities of a bio-controlling agent, *Trichoderma harzanium* would be a promising bio-control agent in elimination of plant diseases and enhance the agricultural output of the country. We have observed the different activities of *Trichoderma harzianum* such as (i) Influence of *Trichoderma harzianum* and Azotobacter on decomposition of organic substances of straw and flex of Wheat. (ii) Effect of *Trichoderma harzianum* and different doses of fertilizer on nitrogen fixation. (iii) Optimization of antagonistic effect of *Trichoderma harzianum* etc. In conclusion, *Trichoderma harzianum* possess the antagonistic activities and it can be used as bio-control agent against wide range of plant diseases.

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Invited Lecture

IL012a

Forlorn Calls to Pakistan for Composting – A Thus Far Unachievable Ideal

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The organismic materials, generally called organic biodegradables (OBs), were introduced first on earth or its degradation is a circular reference just like, what came first, the chicken or the egg. However, there have been time-based disorientations induced by human into the cyclical movement of OBs-based organic carbon (OC) into and from the soil; mainly in the form of land use evolution history. The so-called development-driven land use patterns have ever modified the forest and agricultural fronts of the world. Such grievous modifications have been very intense in Pakistan just like other parts of the developing world. The managed decomposition of OBs is called composting i.e. bringing the disoriented cyclical movement of OC into and from the soil back into regular cycle. The OC given off from the soil is distributed on earth surface in the form of woody and non-woody biomass; crop residues; weed biomass; processable wastes like cereal husks, bagasse, nut shells, sawmill residues, black liquor from paper mills, ≥ 70% OBs in the municipal solid waste (MSW); the silt of fresh water bodies like canals, biosolids from municipal sewerage, etc. Channeling all the given off OC from soil back into it could best be achieved through composting only. Here, the emphases are very much on managing crops residues and OBs of MSW through composting. The biggest drivers behind composting as need of the hour are described. Looming threats of recurrent seasonal smog due to mass scale open burning of crop residue and economically unviable MSW management of Lahore as a model city for six major cities of the Punjab can prove a further potential replicative approach in rest of the provinces of Pakistan. The emerging federal efforts to convert >75 % virgin lands of Baluchistan (43 % area of Pakistan) into cultivable soils creates big local market for consumption of compost beside its dire need in Azad Jammu Kashmir, Gilgit Baltistan, Khyber Pakhtunkhwa, Sindh and Punjab. Composting is an essential factor of decentralized MSW management if not the only option in all the provinces of Pakistan owing to the below average gross calorific value of MSW. The causes of failures of the past large-scale composting endeavors in Pakistan and their workable shortcomings have also been considered. Most of all, it is envisaged that through composting, controlling climate change lies under the feet of man.

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Invited Lecture

IL013

Immobilization of Uranium by Biofilm Forming Strains of *Bacillus* strains

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Bacterial biofilms has a ability to convert heavy metals less toxic elemental metals. *Bacillus cereus* SS5, *Bacillus subtilis* SN5 and *Bacillus pumilus* FR2 isolates from SDB, DDB and FWB respectively, showed prominent EPS production, were selected for further experiments involving *in vitro* biofilms formation potential and metal resistance studies. The three strains SS5, SN5 and FR2 were grown in flat plate Open Channel Reactor on glass slides to assess biofilm formation by these strains, the biofilms were then imaged through CLSM and image structure was analysed with the help of ISA-2 software. All the strains expressed optimum biofilm formation potential. These strains were then monitored for their growth response under different uranium concentrations. SS5 showed prominent growth in this experiment, hence was selected for further uranium immobilization experiments through biofilm formation. *Shewanella oneidensis* MR1, an isolate from the Hanford Reach of Columbia River, served as a model organism in these experiments. Biofilms were grown in Flat Plate Open Channel Reactor fed with Uranium at a concentration of 126 µM per litre. Uranium measurements were accomplished through Kinetic Phosphorescence Analyser (KPA). Biofilm imaging involved light microscopy, Confocal Laser Scanning Microscope (CLSM) and Scanning Electron Microscope (SEM). LIVE/DEAD staining kit was used for the assessment of viability in the biofilms. Biofilm Immobilized uranium was also observed through florescence. EPS abundance in such biofilms was confirmed through a succinyl conjugated Lectin Alexa Fluor 488, glycoprotein binding green florescent dye. Over all study in this work revealed that biofilm inhabiting bacteria are fenced with EPS that impart unique characteristics to these bacteria not only to cope with harsh environment but also to immobilize toxic metals. *Bacillus cereus* SS5 promising uranium immobilization through biofilms and associated EPS. *Bacillus cereus* SS5 biofilms and EPS showed encouraging uranium immobilization potential like the indigenous bacterium *Shewanella oneidensis* MR1.

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Invited Lecture

IL014

CRISPR-CAS System: Discovery, Mechanism and Significance

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Viruses are acellular microorganisms that not only pose threat to humans but also to prokaryotes. Bacteria have certain protective measures to combat the external invasion, such as adaptive immune system called (clustered regularly interspaced palindromic repeats) CRISPR- Cas system. In this defence system bacteria incorporate the invading genetic sequence called spacer into the CRISPR locus via spacer acquisition upon second attack bacteria recognizes the invading sequence and degrades it by various Cas proteins. Three main CRISPR systems have been identified by researchers. This paper summarizes various important discoveries regarding this natural immune system of bacteria, how it works also discusses its function other than immunity and also focus on significance and applications of the system as a DNA editing technology and antimicrobial agent.

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Invited Lecture

IL015

FECPAK; World Leading Online Parasite Diagnostic System, The future In Parasite Management

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The Parasitology Section of the College of Veterinary & Animal Sciences, Jhang is a full-service parasite diagnostic laboratory capable of diagnosing infections in domestic and wild animals. This service is provided in an accurate, timely and cost efficient manner. Accurate identification of parasites benefits animals, their owners and veterinarians by helping to improve animal health. Services provided by the Parasitology Section include: Detection and identification of domestic and wild animal parasites from animal feces, animal tissues, fluids and environmental samples. Tests conducted to identify parasites in feces include: Qualitative and quantitative centrifugation concentration flotation techniques, Nematode larvae cultures, Direct fecal smears (wet mounts), Baermann technique for recovery of nematode larvae. FECPAK is a complete remote-location parasite assessment tool. FECPAK generates results quickly and easily for the farmer, manager, vet or adviser so they can make informed parasite management decisions without delays. The Programme is based on the THE FUTURE IN PARASITE MANAGEMENT; that leads to Simple Fecal Egg Count (FEC) processing on-farm – no microscope required, Prompt FEC results via email, Online access to all your FEC information, No special technical skills or knowledge needed, Easy to use, fully auditable, centralized FEC information reporting and sharing options, Animal health alerts, Complete technical product backup and client support. The big question **Why Use FECPAKG2 To Monitor Parasites?** The answer is Increase financial returns, Increase animal performance while decreasing workloads and drench costs. Drench resistance, Reduce the risk of drench resistance by knowing which drenches work. Monitor performance and minimize drench use by only treating animals that require it. Empowered management, Gain valuable information to enable a best practice approach to parasite management. Meeting consumer needs, Reduce chemical usage to meet customer demands in the meat industry.

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Invited Lecture

IL016

From Technology to Treatment –Nano Structures at Work

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From memory chips to drug delivery, energy sources to agriculture, sensors to water treatment, world's largest issues can be addressed using tiny structures at nano-scale. Naturally available and synthesized nanomaterials all exhibit unique chemical and physical properties because of large surface/volume ratio. Nanoscience gives the advantage of dealing electrons as two different charge carriers by considering nature of their spins i.e. spin up and spin down. Mott was the first scientist who gave this idea in mid-thirties. This idea came up with the advantage that spin flip scattering is dimension dependent and is rare on time scale. JMD Coey, in 1987, postulated as "Conventional electronics has ignored the concept of spin of electrons". Such behavior arises because of the difference in mobility of two types of electrons i.e. spin up and spin down. Above mentioned concept is being utilized through the use of nanomaterials. Two types of approaches for nanotechnology including top-down approach and bottom up approach. Bottom up approach or self-assembly includes sol-gel synthesis, autocombustion, co-precipitation, electrodeposition, green synthesis, bio-synthesis, etc. Nanoparticles synthesized using green approach have advantages over the physical and chemical synthesis procedures, as this is cheap, eco-friendly, convenient single-step method, that can be easily scaled. Moreover, it does not require high vacuum, temperature, toxic gases and chemicals. Environmental friendly and green processes for the synthesis of metal and metal oxide nanoparticles are growing into an important branch of nanotechnology. Furthermore, biosynthesis uses the advantage of micro-organisms, plants and other naturally available species for the synthesis of nanomaterials. We, at the COE in Solid State Physics started MS Nanotechnology in 2010 in order to implement the ideas developed during the over 35 years' experience of materials preparation and characterization in the solid form. We, now, have well established chemical synthesis (conventional-, green-, bio-) labs and characterization facilities to move towards implementing nano ideas for high-tech applications in various fields. Over the years we have optimized the preparation conditions, through characterizations, of carbon, iron oxide, zirconium oxide, titanium oxide, zinc oxide and various metals in the nanostructure form. These nanostructures can be tailored with required properties for different applications, for example iron oxide can be synthesized in the ferromagnetic, paramagnetic or superparamagnetic forms. Superparamagnetic is useful for hyperthermia treatment, and for antifungal activities in various crops. Whereas, Zinc oxide nanostructures can be prepared to be used as dilute magnetic semiconductors or large band gap transparent conducting oxide. We have recently demonstrated that zinc oxide nanoparticles can be used to destroy cancer cells using unharmful rays of photons. Zirconia nanoparticles can be optimized for use as tough ceramic coatings or as protective teeth coatings to slow down the decay processes.

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IL017

Radioprotective Effect of Amifostine on Ccells from Cancer Prone Patients and Healthy Individuals Studied by the G2 and PCC & Cytogenetics G2 Assays for Cancer Diagnosis at High Cytotoxic Level

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To investigate whether amifostine is effective at reducing the yield of chromatid breaks when present during G (2)-phase irradiation of human normal cells and cells from cancer prone patients, as well as to study the mechanisms underlying the radioprotective effect of amifostine. G (2) chromosomal radio sensitivity in the presence or absence of amifostine was studied in healthy donors, cancer patients, ataxia-telangietasia (A-T) patients and five human lymphoblastoid cell lines with genes predisposing to cancer. The yield of chromatid breaks following gamma-irradiation in G (2) phase was obtained at the subsequent metaphase using the G (2) assay. For scoring chromatid damage directly in G (2) or G (0) phase, premature chromosome condensation was used. When amifostine was present during irradiation, the mean yield of radiation-induced chromatid breaks as visualized by the G (2) assay was significantly reduced in healthy donors (t-test, $p=0.001$), in cells from cancer patients ($p=0.001$) and in cell lines from patients with genes predisposing to cancer ($p=0.01$) except ATM (-/-) ($0.1<p<0.2$). However, when chromatid breaks were scored directly in G (2) or G (0) phase using premature chromosome condensation, the presence of amifostine did not affect the yields obtained. Amifostine reduces the mean yield of chromatid breaks in normal cells and in cells from cancer prone patients when present during G (2) irradiation. Although the precise mechanisms of radioprotection caused by amifostine remain unclear, the results obtained using premature chromosome condensation reveal that amifostine does not act on cells only as a free radical scavenger and as a repair enhancer of DNA damage.

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IL018

Salmonella typhi: Prevalence and Drug Resistance Signature

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Typhoid is a common infection in Pakistan caused by *Salmonella typhii*. With the passage of time *Salmonella typhii* develops resistance to antibiotics used to treat typhoid. A total of 2138 blood samples from suspected typhoid patients were collected from North Pakistan region. Blood samples were cultured on bismuth sulphite selective agar and were then identified by biochemical (API) and serological tests. The isolates were further confirmed using *Salmonella typhii* species specific *fliC* gene amplification using PCR. The antibiotic susceptibility of all the isolates was analysed using disc diffusion method. Among the 2138 blood samples, 206 were positive for *Salmonella typhii.* All the *Salmonella typhii* isolates were confirmed by fliC gene amplification. The antibiogram profile of the *Salmonella typhii* isolates exhibited resistance to ampicillin, chloramphenicol and cotrimoxazole. While some of the isolates were susceptible to ceftriaxone. Surprisingly 11 isolates exhibited resistance to all the tested drugs. Findings of the current study showed alarming situation of antibiotic resistance among the *Salmonella typhii* clinical isolates.

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Invited Lecture

IL019

Proteomic Analysis Reveals Novel Binding Partners of SSAO in Human Endothelial Cell

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The existence of semicarbazide-sensitive amine oxidase (SSAO) has been implicated for a long time with diabetes, atherosclerosis, inflammatory conditions and others. However, the molecular mechanism that leads its biological functions is yet little defined. In order to develop a greater understanding of molecular mechanism we employed a proteomic approach to identify SSAO interacting candidates. Firstly, human SSAO was expressed in human umbilical arterial endothelial cell (HUAEC) and then it was pulled down together with its interacting partner using monoclonal anti-SSAO antibody. Mass spectrometry was performed on co-precipitation elutes. Proteomic analysis revealed 35 tentatively interacting proteins. The existing interactions of PTPRC, CCL20 and ALDH1A3 were confirmed with western blot of reverse co-immunoprecipitation elutes and their co-localization was observed by fluorescence confocal laser scanning microscopy. The work described here demonstrated first time that PTPRC, CCL20 and ALDH1A3 interact with SSAO and highlighted the preliminary study on the molecular mechanism; that explain its possible involvement in leukocyte trafficking, inflammation, insulin resistance, chemotaxis and others. Additional interacting partners also indicated a broad spectrum of SSAO activities. This study is an important step towards identifying interacting partners of SSAO that could provide a new insight into mechanism for the evaluation of its functional diversity.

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Invited Lecture

IL020

Drug Resistance in *Mycobacterium tuberculosis* Isolates from Pakistan: Current knowledge and Future Perspectives

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Rapid emergences of multi drug resistant *Mycobacterium tuberculosis* (MDR-MTB) are the growing challenge to global TB control. Unfortunately, Pakistan has been categorized 5thamong high TB burden countries. The prevalence of tuberculosis has been recorded as 342 per 100,000 population. MDR-MTB strains exhibit resistance to the 1st line anti-TB drugs such as rifampicin (RIF) and isoniazid (INH). MDR-TB management needs prolong therapeutic strategy, and pose adverse effects on patient health. RIF resistance has been widely used as screening tool for MDR-TB. Gene Xpert assay has been employed for the detection of the RIF resistance-determining region (RRDR) of the *rpoB* gene. High prevalence of rpoB gene in MTB complex has been reported from Pakistan. Furthermore, mutational analysis showed several mutations in rpoB gene thus making MTB more drug resistant. Isoniazid resistance has been associated with mutation in the *kat*G and *inh*A S94A. Significant number of MTB exhibit extensively drug resistance (XDR) pattern as these isolates showed additional drug resistance to fluoroquinolone and amikacin, and kanamycin. Various efflux pump genes (*Rv2688 and drrB)* have been reported in MTB which also play a vital role in the development of drug resistance. Several factors contribute to high burden of MDR-TB in Pakistan such as absence of effective TB control strategies, lack of accurate diagnosis, lack of awareness, overpopulation and poverty. In addition, drug resistance and virulence is also on the rise. So in depth investigations are needed to explore mycobacterial pathophysiological processes for effective TB control in Pakistan.

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Invited Lecture

IL021

Endophytic Fungi *Fusarium culmorum* Pz11 Senses Flavonoids and IAA Signals to Locate and Colonize Maize Root

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Molecular crosstalk between microbes and their host is the first step toward the establishment of any association. Plant roots release a variety of signal molecules to differentially treat the beneficial and harmful microbes. Current study focuses on the role of flavonoids and IAA as signals for molecular dialogue between endophytic fungus and maize root. Endophytic fungi *Fusarium culmorum* Pz11 was isolated from the roots of drought stressed *Asphodelustenui folius* (wild onion). Identity of the strain was confirmed by homology of the ITS region of 18 S rDNA sequence. The strain was able to produce a number of phyto-stimulants and signaling compounds including indole-3-acetic acid (IAA), flavonoids and sugars. Its culture filtrate contained 33.2±0.8, 275.1±8.7 and 186.6±15.7 µg/mL of IAA, total flavonoids and sugars respectively. The strain effectively colonized the roots of maize and subsequently enhanced growth of its host. To determine the effect of flavonoids and IAA on the ability of the endophyte to colonize maize roots, we inhibited the release of flavonoids and IAA individually which effectively reduced colonization of the endophyte in maize root to 89% of the control. Similarly, colonization of root by endophyte with repressed flavonoids was reduced to 62% of the control suggesting a flavonoids talk between the two partners. Suppression of IAA biosynthesis in the endophyte drastically affected its colonization in the maize root. It is concluded that a molecular crosstalk of maize roots and endophytic *Fusarium culmorum* sp. Pz11 is necessary for subsequent endophytic association between them.

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Invited Lecture

IL022

Synthesis and Exploration of New Antimicrobial Agents: Combined Research from Synthetic and Natural Origins

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Infectious diseases, one of the leading health problems accounting for 41% of disease burden globally. Infections caused by bacteria and parasites are still challenging to public health. Such kinds of diseases are most common in developing countries due to unavailability of relevant medicines or the emergence of drug resistance. The efficiency of many available antibiotics has been endangered by the emergence of multi drugs resistance pathogens. To combat with MDR pathogens, it is important to explore novel and effective antimicrobial agents. We have explored various antimicrobial agents using both synthetic and natural products origins. Using the synthetic approach, we have synthesized various derivatives of succinimides via asymmetric Michael additions using non-covalent bifunctional catalysis approach. We have added various aldehydes, ketones, ketoesters and cyanoacetates to maleimides to produce a diverse array of the respective succinimides. Among all the compounds, we observed that succinimides with ketoester functionality were effective against various strains like *E. sakazakii*, *E. coli*, *S. aureus* and *K. pneumonia*. The in-vitro results were also supported with in-silico studies. High GOLD fitness score data from docking analysis towards the targets represent better protein–ligand binding affinity and thus indicate a high propensity for all the active compounds to bind to the active site. Among the natural products, so far we have explored the antimicrobial potentials of *Notholirion thomsonianum*, *Allium consanguineum*, *Eryngium caeruleum*, *Isodon rugosus*, *Polygonum hydropiper* and *Teucrium stocksianum* against various bacterial and fungal strains.

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Invited Lecture

IL023

Potential Role of microRNAs and Their Target Genes in Metabolic Diseases and Oral Cancer

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MicroRNAs (miRNAs) are short non-coding RNAs, which regulate all major cellular processes by either enhancing or inhibiting target gene expression. In addition, miRNAs play important role in the pathogenesis of various metabolic diseases including obesity, diabetes, chronic kidney diseases (CKD) and cancers. The incidence of metabolic diseases is increasing at an alarming rate worldwide to a highly epidemic level affecting millions of individuals around the globe. We are conducting multiple studies to analyze the role of miRNAs and their target genes in course of pathogenesis of CKD, diabetes, obesity and oral cancer. Briefly, we have generated a list of miRNAs, having a role in each metabolic disease and oral cancer through extensive literature search and few gene expression experiments of each disease were extracted from NCBI database. The list of miRNA target genes and differentially expressed genes (DEGs) from experiments were compared for common gene IDs. Functional enrichment analysis was performed using DAVID, an online functional annotation tool. Networks of genes were generated using Cytoscape (3.2.1). Integrated analyses depicted that miRNAs modulate renal development, homeostasis, various metabolic processes, immune responses and ion transport activities. Furthermore, homology studies of miRNA-mRNA hybrid highlighted the effect of partial complementary binding pattern on regulation of genes by miRNAs. For expression of miRNAs and their target genes in Pakistani population, RT-PCR was used. Expression profilings of miRNA and their target genes were performed by extracting total RNA from whole blood samples of obese, diabetic, CKD patients, saliva samples of oral cancer patients and compared with control samples. Descriptive analysis of each data showed that there was increased prevalence of metabolic diseases and oral cancer among individuals with increase in age. Our findings revealed a significant change in expression of miRNAs and their target genes in patients as compared to controls. Relative expression of target genes was positively correlated with each disease pathogenesis. These results suggest that miRNAs and their target genes may have great value as biomarkers in metabolic diseases and oral cancer diagnosis at early stages of the disease.

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Invited Lecture

IL024

Biochemical and Clinical Outcomes as Potential Indicators of Preeclampsia

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Preeclampsiais a pregnancy specific disorder that complicates 7% of all pregnancies. It is characterized by high blood pressure equal to or above 140/90 mmHg with manifestation of proteinuria after 20 weeks of pregnancy. It is one of the leading causes of maternal and fetal morbidity and mortality and currently there is no treatment other than cessation of pregnancy. Present study was, therefore conducted to detect the women at high risk of developing preeclampsia by assessing the role of biochemical parameters in the development of preeclampsia and to identify early pregnancy plasma/serum markers in women destined to develop preeclampsia. For this purpose, pregnant women were approached at different hospitals of Lahore and private/public obstetric clinics. Subjects for blood sampling were categorized into three groups on the basis of blood pressure and proteinuria. Control group: Healthy control pregnant women at 13-20 gestational weeks with normal blood pressure≤120/80 mmHg and without proteinuria. Risk group I: Pregnant women at 13-20 gestational weeks having blood pressure of ≥ 130/86 mmHg. Risk group II: Pregnant women at 13-20 gestational weeks having blood pressure of ≥ 130/86 mmHg and proteinuria~200mg/l. Women with family history of hypertension, preeclampsia and/or previous history of preeclamptic pregnancy were identified. Venous blood 6cc was withdrawn, after an overnight fasting, from all participants. Serum and plasma was separated. Serum samples were analyzed for Liver function (Alanine aminotransferase or glutamate pyruvate transaminase, Aspartate aminotransferase or glutamate oxaloacetate transaminase, Albumin, Bilirubin, Total protein, Alkaline phosphate), lipid profile (Cholesterol, Low Density Lipoprotein, High Density Lipoprotein, Triglycerides, renal function (creatinine, urea, uric acid) and electrolytes (Na+, K,+ Ca+, Mg+) using commercially available diagnostic kits. The quantitative determination was made by using clinical chemistry analyzer. Inflammatory markers (high sensitive C-reactive protein and interleukin-6) were estimated through ELISA. All tests were performed at Physiology Laboratory, University of the Punjab, Lahore. In lipid profile, TC, TG, LDL-C, and VLDL increased, whereas HDL-C was decreased significantly in the subjects of both risk groups who subsequently developed preeclampsia. In liver profile, ALT and AST levels were significantly elevated during early 2nd trimester in the women who later developed preeclampsia after 20th week of pregnancy. Uric acid showed a significant increase before 20th week of pregnancy in the women who were susceptible to the development of preeclampsia. C-reactive

protein and interleukin-6 were significantly increased in preeclamptic women before 20th week of pregnancy when compared when normotensive pregnant women. It is therefore, concluded that the specific parameters of the study can be helpful in early identification of the subjects at risk of developing preeclampsia before the appearance of clinical symptoms and complications of preeclampsia suggesting that biochemical and clinical outcomes are potential indicators of preeclampsia.

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ORAL Presentations

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Oral

Environmental Microbiology **T001**

Use of Microorganisms: A Strategy to Clean Industrial Wastewater

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This study reports the bioremediation potential of bacteria and yeast to eradicate cadmium from the environment. Spectroscopic analysis clearly illustrated the involvement of bacterial cell wall components in biosorption. Cadmium bioaccumulation was confirmed by TEM, SEM, and EDX examination. Increased biosynthesis of GSH under cadmium stress indicates one of the strategies to cope with metal-stress. Proteins exhibited differential expression and during cellular redox homeostasis are found to involve in nitrogen metabolism, nucleotide biosynthesis, and carbohydrate catabolism. The Cd^{+2} toxicity not only caused growth stasis but also upregulated the cysteine biosynthesis, protein folding and cytoplasmic detoxification response elements in yeast. Yeast was also able to degrade azo dye present in the environment. This study suggests that microorganisms can be used as potential candidates for eradicating the toxic pollutants present in the environment.

Oral

Antimicrobial Agents and Chemotherapy **T002**

Screening of Desert Actinomycetes against Methicillin Resistant *Staphylococcus aureus* (MRSA)

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Actinomycetes have been, for decades, one of the most important sources for the discovery of new antibiotics and their analogs, successfully introduced in the market and still used today in clinical practice. In the present study we have succeeded in isolating 80 strains of actinomycetes from an untapped environment of Cholistan desert located in southern Punjab, Pakistan. The selected strains with potent antimicrobial activity against methicillin resistant *Staphylococcus aureus* (MRSA) were identified by morphological, biochemical and physiological characterization and by 16S rRNA gene sequencing. The antimicrobial activity of the selected actinomycetes isolates was determined by well diffusion and disc diffusion methods (maximum zone of inhibition up to 30mm). These MRSA were recovered from clinical samples and identified morphologically, biochemically and genetically. The isolates were validated as MRSA based on their resistance to standard antibiotics as per CLSI standards 2012 and by mec-A gene characterization. The actinomycetes strains were cultivated as 1 liters shaking flask cultures and subsequently the active compounds were extracted and were characterized by thin layer chromatography (TLC) and HPLC-UV/RI. Most of the isolates exhibited genetic similarity with the genus *Streptomyces* e.g. isolate AFD6 (Accession no. KX131166) showed 99 % similarity with Streptomyces thermolilacinus strain NBRC 14274. The most bioactive actinomycete strains were grown on large scale to get the sufficient amount of the active components which were then subjected to purification through silica gel sephadex columns. These pure components were analyzed through HR-MS and NMR spectroscopy. We can conclude from our results that deserts are very rich and abundant source of actinomycetes with unique and novel secondary metabolites that may play a helpful role to overcome the threatening situation of antibacterial resistance.

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Oral

Public Health **T003**

Association of Hygiene Practices with Frequency, Duration and Severity of Acute Diarrheoa among Children of Lahore, Pakistan

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Childhood diarrhea is a well- known public health issue. Diarrheal disease accounts for high child mortality rates, especially in the developing world. Personal and food hygiene has been studied as probable factors responsible for increasing the diarrheal burden. This study aimed at determining the knowledge and practices of respondents regarding food hygiene and its link to childhood diarrhea. For this study, a cross sectional (analytical) study design was adopted. Data was collected from caregivers of 322 under five children, suffering from acute diarrhea, who visited the OPD of a public sector hospital in Lahore. Data was collected using an interview guideThe knowledge about hygiene among the caregivers was found to be very low; only 19.9% mothers regarded the probable cause of childâ€™s diarrhoea as related to unhygienic factors. The caregivers practised unsafe handling of food; especially the feeding bottle hygiene was being inadequately maintained. The hygiene practices were significantly correlated with childhood diarrhoea. Negative correlation was found between food hygiene scores and diarrhoea frequency ($r = -0.088$, $p > 0.05$), duration ($r = -0.101$, $p > 0.05$) and severity ($r = -0.185$, $p = 0.000$). The regression analysis showed food hygiene as one of the most important independent determinants of childhood diarrhoea (OR=2.7, p=0.031). Childhood diarrhoea is one of the leading causes of under-five mortality and morbidity. The strong associations of food hygiene practices with diarrhoea and lack of knowledge regarding food safety, call for immediate response. Interventions for enhancing the knowledge and promoting hygienic practices can promise cost effective solution to high diarrhoeal rates for the developing world.

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Oral

Environmental Microbiology **T004**

Auxin-Producing Rhizobacteria as Bioremediating Agents to Minimize Chromium Toxicity in Chromium Contaminated Areas

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Heavy metals contamination in the environment is a serious threat causing health risks to the living organisms. Hence, it is very necessary to get rid of these chemical contaminations to have a safe environment. In this regard, bioremediation is considered to be an economic and sustainable agricultural techniques rather than use of other chemical methods to mitigate chromium toxicity from agricultural lands together with improvement in plant growth. Present work is focused on the use of plant growth promoting, chromium-resistant bacteria for enhancing the growth of *Zea mays L*. under stress conditions by using six efficient auxin-producing, chromium-resistant bacteria *Bacillus pumulis* (ALa), *Bacillus atrophaeus* (BL2), *Bacillus cereus* (AR), *Staphyllococcus lentus* (E3), T2aii and W6ii grown under various concentrations of chromium stress i.e., 0, 200, 400 and 600µg/ml by giving inoculation treatments to Zea mays L. seeds and different growth and biochemical parameters were recorded. Analysis of the experiment demonstrated that application of auxin-producing rhizobacterial inoculation treatment enhances the growth of plants prominently by alleviating chromium toxicity.

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Oral

Environmental Microbiology **T005**

Cr^{+6} Reducing Potential of *Staphylococcus sciuri* (A-HS1) Isolated from Industrial Wastewater

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A chromium-resistant bacterium was isolated from industrial wastewater effluent and identified as *Staphylococcus sciuri* (A-HS1) on the basis of morphological, biochemical tests and 16S rRNA ribotyping results. *S. sciuri* (A-HS1) demonstrated optimum growth at 37 °C and pH 7. *S. sciuri* (A-HS1) was able to resist Cr^{+6} (25 mM) as well as other heavy metals such as As^{+2} (19 mM), Pb^{+2} (18.5 mM), Zn^{+2} (17 mM), Cu^{+2} (2.5mM), Cd^{+2} (3 mM), and Ni^{+2} (1.5 mM). Biochemical characterization of chromate reductase enzyme showed its optimal pH as 8.0 and optimal temperature as 40â˚C. Chromate reductase enzyme activity was stimulated only by Mg^{+2} among other metals tested. Chromium biosorption efficiency (q) of *S. sciuri* A-HS1 was 42, 73, 85 and 31 mM/g after 2, 4, 6 and 8 days, respectively. Hexavalent chromium presence did not stimulate activities of APOX, SOD and CAT in significant quantities however a decrease in activities of these antioxidants were observed i.e., APOX (11%), SOD (8%), and CAT (3%), respective to the normal growth conditions. An increase in glutathione and other non-protein thiols levels played a significant role in combating the oxidative stress generated by the toxic metal cations. Pilot study demonstrated that S. sciuri A-HS1 was able to remove 87% Cr^{+6} from tannery effluent and 97 % Cr^{+6} from industrial effluent within 6 days of incubation. The present study revealed that S. sciuri A-HS1 may act as a potential candidate for the bioremediation of hexavalent chromium contaminated environmental sites.

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Oral

Environmental Microbiology **T006**

Newly Developed Multi-Species Bio-Testing Approach for Toxicity Assessment of Biodegraded Xenobiotics

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Environmental pollution with oil-based paints is underexplored and is a major concern nowadays due to their significant health hazards. Although, the bioremediation is supposed to be a cheap and environmental friendly method for biodegradation of pollutants, however novel, simple, reliable and cost-effective validation tools are need of hour.To design multi-species bio-testing model for toxicity assessment of biodegraded xenobiotics.Oil-based-paint degradation potential of strain KJ872855 was executed in an aqueous medium at culture conditions (37°C, 14 days, 160rpm) containing oil-based paint (Conc. 300ppm). The newly developed multi-species bio-testing approach included bio-assays to determine phytotoxic, cytotoxic and antimicrobial effect of treated and untreated oil-based paint using cell free supernatant. Additionally, the antioxidant activity of treated and untreated samples was also detected to confirm detoxification. The untreated oil-based paint displayed significant toxicity against all life forms. However after degradation, the cytotoxic effect against *Artemiasalina* revealed substantial detoxification of oil-based paint with LD50 of 121 µl ml-1 (without glucose) and > 400 µl ml-1 (with glucose). Similarly, the reduction in toxicity against *Raphanusraphanistrum* seeds germination (%FG = 98 to 100%) was also evident of successful detoxification under experimental conditions. Moreover, the toxicity against test bacterial strains and fungal strains was completely removed after bioremediation. Additionally, the biodegraded samples exhibiting reduced antioxidant activities (% scavenging = 23.5 ± 0.35 and 28.9±2.7) in both setups, respectively. Present multi-species bio-testing model in addition to antioxidant studies could be suggested as a reliable and cost-effective validation tool for biodegradation experiments.

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Oral

Soil & Agricultural Microbiology **T007**

Fungal Metabolites as Natural Herbicides

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Rumex dentatus is a broad-leaf weed of wheat. Chemical herbicides being used to combat the menace also cause damaging effects on environment and health. Scientists are in search of alternatives to safe environment and human health. The present study was, therefore, carried out to investigate herbicidal potential of culture filtrates of a phytopathogenic fungus *Drechslera australiensis* for management of *R. dentatus*. Fungal culture filtrates were prepared by incubating it at 26°C for 21 days using M-1-D broth as growth medium. The effect of these filtrates was studied on germination and growth of *R. dentatus* in laboratory bioassays and pot trial. For isolation of herbicidal constituents, culture filtrates were fractionated using four organic solvents. Herbicidal activity of crude fractions was assessed using leaf disc method. From crude chloroform fraction, 6 compounds were isolated with the help of preparative TLC followed by reversed phase HPLC. Herbicidal activity of the isolated compounds was determined by leaf disc bioassay using a synthetic herbicide 2-4-D as reference compound. Structure of the most herbicidal compound was elucidated by MS and NMR. In laboratory bioassay, culture filtrates significantly reduced seed germination, shoot biomass and root biomass of *R. dentatus* seedlings by 40%, 88% and 87%, respectively. In pot trials, culture filtrates significantly reduced shoot biomass of one- and two-week old plants by 60% and 46%, respectively. Among the various organic fractions, chloroform fraction exhibited the best herbicidal activity. From this fraction, 6 compounds were identified. One compound exhibited the best herbicidal activity resulting in high necrogenic activity accompanied by severe discoloration of leaf sections. This compound was identified as holadysenterine. The present study concludes that herbicidal activity of culture filtrates of *D. australiensis* was due to holadysenterine.

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Oral

Microbiology **T008**

Biological Control of Potato Common Scab with Rare Isatropolone C Compound Produced by Plant Growth Promoting *Streptomyces* A1RT

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Potato is prone to many drastic diseases like potato common scab (CS) in Pakistan. As no highly effective methods for managing CS, this study was to explore the possibility of biological control. Ten bacterial strains were isolated from CS-infected potato tubers from four different locations of Punjab, Pakistan and identified based on biochemical and molecular analysis, particularly polymerase chain reaction (PCR) and RFLP analysis of the amplicon. 16S rDNA region was amplified using species-specific primers. Isolated *Streptomyces* strains were confirmed as *Streptomyces scabies, S. turgidiscabies* and *S. stelliscabies*. Pathogenicity island was confirmed among the isolates after identification of *txtAB, nec1* and *tomA* genes with PCR amplification. One strain isolated from soil was antagonistic to the pathogenic Streptomyces spp., and confirmed as Streptomyces A1RT by 16s rRNA sequencing. Methanolic extract of *Streptomyces* A1RT showed a rare Isatropolone C compound production which was confirmed by 1H-NMR, 13C-NMR and 1H/1H-COSY, HMQC and HMBC techniques. Production of Indole-3-acetic acid (IAA) from *Streptomyces* A1RT was measured by spectrophotometry (26µgml^{-1}) and analyzed by HPLC. In a greenhouse assay, disease severity index was established from 0 to 500. Average diseases severity indexes were recorded as 63, 130.5 and 78 for *Streptomyces scabies, S. turgidiscabies* and *S. stelliscabies*, respectively. When *Streptomyces* A1RT was applied in soil that contained one of these pathogenic isolates, the average disease severity indexes were significantly ($P < 0.05$) reduced to 11.1, 5.6 and 8.4, respectively. A significant increase in tuber weight and shoot development was also observed with the tubers treated with *Streptomyces* A1RT.

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Human Genetics, Immunology **T009**

Role of Rs8177374 Single Nucleotide Polymorphism in TIRAP Gene in Modulation of Cytokines Level upon Plasmodium Exposure in the Pakistani Population

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Malaria is a mosquito borne infectious disease. Clinical outcomes of the disease vary among individuals infected with *Plasmodium*. Genetic back ground of the host modulates the immune response towards Plasmodium infection. TLR receptors of the host immune system recognize PAMPs and initiate the intracellular signaling, which culminates on the activation of NF- κBpathway. Activation of NF-κBupregulates the expression of inflammatory cytokines. Balance between the pro-inflammatory and anti-inflammatory cytokines mediates the level of disease severity. Variations in the TIRAP gene may modulate the immune response towards Plasmodium rather than the species of *Plasmodium*. This study investigates the level of cytokines in various genotypes of rs8177374 in malaria patients and healthy individuals.The study involves 454 samples including, 228 malaria patients and 226 healthy individuals. Malaria samples were divided into mild and severe malaria patients based on clinical symptoms and *P. vivax* and *P. falciparum* infected individuals based on the species of *Plasmodium* responsible for infection. rs4986790 polymorphism of TIRAP gene was investigated via allele specific PCR and RFLP based analysis. IFN-γ, TNF-α, IL-10 and TGF-βlevels were analyzed via ELISA kits (BioLegend).Increased levels of IFN-γand TNF-αwere observed in CC carriers of rs8177374 as compared with TT carriers ($p<0.05$). Increased IFN-γlevel was found in CC carriers of severe malaria and *P. vivax* exposed severe malaria groups.Presence of CC genotype may increase the level of inflammatory cytokines, causing severe malaria. Appropriate inflammatory response helps in parasitic clearance without severe systemic inflammation.

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Oral

Nanobiotechnology **T010**

Antifungal Activities of TiO_2 Nanotubes

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The ever-increasing interest in titanium oxide (Titania) is motivated by its applications in solar cells, biomaterials and photo-catalytic activities. Nanocrystalline titania is preferred in these applications dues to chemical stability, mechanical hardness, high refractive index and excellent transmission in the visible region. Titania exists in three different crystallographic phases i.e. anatase, rutile and brookite, amongst which brookite is the most difficult to synthesize. Anatase and rutile crystallize in tetragonal phase whereas brookite has orthorhombic phase. In the present work, titania nanoparticles are synthesized following sol-gel approach. $TiCl_4$ is used as precursor and ammonia as a gelation agent pH 1. pH of the sol was varied from 1 to 11. At pH 1 nanotubes show amorphous behavior whereas increasing the pH induces crystallinity in nanoparticles. The presence of (020), (202) and (321) confirms the formation of pure brookite phase at low synthesis temperature of 60 ºC. The absorption bands in FTIR analysis in the range of 450–700 cm^{-1}cm-1 corresponded to Ti–O–Ti stretching of titania. SEM micrographs show the formation of nanotubes with diameter 20nm and length ~1μm. Antifungal activities of the samples have been studied and remarkable behavior has been observed. Nanotubes synthesized at higher pH show the 100% resistance against fungal. The current investigation highlights that the synthesized nanotubes are promising in targeting fungal infection.

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Pharmaceutical Microbiology **T011**

Partial Purification of Bioactive Compounds from *Streptomyces* and their Evaluation for Antimicrobial, Antioxidant and *In-Vitro* Antitumor Activity

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Natural products are continuously providing their fair share for development of new clinical drug candidates against drug resistant pathogens. The actinomycetes from neglected habitats may provide high quality biological material for screening programs designed to detect novel bioactive secondary metabolites. In this study four selected Streptomyces strains including 10A, 9A, 15C and 20C have been investigated at preparative screening level for purification of active compounds and biological activities. Each strain cultivated as 10 L shaking flask cultures and crude extracts were prepared by bed extraction using XAD16-N resin from fermentation broth. The powder crude extracts of strains 10A and 15C were fractionated by manual silica gel columns (70–230 mesh), followed by separation on preparative thin layer chromatography (PTLC), and finally components were resolved by size exclusion chromatography using sephadexLH-20 column. In biological screening some partially purified fractionations were found active against the test organisms, *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis* and *Acinetobacter baumannii*, whereas crude extracts from strains had MIC value up to 3.2 mg/mL. The partially purified fraction (F2A1) from strain 10A exhibited 90% inhibition of DPPH free radical. In brine shrimp cytotoxicity assay strain 10A had moderate lethality and its partially purified compounds had varied % mortality in range of 0% to 94%. In MTT assay for *in vitro* antitumor activity against ATCC's HCT116 human colorectal carcinoma cell line, extract of strain 15C and partially purified compounds from it, exhibited high % cell mortality. Overall, among 37 partially purified fractions, 15 exhibited promising biological activity, while 5 fractions have maximum purity when examined by TLC and HPLC. The partially purified components can be further purified for spectroscopic studies (mass spectrometry and NMR spectroscopy) for final identification and structural elucidation.

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Oral

Environmental Microbiology **T012**

Evaluation of Growth Promotion in *Lens culinaris* by *Bacillus Spp*. with Inorganic Fertilizers and Subsequent Selenium Bio-Fortification

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Malnutrition is the global issue that is resulting into the micronutrients deficiencies worldwide. These deficiencies are causing incurable mental and physical disorders. Several strategies are devised to fulfill the nutritional requirements of the human race. Most of them are too expensive to be implemented in the far away areas. However biofortification is an easy, widely applicable and inexpensive way to increase the nutritional content of food. Biofortification of Selenium is substantial as it has prevalent use in our body at cellular and molecular level and is also preventive from several metabolic and physiological diseases. Various microbial species are capable of hyperaccumulating the Se in different types of plants. *Lens culinaris* is proved to successfully biofortify Se by agronomic biofortification and in present study it provided remarkable increase in the Se content with the help of biofertilizers. The results presented the exceedingly increased selenium level in bacterial inoculated and selenium treated plants as compared to selenium treated control plants that was checked by inductively coupled plasma atomic emission spectroscopy (ICP-AES). This study shows that the Se biofortification by using biofertilizers is a cost effective method to reduce the prevalence of Se deficiencies worldwide.

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Oral

Nanobiotechnology **T013**

Microwave Assisted Sol Gel TiO_2 Doped Zirconia Nanocrystallites for Dental Fillers

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To obtain dense and monodispersed nanometric particles with phase uniformity is a crucial factor for obtaining nanocrystalline ceramics. For this purpose microwave assisted sol gel has gain much attention due to its rapid heating, improved production rate, enhancement in densification, grain growth inhibition. In a microwave oven, ceramic absorb energy though dielectric heating, and the temperature is more uniformly distributed in the ceramic material. The increasing demand for ceramic restorations with a Zirconia (ZrO_2), the micro wave assisted sol gel fulfill all requirements. Moreover, the basic problem with zirconia is existence of mix phases. Hence formation of single phase zirconia at relatively low microwave powers is still a task. Among different dopants TiO_2 has stabilized zirconia at relatively low temperatures along with its biocompatibility. In this research work TiO_2 has doped with 1wt% and subjected to different microwave powers (140, 280, 420, 560 and 700W). XRD rietveld refinement results show the formation of pure tetragonal zirconia (t-ZrO_2) at relatively lower microwave powers (420W, 560W and 700W). Crystallite size of TiO_2 doped zirconia is consistent with literature for occurrence of t-ZrO_2. Mixed phases of zirconia exhibit relatively higher crystallite size at low microwave powers. Further increase in microwave power leads tov sharp decrease in crystallite size with phase shift from mixed to phase pure t-ZrO_2. After stabilization of t-ZrO_2 crystallite size began to increase. SEM shows the formation of dense and monodispersed nano sized particles. Formation of dense particles leads to higher value of hardness confirmed by Vickers Micro indenter. Dielectric constant shows a very high value ~625 at low frequencies. Zone of inhibition for these nanoparticles was found to be 28mm against bacteria.

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Oral

Human Genetics **T014**

Effect of Incubation Time of Adipose Tissue on Stem Cell Regenerative Potential

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Human adipose tissue is an ideal source of adult stem cells for cell based regenerative therapies. Adipose tissue derived mesenchymal stem cells (AT-MSCs) are ideal for treatment of several diseases and disorders. The main advantage of using adipose tissue over other sources e.g. bone marrow, includes of less invasive harvesting procedures and high number of regenerative cells. In current study, we examined the effect of incubation time (before processing of adipose tissue) on cell viability, proliferation and differentiation potential of AT-MSCs. Harvested adipose tissue was incubated at 4oC for either 2 hours (group 1) or 24 hours (group 2). Adipose tissue was enzymatically digested after incubation period and total cell count and CFU was performed with haemocytometer and colony forming assay for each group. Effect of incubation time on population doubling, doubling time and proliferation potential was determined for each group. AT-MSCs were differentiated into adipose, bone, cartilage and neurons using respective differentiation media. Differentiation into adipose, bone, cartilage and neuron was analysed quantitatively and qualitatively. AT-MSCs extracted from each group showed spindle shape morphology and plastic adherence growth with great proliferation potential. Percentage viability, total cell count and number of CFU were similar in each group. Similarly proliferative potential, population doubling and doubling time were unchanged even after 24 hour incubation. In both groups the differentiation of AT-MSCs was similar as indicated by morphological changes and number of differentiated cells24-hours delay in adipose processing and MSCs extraction has no difference on cell viability, proliferation and differentiation potential of MSCs from those extracted immediately after sample collection.

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Oral

Immunology **T015**

Prevalence of IgM and IgG in Pediatric Patients Suspected of Having Typhoid Fever

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Typhoid fever is a systemic infection caused by *Salmonella enterica* serotype *Typhi*. This bacterium has remarkable mechanisms for persistence in its host. Typhoid fever is of major concern in tropical regions of the world. Highest episodes of typhoid fever occur in Asia i.e.93%. Early diagnosis of the disease is mandatory to lower the mortality rate associated with it as well as to prevent the emergence of antimicrobial drug resistance by *Salmonella typhi*. Research work was conducted in Immunology Department of the Children's Hospital, Lahore for the period of one year. The study includes a total of 60 patients suspected of having typhoid fever. Serum samples of these patients were tested for Typhidot IgG and IgM antibodies as well as for the antibodies against TO and TH antigens using Widal test. Of the total 60 patients, 10 (16.7%) were positive for both Typhidot IgG and IgM, 16 (26.7%) were positive for Typhidot IgM, 3 (5%) were Positive for typhidot IgG and 31 (51.7%) were negative for both Typhidot IgG and IgM. While concerning the results of Widal test, 8 (13.3%) were positive for Widal TO and TH antigens, 3 (5%) were positive for Widal TO antigen, 10 (13.7%) were positive for Widal TH antigen and 30 (50%) were negative for Widal TO and TH antigens. IgM is positive at the early stage of acute typhoid fever, IgM along with IgG is positive at the middle stage of acute illness. In areas, where typhoid fever usually persists, only IgG positive cases are also observed. The detection of only IgG cannot discriminate between acute and convalescent phases as it can stay in the serum for at least 2 years and above. The Typhidot test is much helpful for the rapid diagnosis of typhoid fever as compared to Widal test. By testing the rise of IgM and IgG antibodies against *Salmonella typhi*, we are able to detect the infection at early and late stages, respectively.

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Oral

Virology **T016**

First Report Molecular Characterization of Complete Monopartite Begomovirus Complex Infecting Parthenium Hysterophorus in Pakistan

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During a survey in December 2013, severe viral disease leaf curling was observed in *P. hysterophorus* weed from the vicinity of University of the Punjab, Lahore, Pakistan. Genomic DNA was extracted by Doyle and Doyle method then rolling circle amplification (RCA) subjected using phi29 DNA polymerase (Thermo-Scientific). The diluted RCA product was exploited in PCR to amplify whole begomovirus complex. In present study sequenced old world begomovirus genome shared nucleotide sequence identity at 94.2% with Cherry tomato leaf curl virus (CToLCV). The alphasatellite shared maximum nucleotide sequence 94.3% to Tobacco curly shoot alphasatellite (TbCSA). The identified betasatellite shared maximum nucleotide sequence identities at 97.5% to Papaya leaf curl betasatellite (PaLCuB). To the best of our knowledge this is a novel begomovirus disease complex infecting *P. hysterophorus* in Pakistan. Furthermore, this is the first report of CToLCV associated with DNA satellites and infecting a weed host in Indo-Pak subcontinent.

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Oral

Environmental Microbiology **T017**

Isolation and Molecular Characterization of Toluene Metabolizing Bacteria from Tannery Effluent

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Toluene is a highly toxic and hazardous substance and is classified as priority pollutant by U. S. Environmental Protection Agency (EPA). For its amputation from the environment, bioremediation using toluene metabolizing bacteria is one of the emerging steps. The present study was conducted to isolate toluene degrading bacteria from some tannery effluents. For isolation of bacteria selective enrichment and serial dilution methods were employed. Growth curves and toluene removal efficiencies of the isolated bacteria were documented. Molecular characterization involved DNA extraction followed by PCR amplification of 16S rRNA gene. The amplified PCR products were sequenced and retrieved DNA sequences were subjected to BLAST analysis for molecular identification of these bacteria. In the present study, total 21 toluene metabolizing bacteria were isolated and characterised. Among these isolates few (i.e., IUBT2, IUBT3, IUBT5, IUBT8, IUBT9, IUBT10, IUBT12, IUBT14, IUBT15, IUBT16, IUBT17, IUBT18, IUBT19, IUBT21, IUBT22, IUBT23 and IUBT26) exhibited similarity to *Brevibacillus agri* strain NBRC 15538 while IUBT4, IUBT24 and IUBT28 shared significant homology with *Bacillus paralicheniformis* strain KJ-16. However, IUBT11 was identified as a unique bacterium sharing lower sequence identity with the available database sequences. Many of the bacterial isolates possessed high toluene removal efficiency. Using bioremediation approach, potential of these extremely efficient toluene metabolizing bacteria can be harnessed to mitigate toluene pollution in the environment.

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Oral

Microbial Genetics **T018**

Analysis of Mutations Associated with MDR *Mycobacterium tuberculosis* Strains from Punjab

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Tuberculosis was the cause of 1.5 million deaths in 2015. Pakistan is among high burden TB countries. This high burden is attributed to lack of rapid diagnostic methods. Molecular methods provide a solution to this malady. These methods not only help in early detection, but also in correct prescribing of the drug regimen. This study was aimed to find out the mutations in genes associated with resistance to first line anti-TB drugs. Sputum samples were collected from different cities of Punjab and processed by fluorescent staining, LJ culturing, and proportion method to determine the drug resistance. DNA was extracted by Genolyse DNA isolation kit. For mutational analysis of Isoniazid and Rifampicin, Line Probe assay was performed and for streptomycin & ethambutol, amplification of rrs, rpsl and embB genes was done. Purified amplicons were sent for sequencing to First Base Asia Singapore. Sequences were analyzed by Bio-informatics tools including conserved domains and Phylogenetic analysis. Total 625 samples were collected but 153 samples were rejected as they didn't provide the complete information. Mean age was 35.6±14.7 years. Culturing was found to be more sensitive than microscopy. Out of 522 samples, 174 strains were found to be MDR on DST and 117 strains on LPA. S531L, S315T1 and C15T were the most common mutations identified. Sensitivity of rifampicin and isoniazid was 87% and 84.47% respectively. The mutations identified by LPA was 85%, 70% and 20% in rpoB, katG and inhA genes respectively. Sequencing of rrs gene showed many frame shift mutations. G76T and M306V were most prevalent mutations identified in rpsl and embB genes respectively. In some phenotypic resistance strains, no mutation was identified on sequencing. In Punjab, Pakistan the most prevalent mutations were S531L, S315T1 and C15T in rpoB, katG, and inhA genes respectively. Whereas the most common mutations identified in rrs, rpsl and embB genes were A429-, G76T and M306V respectively.

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Oral

Bioinformatics/ Computational Biology **T019**

Comparative *In Silico* Screening of Synthetic Molecules and Dietary Phytochemicals as Inhibitors of Human Acetylcholinesterase (hAChE) and Designing of New More Potent Anti-Alzheimer Drug Candidates

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Alzheimer's disease (AD), the most common cause of dementia is a progressive neurodegenerative disorder leading to irreversible loss of 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 with a group of chemicals known as acetylcholinesterase (AChE) inhibitors. Drug discovery today has become immensely dependent on computational approaches. Present study presents in-silico screening for potential anti-Alzheimer drug via molecular docking and estimation of optimal solubility and permeability. Six FDA approved drugs for AD were compared with their 300 derivatives and dietary phytochemicals present in online databases on the basis of docking score as inhibitors of modeled human acetylcholinesterase (hAChE). The 42 lead derivatives were subjected to Lipinski's 'rule of five'to determine their oral bioavailability. Inhibitors obtained after screening were mostly dual binding site inhibitors having two binding subunits with a usually 8-12 carbon chain in between (second generation AD drugs). Yohimbine, Berberastine, Berberine, Sanguinarine, Elemol, Naringenin and Viridiflorol are the worth mentioning phytochemicals as anti-Alzheimer drugs. 15 new second generation AD drugs were designed that are significantly more potent than previous drugs. Using in-silico drug discovery methods, bioactive compounds present in online chemical databases can be screened to develop more efficient and safer drugs against cognitive symptoms of AD.

Oral

Nanobiotechnology **T020**

Green Synthesis, Characterization and Toxicity Studies of Plant Synthesized Gold Nanostructures

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Advancements in the field of nanobiotechnology has enabled the scientists to look for benign methods of nanoparticle production as the conventional chemical approaches are potentially harmful to both environment and humans. The biocompatibility of chemically synthesized particles remains questionable forcing researchers to invent nontoxic and cheap synthesis methods. Green technology is a new approach employed by scientists to synthesize biologically compatible and ecologically friendly nanoparticles. The study focuses on the green synthesis of gold nanocrystals using three different and completely unrelated plants. Aqueous leaf extracts of Clove, Basil and Pomegranate were used as the reducing and stabilizing agents. The extracts reduced HAuCl4 to Au0 giving an instant violet/purple color with a defined absorption peak at 545 nm on the UV-Visible spectrum. Characterization of particles using Scanning electron microscopy showed spherical and elliptical shaped particles with size ranges between 50-150 nm. Energy dispersive X-ray diffraction pattern of particles confirmed the elemental origin of the particles giving prominent Au peaks. The antimicrobial activity and anticancer potential of the naked green synthesized particles showed Pomegranate synthesized Gold particles to be the most effective against marine isolated Bacillus and Vibrio strains. While no defined anticancer potential was notable from time dependent in vitro application of the particles on Leukemia cell lines. Cellular morphology after particle application showed aggregates but the MTT assay showed no concentration dependent cell death. Nontoxic gold particles were generated with poor bactericidal effects and no cytotoxic effects. This makes these benign particles as prospective carriers of drugs and genes opening new arenas for their application in imaging studies as well as diagnostics.

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Oral

Cancer Genetics **T021**

Decreased Cellular Cholesterol Levels in Acute Leukemia Cells

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Cancer cells are shown to deposit intracellular cholesterol but there are inconsistent evidences available on intracellular cholesterol levels in leukemia cells. For instance, according to a relatively recent study by Casalou *et al*., (2011), primary myeloid leukemia cells display significant increase in intracellular cholesterol deposits. Another study by Mulas et al., (2011) indicated that total cholesterol levels in primary acute lymphocytic leukemia cells are not significantly different than in PBMCs from healthy donors. On the other hand, some earlier works reported decrease, in intracellular cholesterol levels of leukemia cells in comparison to peripheral blood mononuclear cells (PBMCs) from healthy donor. Intracellular cholesterol levels were compared among acute lymphocytic leukemia (ALL) cells, acute myeloid leukemia (AML) cells and peripheral blood mononuclear cells (PBMCs) from the healthy subjects by using multiple methodological approachesSignificantly lower intracellular levels of total cholesterol were found in PBMCs from ALL (n=7) and AML (n = 7) patients as compared to PBMCs from the healthy subjects (n = 26). Similarly, acute leukemia cell lines also displayed significantly lower intracellular levels of total cholesterol. In addition, free cholesterol and cholesteryl ester were also quantified in both ALL and AML cell lines. Again it was noticed that levels of free cholesterol and cholesteryl ester contents were significantly lower in leukemic cells as compared to normal cells. Approximately 90% of the total cholesterol was found in the form of cholesteryl esters in leukemia cells as well as in normal PBMCs. Present work provides convincing evidence to confirm lower levels of intracellular cholesterol in acute leukemia cells. Moreover, most of the cholesterol was found in the form of cholesteryl esters in leukemia cells.

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Oral

Human Genetics **T022**

Distribution of XmnI-158 I^{3}G Variant in Beta Thalassemia Major in Pakistan

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β-thalassemia is a monogenic inherited quantitative blood disorder with considerable heterogeneous clinical manifestations. A multicenter study aimed to investigate frequency of HBG2:c.-211C>T base-pair substitution (historically described as -158 G XmnI polymorphism) among homozygous or compound heterozygous beta thalassemia patients and relationship between beta and alpha genotypes. One hundred and sixty one molecularly identified beta thalassemia patients were Gamma (HBG2) Globin gene XmnI polymorphism using a restriction fragment length polymerase (RFLP) based PCR. All data were compared and analysed by SPSS software. The -158 GγXmnI polymorphism was present in 57 (36%) patients in which HBB:c.27_28insG (p.Ser10Valfs*14)was the most common beta-mutation 33 (34.5%). In patients with both the alpha globin deletion and XmnI polymorphism, the clinical onset of thalassemia occurred later in life (e.g. 1.8 to 2 years of age). A statistically significant (p-value 0.01) relationship was found between XmnI polymorphism and beta thalassemia while no significant association between XmnI polymorphism and FBT was observed. Monogenic disorders showing a wide spectrum of disease severity demands a better prognosis and treatment to ameliorate the disease in the long run.

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Oral

Virology **T023**

A Single Dose of Lytic Bacteriophage RLP can Inhibit Growth of Multi Drug Resistant *Pseudomonas aeruginosa* for 20 Hours

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Antimicrobial resistance is a serious threat to health around the globe, according to world health organization (WHO), we will enter pre-penicillin era by 2050 if no new antimicrobials are discovered. The search for new antimicrobials and the alternative is of prime importance. Among some alternatives Phage therapy is a potent candidate for treatment of MDR infectious agents. The bacteriophage RLP has been isolated from River Ravi Near Lahore. Its host is *Pseudomonas aeruginosa*. It forms clear, transparent circular plaques with *P. aeruginosa*. RLP is stable at PH of 5-9 and temperature range of 4-65^{o}C. It can inhibit bacterial growth upto 20 hours at MOI 10 in vitro. RLP infects mostly *P.aeruginosa* strains and some others Gram Negative bacteria. The Genome of RLP is double stranded DNA and is 43Kbps in size. The genome was sequenced using illumine Next Generation Sequencing (NGS) and assembled using Abbyys, Velvet and CLC workbench softwares. The annotation was done using RAST server and Phaster (www.phaster.ca). The annotation revealed 52 genes in RLP genome. Out of these 52 CDS (coding DNA sequences) 24 codes for hypothetical proteins and 28 codes for proteins of known functions. Out of these 28 known proteins, 18 codes for structural proteins whereas 10 codes for enzymes. A cascade system of the lytic machinery is also present in the form of 4 CDS i.e R/Z, R/Z1, holin and endolysin. Endolysin and Holin genes are amplified using PCR and cloned in pet-28a vectors. The expression of endolysin and Holin is done using BL-21 cells. The protein are separated by Ni-affinity chromatography. The activity of the enzymes is tested through zymography.

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Oral

Metabolomics **T024**

Metabolic Fingerprinting: Classification of Grapevine using NMR and Multivariate Data Analyses

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Grapevine (*Vitis vinifera* ssp.) and grapes have been extensively studied due to their numerous nutritional benefits and health affecting activities. In this study, metabolite fingerprinting of crude leaf extracts, based on 1H nuclear magnetic resonance (NMR) spectroscopy and multivariate data analyses, has been used for the metabolic characterization of different grapevine cultivars. Several two-dimensional (2D)-NMR techniques were also employed leading to the identification of a number of different types of compounds. Various multivariate data analyses of the processed 1H NMR data revealed clear differences among the samples. Metabolites responsible for the discrimination in different grapevine cultivars belong to major classes, that is, organic acids, amino acids, carbohydrates, phenylpropanoids and flavonoids. A differentiation of the cultivars based on their resistance to pathogen was also achieved, and metabolites associated with this trait, namely, quercetin-3-O-glucoside and a trans-feruloyl derivative, were identified. On the basis of these results, the distribution of different plant metabolites among the different grapevine cultivars is presented.

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Oral

Antimicrobial Agents **T025**

Appraisal of Antifungal Potential of Selected Plants against *Rhizoctonia solani* Kuhn

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In present study the antifungal activity of *Oxalis corniculata* L. and *Hibiscus rosa-sinensis* L was assesses against *Rhizoctonia solaniKühn*. In vitro bioassay with methanolic leaf and stem extracts was carried out with six different concentrations viz. 1%. 1.5%, 2%, 2.5%, 3% and 3.5%. Maximum inhibition in *R. solani* growth was observed in 3.5% treatment in *O. corniculata*. Various organic fractions viz. n-hexane, chloroform, ethyl acetate and n-butanol of methanolic extracts of *O. corniculata* were isolated and in vitro antifungal potential of each fraction was assessed. n- hexane fraction of *O. corniculata* significantly inhibited the test fungal growth upto 81%- 89%. GC-MS analysis of n- hexane fraction of *O. corniculata* was done and twenty two compounds belonging to alkane, acyclic, aromatic hydrocarbons, aromatic carboxylic acid, aromatic carboxylic hydrocarbons and saturated fatty acids were identified. Reverse transcriptase polymerase chain reaction of cDNA of *R. solani* grown in 3.5% methanolic extract of *O. corniculata* and control treatment was executed. The selected gene specific primers (ACT & CAT) were used to determine transcript levels for known genes in the genome of *R. solani*. No change was observed in the expression of housekeeping genes in control while expression level of catalase gene enhanced under stress.

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Oral

Diagnostic Techniques **T026**

Multi-Epitope Fusion Antigens of *Mycobacterium tuberculosis* for Enhancing Sensitivity in Serodiagnosis of Tuberculosis

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Serodiagnosis of tuberculosis requires detection of antibodies against multiple antigens of *Mycobacterium tuberculosis*, because antibody profiles differ among patients. Using fusion proteins with epitopes from two or more antigens would facilitate in detection of multiple antibodies. Fusion constructs tn1FbpC1-tnPstS1 and tn2FbpC1-tnPstS1 were produced by linking truncated regions of variable lengths from FbpC1 to the N-terminus of truncated PstS1. Similarly, a truncated fragment of HSP was linked to the N-terminus of a truncated fragment from FbpC1 to produce tnHSP-tn1FbpC1. ELISA analysis of plasma samples of TB patients against tn2FbpC1-tnPstS1 showed 72.2% sensitivity which is nearly the same as the expected combined value for the two individual antigens. However, sensitivity of tn1FbpC1-tnPstS1 was lowered to 60%. tnHSP- tn1FbpC1 showed 67.7% sensitivity which is slightly less than the expected combined value for the two individual antigens, but still significantly higher than that of each of the individual antigen. Data for secondary structure analysis by CD spectrometry was in reasonable agreement with the X-ray crystallographic data of native proteins and predicted structure of fusion proteins. Comparative molecular modeling suggests that epitopes of constituent proteins are better exposed in tn2FbpC1-tnPstS1 as compared to those in tn1FbpC1- tnPstS1. Therefore, removal of N-terminal non-epitopic region of FbpC1 from 34–96 amino acids seems to have unmasked at least some of the epitopes, resulting in greater sensitivity. High level of sensitivity of tn2FbpC1-tnPstS1 and tnHSP-tn1FbpC1, not reported before, shows that these fusion proteins have great potential for use in serodiagnosis of tuberculosis.

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Oral

Nanobiotechnology **T027**

Effect of Microwave Radiation on Structural and Magnetic Properties of Fe_3O_4 Doped Zirconia Nanoparticles and their Antibacterial Study

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This study aimed to identify the effect of the microwave powers on the structural and magnetic properties of Fe_3O_4doped zirconia for biomedical applications. Fe_3O_4 doped zirconia was heated at different microwave powers (136, 440, and 800W). X-Ray Diffraction (XRD) results demonstrate the formation of pure tetragonal zirconia (t-ZrO_2) at microwave power 440W without any further heat treatments. Crystallites size calculated from XRD data (~23nm) is in good agreement with literature for stabilization of t-ZrO_2. Microwave energy dissipation results in stresses, generated in the heated zone cause shrinkage in volume, consequently leads to transformation of monoclinic phase to tetragonal. Higher x- ray density with pure tetragonal phase has been observed. VSM results show ferromagnetic behavior with relatively low value of coercivity (600Oe) and saturation magnetization (~2emu/g). Scanning electron microscopy reveals the formation of well separated spherical nanoparticles (NPs) with diameter about 35nm. It is worth mentioning here, to the best of our knowledge these Fe_3O_4 doped ZrO_2 NPs have never been reported at very low microwave power (440W) with phase purity without any further heat treatment and modifications. Antibacterial study of Fe_3O_4 stabilized zirconia NPs was carried by streaking method and they show inhibition zone up to 24 mm.

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Oral

Soil & Agricultural Microbiology **T028**

Evaluation of Genotoxicity of Pesticides (Lufenuron And Emamectin) using the *Allium cepa* Assay

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Over the past few decades, the use of pesticides has been increased enormously in agriculture. These pesticides help in combating pests (insects, weeds etc), but at the same time, have potential threats for the plant itself and its consumers. In this, study two pesticides, lufenuron and emamectin were evaluated for genotoxicity and mutagenicity using Allium cepa chromosomal aberration test and Ames test, respectively. For genotoxicity evaluation, 7 concentrations (D1= 0.05%, D2= 0.1%, D3= 0.15%, D4= 0.2%, D5= 0.4%, D6= 0.6%and D7= 0.8%) of both pesticides were used. Both pesticides were involved in decreased germination rates for onion seeds, inhibition of root length and decreased mitotic indices when compared to negative control (autoclaved water) in dose dependant manner. While the chromosomal aberrations (micro nuclei, vacuolated nuclei, chromosomal breakage, sticky chromosomes, vagrant chromosomes and chromosomal bridges) increased as the pesticide concentration increased. For mutagenicity detection, Ames test was performed using two *Salmonella typhimurium* strains i.e. TA98 and TA97a to detect frame shift mutations. For lufenuron doses used were LD1= 50%, LD2= 75% and LD3= 100%. For emamectin doses used were ED1= 0.05%, ED2= 0.1% and ED3= 0.5%. For both pesticides at least one concentration showed high mutagenicity. So, it was confirmed by that both pesticides have genotoxic and mutagenic effects. Additional mutagenicity tests should be conducted for further validation of these results which will confirm the predictions for pesticide's effect in an organism.

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Oral

Environmental Microbiology **T029**

Assessment of Airborne Bacteria from Orthopedic Wards of Different Hospitals in Lahore

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Investigation of hospital air quality is necessary because patient health is more affected by indoor air as compared to outdoor air. In this study, airborne bacterial load of orthopedic wards of seven hospitals in Lahore was investigated. 28 samples were collected using filtration technique for bioaerosol sampling. Blood agar and MacConkey agar were used to culture bacteria. The average airborne bacterial load investigated was 5.2— 106 CFU/m^3 which was much higher than the indoor air quality standard. Average airborne bacterial load was highest for emergencies (9.7— 106 CFU/m^3). Minimum average bacterial load was calculated for operation theatres i.e., 5.9 — 105 CFU/m3. Total 143 bacterial strains were isolated. Different bacterial genera isolated were *Bacillus* (36.36 %), *Staphylococcus* (45.5 % including 4.9 % *S. aureus*), *Micrococococcus* (5.9 %), *Corynebacterium* (4.2 %), *Pseudomonas* (4 %) and *Enterobactericeae* (8.4 %). Most of the strains were multiple drug resistant. 40 % *S. aureus* strains were MRSA. Among Gram negative bacteria, 50 % were suspected to be extended spectrum beta lactamase producers. This study investigated a high airborne bacterial load and presence of multiple drug resistant bacteria (including MRSA and ESBLs) in hospitals of Lahore. Our findings represent the potential risk of nosocomial infections and possible transfer of resistance genes to pathogenic bacteria.

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Oral

Environmental Microbiology **T030**

Bacterial Enhanced Selenium Biofortification: Raising Selenium Content of Crops in Se-Deficient Soils

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Toxic heavy metals in air, soil, and water are global problems because of industrialization that is a growing threat to humanity. Selenium has become the element of interest to many investigators because of its toxicological and physiological importance. Selenium is a trace element that is essential in small amounts, but like all essential elements, it is toxic at high concentration. Most important forms of selenium are Selenite, selenate and elemental selenium. Both selenite and selenate are toxic. It is used in pesticides, herbicides, oil refineries, coal carbonization, photocopy machines, electronics, and glass manufacturing as well as production of selenium-containing wastes during smelting and mining operations. Several studies show that selenium is highly toxic to animals including humans. Chemical detoxification of metal- has proven to be very expensive and often results in secondary effects on the environment. Consequently, more sustainable biological solutions need to be found. Bacteria have ability to transform toxic selenium oxyanions (Selenate and Selenite) into less toxic elemental selenium. Elemental selenium being the insoluble in water is less mobile and usually remains in soils posing a smaller risk of exposure. Present study deals with the selenium reducing bacteria and their impact on plant growth and bio-fortification of selenium.

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Oral

Biochemistry **T031**

Production and Characterization of Protease from Geobacillus SBS-4S

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Proteases are hydrolytic enzymes responsible for proteolytic cleavage of proteins by breakdown of the peptide bonds. Current study was designed to get hyper production of protease from *Geobacillus* SBS-4S through submerged fermentation. Different agricultural wastes i.e molasses, wheat bran and rice bran were utilized as carbon substrates and yeast extract, peptone and tryptone were utilized as nitrogen substrates, through fermentation with *Geobacillus* SBS-4S.The maximal protease production (178.46g/mL) was recorded when 5% wheat bran was used as carbon source with a growth at 60°C, pH 7.0, 19 hours of incubation period with 1% (v/v) inoculum size and agitation speed of 120 rpm. Among different nitrogen sources investigated, 2% yeast extract produced higher amount of protease (126.75Î¼g/mL) with an activity of (24.4 units/mL). The protease produced by *Geobacillus* SBS-4S was purified through column chromatography and the purity of enzyme was analyzed through SDS-PAGE. The characterization studies demonstrated that enzyme showed maximal activity at 60oC and pH 9.0. Triton X-100, Tween-80, Tween-20 and SDS in assay mixture resulted in the reduction of enzyme activity to 87, 69, 82 and 92% respectively. The locally produced protease was utilized to examine its efficacy on the growth of poultry birds. The results showed that the supplementation of feed with the locally produced protease showed significant effect on weight gain, feed consumption and feed efficiency of birds in third, fourth and fifth weeks of trials. The higher level stability at alkaline pH and poultry trials demonstrated that this enzyme is suitable for its use in detergent and poultry feed industry, however further experiment at large scale will be required for its use in the above said industries.

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Oral

Environmental Microbiology **T032**

Isolation and Molecular Characterization of Chromium Resistant *Staphylococcus aureus* from Industrial Effluent

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Chromium is of great economic importance due to its uses in different industries; however, it is also becoming a major environmental pollutant due to uncontrolled discharge in industrial effluent. This is due to lack of proper legislations and monitoring systems in developing countries such as Pakistan. Therefore, bioremediation of contaminated wastewater using metal resistant bacteria could be considered as cost-effective and environmentally benign approach. This is because the bacteria present in extreme environment(s) such as contaminated effluents could have well adapted and might be exploited for the bio-treatment of metal contaminated industrial effluents. For this purpose, sample from metal contaminated effluents (n=18) were collected from discharge point of different industries from Kala Shah Kaku city. Culture-able bacteria (n=40) were isolated through serial dilution method and were evaluated for chromium tolerance ability by enrichment medium. Only one bacterium (strain K1) was able to tolerate 22mM Cr^{6+} that was further characterized by VITEK® 2 system and 16S rRNA gene sequencing. Both biochemical and phylogenetic analyses confirmed that bacterium strain K1 belonged to Staphylococcus aureus exhibiting optimum growth at 35°C, (pH =8.0). Under optimum conditions, it could remove about 80% chromium (Cr^{6+}) after 16 hours. Data regarding SEM-EDX analyses confirmed enlargement of cells with irregular surface in presence of chromium. FTIR results assumed that carboxyl, amino and phosphate groups of cell wall were involved in complexation with chromium. Our results have suggested that metal tolerant bacteria can be isolated from contaminated environment that might be used for bioremediation of chromium present in industrial effluents.

MMG 2018
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Oral

Antimicrobial Agents and Chemotherapy **T033**

Antibacterial Activity of Endophytic Fungi Isolated from Local Medicinal Plants of Mansehra

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The secondary metabolites secreted by entophytic fungi of medicinal plants showed many biological activities. The problem of antibiotic resistance in many common human pathogenic bacteria can be solved by checking antibacterial activities of endophytic fungi. In present investigation the endophytic fungi of *Hordeum vulgare* and *Hyoscyamus niger* were evaluated for their antibacterial activities against 6 human pathogenic bacteria. 12 endophytic fungi were isolated and identified from different plant tissues. The antibacterial activity was evaluated by using agar well diffusion method. Minimum inhibitory concentration was determined by using microbroth dilution method. Out of the total 12 different endophytic fungal strains 5 endophytic fungal extracts showed activity against all tested bacterial strains. The endophytic fungi which belong to *Aspergillus* and *Curvularia* genus comparatively showed good antibacterial activity. Maximum zone of inhibition (16.25 mm) was shown by extracts of *Aspergillus flavus* against *Salmonella typhi*. The fungal species *Fusarium oxysporum, Aspergillus fumigatus, Curvularia lunata* and *Pernicillium Chrysogenum* also showed good inhibitory effect against selected bacterial strains. The MIC values were ranged between 14.8 µg/well to 185 µg/well. The endophytic fungal extracts showed maximum zone of inhibition against gram negative bacteria. Ethanolic extracts of *Aspergillus flavus, Curvularia lunata, Penicillium chrysogenum* and *Mucor hiemalis* revealed significant antibacterial activity against the tested bacterial strains as compared to methanolic and n-hexane extracts. The comparative phytochemical screening of crude extracts of isolated endophytic fungi and plant extracts was also carried out. Many significant secondary metabolites were commonly found in crude extracts of plants and endophytic fungi.

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Oral

Biochemistry **T034**

DNA Methyltransferase 3b (Dnmt3b) Mediated Regulation Of Dipeptidyl Peptidase 6 Expression and Its Novel Function During Retinoic Acid Induced Neuronal Differentiation of P19 Cells

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DNA methylation is carried out by a group of DNA methyltransferases enzymes that include Dnmt1, Dnmt3a, and Dnmt3b which are involved in the establishment of genomic methylation patterns during development and differentiation. The present study deals with the role of Dnmts during RA induced neuronal differentiation of P19 stem cells. As a first step, we studied the expression of Dnmts during neuronal differentiation of P19 cells by using real time PCR and western blot analysis. The results showed that the mRNA as well as protein level of Dnmt3b is increased while the expression of Dnmt1 and Dnmt3a is decreased following RA treatment of P19 cells. These results lead us to construct a chromatin library of Dnmt3b enriched DNA fragments which identified several novel targets of Dnmt3b including Dpp6. In this report, we described the regulation of Dipeptidyl peptidase 6 (Dpp6) expression and its novel function during RA induced neuronal differentiation of P19 stem cells. Dpp6 was verified to be a target of Dnmt3b by using ChIP analysis followed by PCR analysis. Dpp6 gene promoter was heavily methylated in P19 cells as demonstrated by bisulfite genomic sequencing, COBRA, and methylation specific PCR. Specific depletion of Dnmt3b using shRNA based silencing resulted in increased mRNA and protein expression of Dpp6. In agreement with the expression analysis, the average methylation level of Dpp6 gene promoter was reduced to half in Dnmt3b knockdown cells. We also showed that in the absence of Dnmt3b, Dnmt3a was recruited to the Dpp6 gene promoter and regulated its expression and methylation in P19 cells. Ectopic expression of Dpp6 resulted in impaired neuronal differentiation, altered cell proliferation and apoptosis following RA treatment of P19 cells. Taken together, the present study described the regulation of Dpp6 gene and demonstrated a novel role of Dpp6 in RA induced neuronal differentiation of P19 cells.

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Oral

Cancer Genetics **T035**

Effects of Aunps on Transient Receptor Potential (TRP) Channel Gene Expressions In SKBR3 Breast Cancer Cell Line

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The most abundant malignancy observed in women is breast cancer and in metastatic disease its mortality rate is high. Cancer progression and malignant growth is related to ion channels like transient receptor potential (TRP). In recent study TRP channels are considered as novel treatment regime in breast cancer. In our hypothesis, effect of gold nanoparticles (AuNPs) can be effective on the expression levels of TRP channel genes in breast cancer line (SKBR3) and mammary epithelial cell line (CRL-8798). Real-time PCR (RT-PCR) was used to measure the expression level of TRP genes. We observed that TRPA1 and TRPM8 expression levels in SKBR3 breast cancer cell line was decreased due to AuNPs. While on the other hand expression levels of TRPC5, TRPM1, TRPM2, TRPM3, TRPM7 and TRPMV5 in SKBR3 breast cancer cell line were increased. In addition, there was no significant change in TRPC7 and TRPV7 genes. This is one of the pioneer studies in its uniqueness and it depicts a relation between ROCK pathway and expression levels of TRP channel genes. Our results demonstrated that expression levels of TRP genes can be modified following the treatment of AuNPs.

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Oral

Virology **T036**

A Computational Approach to Gene Silencing of *Cotton Leaf Curl Kokhran Virus-Lucknow* Determines Potential Host-Derived MicroRNAs in *Gossypium hirsutum*

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Cotton Leaf Curl Kokhran Virus-Lucknow (CLCuKoV-Lu) is an emerging dangerous whitefly-transmitted monopartite begomovirus which causes Cotton Leaf Curl Disease (CLCuD) that result a substantial hindrance generating high cotton earnings. The single-stranded DNA genome of CLCuKoV-Luc (2.7Kb) encodes six open reading frames; C1 (Rep), C2 (TrAP), C3 (REn), C4, V1 (CP) and V2 which have bidirectional transcription mode of replication from the LIR. The present study is designed to locate the potential attachment sites of *Gossypium hirsutum* in the genome of *CLCuKoV-Lu* applying four diverse computational algorithms for microRNA (miRNA) target prediction. A total of 78 potential mature target miRNAs were retrieved from miRBase database and were further analyzed for hybridization of *CLCuKoV-Lu* genome. Employing computational approach, miRNA-target seed pairing, multiple target positions, minimum free energy, target site accessibility, maximum complementarity, pattern recognition and minimum folding energy for attachments were considered by all algorithms. Out of 78 micoRNAs, only 2 *Gossypium hirsutum* miRNAs (ghr-miR390a, ghr-miR390b, ghr-miR390c at locus 2278, and ghr-miR7513 at locus 1350) are predicted for gene silencing of *CLCuKoV-Luc* by all four algorithms. This is the first report of identification of *Gossypium hirsutum* microRNAs which have potential to target *CLCuKoV-Lu* genes AC1, AC2, AC3 and AC4 involved in virus replication, virus pathogenicity/suppression of gene silencing, virus DNA accumulation and symptom development/ virus movement respectively. The present study concludes the first step towards development of *CLCuKoV-Lu* resistant cotton using genome engineering techniques through expression of the predicted microRNAs.

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Oral

Nanobiotechnology **T037**

Glaire Mediated Biocompatible Synthesis of Fe_3O_4 Stabilized Zirconia Coatings for Dentistry

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Zirconia has gained much attention due to its excellent mechanical, optical, thermal properties such as high refractive index, good ionic conductivity, high melting point and biocompatibility. Recently, zirconia coatings are used in myriad applications such as protective and thermal barriers, biomaterials and prosthetic coatings etc. Enhanced surfaces with increased bioactivity along with chemical and structural changes on metals and ceramic implants have been the key focus of a huge number of researchers in biomedical field. Zirconia has three polymorphous: monoclinic, tetragonal and cubic. Among these tetragonal zirconia has good mechanical and optical properties but it is not stable at ambient conditions . For stabilization of tetragonal zirconia (t-ZrO_2), metal oxides have been extensively used and stabilization using with Fe_3O_4 has already been proven but with compromising hardness. In order to increase its hardness and bioactivity different protein rich organic additives have been added into zirconia. Since glaire is a rich source of protein therefore in this research paper, acidic (pH 4) and basic (pH 10) Fe_3O_4 was doped into pre-synthesized zirconia sol with 2, 4, 6, 8 and 10wt%, whereas 2ml glaire was added in each sol during stirring. Structural properties show that formation of phase pure t-ZrO_2 with amorphous behavior in case of basic dopant at all concentrations due to presence of organic additives. Dense and low porous coatings lead to higher refractive index which increases with Fe_3O_4 concentration. Vickers micro indenter shows the higher value of hardness up to ~1500HV due to presence of glaire. These optimized coatings were applied on sterilized teeth for in vitro study. Smooth coatings have been observed under optical microscopy results. Antibacterial study of glaire added zirconia coted teeth showed high inhibition zone of 30 mm. Teeth show high corrosion and crack resistance when dipped in vinegar overnight as compare to un-coated teeth.

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Antimicrobial Agents & Chemotherapy **T038**

Himalayan (Pakistan) Actinomycetes as a Source for Novel Antimicrobial and Anticancer Agents

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The actinomycetes flora of Himalayan forests Pakistan is not yet explored, and has been selected herein for the isolation of actinobacteria. More than 93 pure bacterial cultures were isolated from the soil samples collected from different sites of Himalayan mountains range (Pakistan), and followed by biological and chemical screening. A comparison of HPLC-MS and UV absorption profiles from isolate extracts to natural product databases suggested the Himalayas isolate Streptomyces sp. PU-14-G to produce unique secondary metabolites. Scale-up fermentation, isolation and purification of the organic extract produced by this strain afforded four new naturally-occurring amino-nucleosides [puromycins B-E (1-4)]. Additionally, three other known compounds namely 5-methylthioinisine, nocardamine and ferrioxamine were also isolated and identified. Structures of the new compounds were elucidated on the basis of comprehensive 1D and 2D NMR and mass spectrometry data analysis, and by chemical methods. The isolated compounds 1-4 and 6 were biologically evaluated in comparison with puromycin (5) against a prostate cancer cell line (PC3) and non small cell lung cancer cell line (A549) for 48 hrs, and against several bacterial and fungal strains. Puromycin C (2) was the most active of our new isolated puromycin-analogues against both PC3 and A549 cell lines, however it was less active than the parent compound 5, which indicates, a replacement of an amino-group with an hydroxyl reduce the cytotoxicity as well as the antibacterial effects, and thereby extends the anticancer/antibacterial SAR for this privileged scaffold. Taxonomically, the amplified 1,377 bp 16S rRNA fragment of the *Streptomyces* sp. PU-14G had 99% identity (BLAST search) to the 16S rRNA gene sequence of Streptomyces flocculus strain NBRC 13041.

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Virology **T039**

Isolation, Chracterization and Genome Analysis of Lytic Bacteriophage TAC1: An Exceptionaly High Burst Size Dna Phage

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After serving for about a century, antibiotic are losing their effect. Multi drug resistance (MDR) and even pan drug resistance (PDR) are being reported worldwide. Development in newer antibiotics is at its lowest due to decreased output in terms of income. Bacteriophages are so for most studied alternate to antibiotics. TAC1 phage is isolated against AC5, a MDR Acinetobacter baumannii strain from tracheal aspirate. It showed narrow interspecies and broad intraspecies host range. It infects 67% Acinetobacter bumanni isolates. According to our best knowledge that's second highest activity phage reported till date. It is thermally stable and can withstand at varying pH. Two month storage at 4oC and -20^{o}C has now significant effect on its titer. It has very short latent period (10 minutes) with exceptionally high burst size (9977). It is the first DNA phage with such high burst size. It controlled in-vitro bacterial growth efficiently for entire 24 hour period at MOI 1 and MOI 100. TAC1 has double stranded DNA genome with 101.77 KB size. A total of 161 genes were predicted by phaster which comprise for 18 structural proteins (capsid 5, tail 6, base plate 2, others 5), 2 genes for cell lysis proteins (one for holing and one for endolysin), 27 genes for regulatory enzymes, 2 repressor genes and 2 recombination specific genes. Proteins with unknown functional homology in data bases were termed as hypothetical proteins. There are 141 hypothetical proteins. Phylogenetic analysis of TAC1 showed it belongs to class caudivirales and family myoviridae. The characterization and genome sequencing of TAC1 has revealed some significant results like extraordinary high burst size with short latest period and close location of holon and endolysin genes. Such information helps us to better understand the biology of phages and phage virus interaction. It will add up in scientific efforts to employ phages as therapeutic agents in future.

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Oral

Diagnostic Techniques **T040**

Cell Free DNA Quantification and Methylation Status of DCC Gene as Predictive Diagnostic Biomarkers of Lung Cancer

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The worldwide high mortality rate of lung cancer could be reduced significantly by its non-invasive early detection. The quantitative analysis of cell free circulating DNA (fcDNA) in plasma presents a potential noninvasive approach for liquid biopsy of tumor. In this study, real time PCR based approach was used to quantify fcDNA in plasma. The concentration of fcDNA was checked using hTERT (human-telomerase-reverse-transcriptase gene) as marker and amplification status of oncogene AKT2 (RAC-beta serine/threonine-protein kinase) along with the DNA methylation status of tumor"suppressor-gene DCC (Deleted in Colorectal Cancer) was assessed. The concentration of fcDNA in lung cancer patients (22.8 ng/mL) was found approximately six times above than the value detected in controls (2.8 ng/mL). Considerable variation in the AKT2 copy number was observed in lung-cancer-patients and controls ($p < 0.000$). Aberrant methylation of the DCC promoter was found to be highly specific (100%) as none of the control plasma samples showed aberrant methylation but was less sensitive (55.88%) ($p < 0.001$). The quantification of fcDNA along with determination of AKT2 amplification and DCC promoter methylation status appears promising to differentiate lung-cancer patients from healthy individuals.

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Antimicrobial Agents & Chemotherapy **T041**

Induction of Ampicillin Resistance in *Escherichia coli* and *Salmonella typhi*

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One of the major concerns for the emergence of antibiotic resistance is the extensive use and abuse of antibiotics in agriculture, poultry and in hospitals. This eventually creates selective pressure of low concentrations of antibiotics on bacteria. This stressful condition induces mutations in bacteria responsible for antibiotic resistance or just induces phenotypic adaptation/tolerance (reversible) responsible for an appreciable increase in MICs. Similarly, when bacterial infections are treated with inadequate or inappropriate dosage of antibiotics, patient may maintain low levels of antibiotics in their body fluids which may lead not only to treatment failure but also provides a chance for bacteria to develop antibiotic resistance.The aim of the study was to check (in-vitro) the outcomes of Ampicillin exposure (at sub-minimal inhibitory concentrations) to *E. coli* and *Salmonella typhi* to detect the presence of mechanism(s) contributing to the antibiotic resistance. The identification of *E. coli* and *Salmonella typhi* encountered in urine and blood specimens was carried out on the basis of conventional cultural and biochemical properties on various media, automated profile index (API) and serotyping. Antibiotic sensitivity and minimal inhibitory concentrations (MICs) of *E. coli* and *Salmonella typhi* was detected by "Disc diffusion method" and "Agar dilution method". Only Ampicillin sensitive *E. coli* and *Salmonella typhi* were subjected to induction study by exposing E. coli and *Salmonella typhi* to various sub minimal inhibitory concentrations (sub-MICs) of Ampicillin in Ampicillin incorporated Muller Hinton agar (Agar dilution method). Thereafter, these Ampicillin stress encountered strains were monitored for the formation of biofilm by scanning electron microscopy (SEM). Results indicated the development of cross resistance (resistance to unrelated antibiotics) and increase in MICs of ampicillin and enhanced biofilm production in *Salmonella typhi* and *E. coli.*

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Infectious Diseases **T042**

Detection and Molecular Characterization of Methicillin-Resistant *Staphylococcus aureus* from Table Eggs in Haripur, Pakistan

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A number of medically significant bacterial species such as*Salmonella enterica, Listeria monocytogenes,* and *Yersinia enterocolitica* have already been reported from tableeggs previously. More important is the presence of antimicrobial-resistant bacterial strains in this food source. The presentstudy was aimed at detection and characterization of *Staphylococcus aureus* from table eggs collected fromdifferent retail shops in Haripur city of Pakistan. *Staphylococci* were isolated from 300 eggs collected from December 2015 to May 2016. *S. aureus* isolates were tested for antimicrobial susceptibility using brothmicrodilution and characterized using pulsed-field gel electrophoresis (PFGE), multilocus sequence typing (MLST), staphylococcal cassette chromosome mec (SCCmec) typing, and spa typing. The presence of PantonValentine leukocidin and antimicrobial resistance genes were detected using PCR. *Staphylococci* were isolatedfrom 21.3% (64/300) of the table eggs tested. Of those, 59% (38/64) were identified as *S. aureus*, of which 33 (86.8%) were positive for mecA (MRSA, methicillin-resistant *S. aureus*). All MRSA were multidrug resistant(resistant to two or more antimicrobial classes), contained aac-aph (encoding aminoglycosides), and were pvl+.Using MLST, spa typing, and SCCmec typing, three genotypic patterns were assigned: ST8-t8645-MRSA-IV,associated with USA300; and ST772-t657-MRSA-IV and ST772-t8645-MRSA-IV, both characteristic of theBengal Bay community-associated MRSA clone. Molecular typing by PFGE revealed that the bacterial populationwas highly homogenous with only two patterns observed. This study is the first report of detection ofhuman-associated pvl^+ MRSA from table eggs. The genetic similarities of MRSA present in the eggs to that ofhumans may suggest human to poultry transmission of MRSA via contamination.

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Oral

Antimicrobial Resistance, Clinical Infectious Disease **T043**

Molecular Insight of NDM-1 -5 and -7 Producing Gram-negative Pathogens Isolated from Children

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Pediatric infections caused by New Delhi metallo-Î²-lactamase (NDM) producing bacteria constitute a serious threat to public health sectors globally. Currently, seventeen NDM variants (NDM-1 to NDM-17) have been identified. This study was designed to characterize NDM Gram-negative isolates from pediatric patients in Lahore, Pakistan. Carbapenem resistant clinical isolates (n=117) were collected from different tertiary care hospitals, Lahore. Identification and MIC was determined by Vitek 2 compact system (bioMerieux, France) and MALDI-TOF (Bruker, Germany) respectively. Carbapenamases and Metallo-Î²-lactamases (MBLs) were detected phenotypically by Modified Hodges test and double disc synergy test respectively. PCR and sequencing was performed for the detection of blaNDM and multilocus sequencing typing (MLST) was done by amplifying housekeeping genes for strain typing. Plasmids were characterized by pulsed field gel electrophoresis (PFGE) and In Gel DNA DNA hybridization. Out of 117, 37 (31.6%) were *K. pneumoniae*, 29 (24.7%) *A. baumannii* and 12 (10.2%) *E. coli*. Most of the isolates were recovered from blood (n=40) and urine cultures (n=31). Among these, 108 (92%) were MBL producers. Multiple sequence alignment revealed, 72 (61.5%) were NDM producers and among these 60, 9 and 3 were NDM-1, NDM-5 and NDM-7 respectively. Most of the NDM producing *K. pneumoniae* belong to the ST11, ST273 and ST147. However, NDM positive E. coli were mainly characterized as ST405 and ST101. Most of the blaNDM genes were located on 150kb and 280kb plasmids. MICs of the NDM positive isolates displayed high resistance against beta-lactam drugs including carbapenems and highest susceptibility against colistin. Spread of blaNDM in the clinical isolates is a matter of great concern and can become a major cause of mortality in children.

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Oral

Human Genetics **T044**

Common FTO Gene Variants and Obesity in Pakistani Population

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Obesity has become a global epidemic due to an increase in the number of obese individuals worldwide. There is little research in the field of obesity genetics in Pakistan. The aim of the current study was to analyze the association of common variants in Fat Mass and Obesity associated gene with obesity in Pakistan, find out the effect of the selected SNPs on anthropometric and biochemical traits and to observe whether these variants act synergistically. Samples from 631 subjects were taken after informed consent and were used for serum parameters and genetic analysis. Lipid profile was determined, tetra ARMS PCR was used for genotyping, allele/genotype frequencies and genescore were calculated. All FTO variants were associated with obesity, some biochemical and anthropometric measures and had higher minor allele frequencies than those reported for Asian populations previously. The risk allele of each single nucleotide polymorphism resulted in an increase in BMI in a quantitative manner. Common forms of obesity are due to a combined net effect of many variants presented in same or different genes. The more the number of risk alleles present, the higher is the risk and severity of obesity resulting from an increase in BMI.

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Oral

Biofuels **T045**

Effect of Temperature and pH on Biomass and Lipid Production of *Ulothrix*

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Algae have natural tendency to produce lipids which can be utilized as a source of biofuel. The basic aim of the study was to investigate the effect of abiotic factors on growth and lipids of *Ulothrix sp*. Two a-biotic factors i.e. Fluctuation in Temperature and pH were considered for checking their ultimate effect on both biomass and lipid production . It was observed among the different temperatures, maximum biomass was observed at 400C, while maximum lipid productivity was recorded at 350C. However in case of pH, both maximum biomass and lipid productivity was recorded at pH 7. Hence lipid productivity is significantly affected by these abiotic factors.

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Oral

Diagnostic Techniques **T046**

Optimization of Molecular Based Technique for Rapid and Sensitive Detection of *Citrus tristeza* Virus

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Citrus tristeza virus (CTV) is responsible for worldwide destruction of citrus orchards leading to huge economic losses in citrus growing countries. Pakistan is among those countries which generates huge revenue each year by means of export, but now serious issues are being faced by the cultivators due to the devastating effects of CTV. Spread of CTV is being carried out by infected root stocks and aphid species. Eradication upon early detection and use of virus-free rootstocks is the only solution to this problem, due to which the CTV detection has gained importance. Currently CTV infection is detected by using the ELISA based methods but techniques like PCR based diagnostics are also available. Several limitations such as low sensitivity, expensive instrumentation and poor expertise lead to less reliable and lower usage rate of these techniques causing difficulties in the detection and control strategies of CTV. Analyzing the current situation and danger posed by the CTV infection, we have designed our research to develop an isothermal amplification-based technique for the rapid, specific and sensitive detection of CTV. Development of other molecular variants according to their ease of use, less time requirement, increased specificity and sensitivity is also required for better results. Thus, adoption of the latest molecular based systems will make it a promising candidate for virus detection and for rapid selection of CTV-free planting material for further propagation and distribution to smallholder farmers.

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Oral

Medical Microbiology **T047**

Molecular Analysis of Metallo-Beta-Lactamase Producing Multi-Drug Resistant Clinical Isolates from a Tertiary Care Hospital

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Carbapenems are the major antimicrobials used to treat serious infections. However, their effectiveness is being compromised by the emergence of carbapenemase particularly the metallo-Î²-lactamases. The metallo-β-lactamase producing clinical isolates are very difficult to treat due to limited therapeutic option available. The study was based on analysis of 3000 samples, other than blood cultures, that were processed in Pathology laboratory, Allama Iqbal Medical College, Lahore and Department of Microbiology and Molecular Genetics from July 2015 to January 2017. Biochemical characterization and Antimicrobial Susceptibility Testing (AST) was performed. Based on AST, 143 isolates resistant to imipenem were randomly selected. Phenotypic detection tests, including: Combination Disc Synergy Testing, Modified Hodge test and Epsilometric Test for detection of MBLs were performed. Genotypic analysis of MBL positive strains was carried out by means of Multiplex PCR for OXA, TEM, SHV, IMP-1 and VIM genes. The overall frequency for carbapenem resistance was calculated to be 56%. Molecular characterization of MBLs isolates was performed to evaluate the overall distribution of OXA TEM, SHV, IMP-1 and VIM through PCR. PCR revealed 46% TEM, 34% SHV and 24% OXA among the selected isolates. 60% of the isolates were confirmed for the co-existence of OXA, TEM and SHV genes. 12.5% of the isolates were detected for acquisition of IMP-1 gene. VIM gene was found in 6% of the MBL producing isolates. The co-existence of VIM and IMP-1 genes with OXA, TEM and SHV genes was also observed. The MDR isolates are only susceptibility to few drugs, carbapenems and combination drugs including sulzone (cefoperazone/sulbactam) and tazocin (piperacillin-tazobactam). It is a need of hour to implement supplementary phenotypic diagnostic techniques for screening of the MBL with the routine testing. The molecular detection can prove to be a good tool for management of MBL associated infections.

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Oral

Medical Microbiology/Immunology **T048**

Novel Host inflammatory Responses Against Methicillin Resistant *Staphlococcus aureus* (MRSA)

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Staphylococcus aureus is a Janus faced Gram positive bacterium that causes a number of diseases ranging from mild skin abscesses to lethal infective endocarditis, necrotizing pneumonia and septic arthritis. This range of diseases caused by *S. aureus* reflects that it carries an array of virulence factors for hijacking the host immune system. Upon challenge, host immune cells recognize S. aureus and mount diverse immune responses including production of pro-inflammatory cytokines such as IL-II and TNF-I. These cytokines are important mediators of inflammation which can be detected via various immunological methods such as enzyme linked immunosorbent assay (ELISA). A number of clinical isolates as well as laboratory strains of S. aureus exhibited cross reactivity with ELISA antibodies for murine IL-1Î² and TNF-I. This cross reactivity generates exaggerated false positive signals which can be a source of discrepancy for the understanding of real immune responses against *S. aureus* infection by host immune cells. Moreover, the novel phenomenon of cell death exhibited by methicillin resistant *S. aureus* (MRSA). The clinical isolates of S. aureus had a strong ability of causing cell death in murine primary cells which was quite different from typical cell death modalities such as apoptosis, necrosis and pyroptosis. Mechanistically, this unique phenomenon of cell death induced by MRSA was independent of host inflammasome activation, Toll like Receptors (TLR) signaling, Lysozyme M (LyzM) and necroptosis. However, this kind of cell death can be protected partially after genetical deletion of Gasdermin D (GSDMD). Additionally, this form of cell death can also be rescued by using pharmacological inhibitors against Reactive Oxygen Species (ROS), autophagy and lysosomal protease which determined its complex nature.

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Oral

Antimicrobial Agents & Chemotherapy **T049**

IndoPak Medicinal Plants and their Associated Endophytes

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Ever since the creation of mankind, man has been using medicinal plants for curing different diseases thereby making them a major route towards discovering new drugs. This usage of traditional medicine has been established in the Indo Pak subcontinent for centuries. In both the countries there is a routine usage of such medicinal plants in the daily diet in the form of herbs and spices. In India, the Ayurveda system of medicine is prevalent whereas in Pakistan local physicians called Hakims make local drugs through these plants. Endophytes encompass any organisms that inhabit the internal tissues of the host plant through a considerable time of their life. There such organisms may produce a wide assortment of metabolites that may be bioactive with distinctive structures that in turn may provide advantage to the plant. Endophytes symptomlessly colonize the internal plant tissue without causing any visible change. Since they reside in an unusual habitat, this may support them to acquire the ability to mimic the host plant bioactive compounds production. The study will discuss the bioprospecting efforts related to endophytes of common therapeutic plants of Indo Pak subcontinent. Its will also discuss the reason behind using such plants for endophyte isolation and the scenario of their bioactivities.

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Oral

Biofuel Production **T050**

Biofuel Production from Algae

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Algae have natural tendency to produce lipidic bodies. The basic aim of the study was to extract lipid from *Ulothrix sp*. This strain was selected because of its maximum oil content (62%). Two a-biotic factors i.e. Fluctuation in Temperature and pH were considered for checking their ultimate effect on both biomass and lipid production . Among cultivation of *Ulothrix sp*. The study gave differential results considering comparison of both treated factors. With variation in temperature maximum biomass was observed with 400C, while maximum lipid productivity was associated with narrow but differential temperature 35ºC. However in case of pH, both maximum biomass and lipid productivity was associated with the single value. The study maily resulted in conclusion that fluctuation in pH and temperature might have both positive and negative effect on the accounted/analyzed algal spp.

Oral

Virology **T051**

Isolation and Molecular Characterization of Lytic Bacteriophage against Multidrug Resistant *Klebsiella pneumoniae* and Assessment of its Biofilm Removal Capacity

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Klebsiella pneumoniae, a Gram-negative enteric bacterium, is a common pathogen that causes human nosocomial infections including septicemia, pneumonia and urinary tract infection. Multidrug resistant *Klebsiella pneumoniae* isolates carrying extended-spectrum beta-lactamases (ESBLs) encoding plasmids are the main cause of nosocomial infections. In addition to this, *Klebsiella pneumoniae* have ability to form biofilms which provide resistance to antibiotics and immune systems. So alternative therapeutic approach is required. The present study was conducted to isolate and characterize novel lytic bacteriophage against *K. pneumoniae* and its biofilm removal capacity was also assessed. TSK1 formed clear plaque with halo which is involved in the secretion of exopolysaccharide depolymerase enzyme. The highest antibacterial activity was observed at pH 7 and at temperature 37ºC. TSK1 showed potent lytic activity only *against K. pneumoniae* strains. The reduction in bacterial growth was observed at initial 14 hours of infection, after this the growth increases rapidly but still remain lower than control. TSK1 had short adsorption rate and latent period while burst size of 60 pfu/infected cells. TSK1 belongs to *Siphoviridae* family and has a genome size of 49.74 Kb. Bioinformatic analysis of the genome suggest that TSK1 has all structural and functional genes and show close relation to the members of Siphoviridae phages (KP36 and KP1513). TSK1 significantly reduce 24h old biofilm biomass indicated by %age reduction of bacterial load i.e. 99.9% as compared to control. However, complete removal of biofilm require phage cocktails.

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Oral

Plant Microbe Interaction **T052**

Biological Control of *Fusrium* Wilt of Tomato By Indigenous *Bacillus* Strains

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Fusarium wilt is a serious vascular wilt disease of tomato crop worldwide, *Fusarium oxysporum f. sp. lycopersici* is the causative agent of this wilting disease. Uptil now there is no effective fungicide or resistant cultivar is available, this leads to the need of bacterial use as biocontrol agent. It has long life span and can survive for 7 months to 1 year either by infecting the nonspecific plants or chlamydospores in soils. This study was conducted to determine the potential of indigenous *Bacillus* strain for the biocontrol of this disease. Bacterial strains used in the study were isolated from the drought prone area and checked for their plant growth promoting ability by determining auxin production, HCN and EPS production, antibiotic sensitivity. Biocontrol potential was determined by the antifungal and antibacterial activity, production of volatile organic compound (VOC), these strains were also for the presence of lipopeptide genes of fengycin and Bacillomycin. 8 different strains showing PGPR and biocontrol potential were selected and subjected to plant growth experiments with different treatments as single use inoculum and in consortium. Observed parameters in study includes number of leaves, number of branches, shoot length, plant fresh and dry biomass. The recorded increase in shoot length, number of leaves, number of bunches was up to 1.6 folds while for number of leaves increase was up to 1.98 folds by B. aryabhattai T2S6-3 under natural conditions. Under axenic conditions the recorded increase in the plant growth was 9.09 folds, 14.5%, 27.27%, 36.3% and 100% by strain *Lysinibacillus xylanilyticus* T2S6-1, *B.cereus* T4S1-4, *B.subtilis* T4S5-4 and *B. pumilus* T1S1-1. Whereas consortium gave more promising results by enhancing the growth up to 14.8% and 1.6 folds for number of bunches and number of leaves by consortium T3 and T7. Native strains of *Bacillus* have the potential to be used as biocontrol agent for the fusarium wilt of tomato.

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Oral

Immunology **T053**

Analysis of Novel Single Nucleotide Polymorphisms in Various Exons of SYKGene in Atopic Airways Allergic Patients

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Syk a non-receptor spleen tyrosine kinase enzyme, regulates adaptive and innate immune response, plays crucial role in atopic allergies. Also mediates FcεRI signalling of mast cells and remodeling of airways. In this study, the association of different demographic and genetic variations in Syk gene, among atopic airways allergic patients were observed. To observe polymorphism in Syk, 200 blood samples of airways allergic patients and 200 healthy individuals as control were collected. DNA extraction and PCR was done, followed by SSCP analysis for banding pattern. Samples having altered mobility patterns were sequenced and results were analysed.Sequencing results confirmed polymorphism in exon 1-3 and 14'. SNP identified in exon 14 were C>T and insA, both in intronic region and synonymous mutations. An SNP in exon 1-3 was present in the intergenic region. All these polymorphisms were present in patients between 25-30 years age, having lower socio-economics and positive family history of allergies. The prevalence of airways allergies was high in males (52%), than females. Nocturnal coughing was highly prevalent (67%) among asthmatics. Family history was found significantly associated with LRP/URP, smoke and exercise.The role of modifiable and non-modifiable risk factors was clear, polymorphism in Syk gene may have association with disease severity. Analysis of SNP in Syk gene will provide new avenues, for the development of modern therapeutics to combat the escalating prevalence of atopic airway allergies.

Oral

Cancer Genetics **T054**

To Study the Influence of Metabolic Stress on Growth of Cancer Cells in 2D and 3D Cell Culture Systems

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Previous research reports have shown that metabolic stress conditions in tumor microenvironment significantly modulate the lipid metabolism of cancer cells. 3D cell culture conditions more closely resemble with the in vivo tumor microenvironment. In present research project we aims to study the effect of different metabolic stress conditions (Hypoxia and Low serum concentration) on proliferation of cancer cells both in 2D and 3D cell culture systems. HepG2 and HCT-116 cell lines were purchased from ATCC and cultivated in RPMI 1640 supplemented with 10% FBS and penicillin-streptomycin solution. Cells were maintained at 37°C with 5% CO_2. For low serum culture conditions RPMI 1640 medium was supplemented with 2%, 4% and 6% FBS. For creating hypoxia cell culture plates were sealed with paraffin film and incubated at 37°C. The generation of Multi cellular tumor spheroids (MCTs) was carried out using a liquid overlay cultivation technique. Total protein content was quantified by Bradford assay. Low serum culture conditions significantly affect the proliferation of cancer cells both in 2D and 3D cell culture systems. It also affects the growth of tumor spheroids. But no effect of hypoxia was observed on proliferation of cancer cells. We also compared the proliferation of cancer cells in 2D and 3D cell culture systems. HCT-116 cells shows reduced proliferation rate in 3D cell culture system whereas HepG2 cells shows decreased proliferation in 2D culture system. Multicellular tumor spheroid cultures more closely mimic the in vivo physiological conditions. Therefore, studying metabolic pathways of cancer cells in 3D cell culture system will provide more insights into how tumor microenvironment regulates cancer cell growth and metabolism.

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Oral

Nanobiotechnology **T055**

Effect of Surfactants and Calcination Temperature on the Magnetic Properties of Sol-Gel Synthesized Biocompatible Fe_3O_4 Nanoparticles

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Crystalline superparamagnetic Fe_3O_4 nanoparticles were prepared by simple and low cost sol-gel technique. Iron (III) chloride was used as precursor. Oleic acid, being important fatty acid for human body, is used during NPs synthesis. The amount of oleic acid is varied as 5%, 10% and 15% by volume. Shape and morphology of iron oxide nanoparticles strongly depend on the synthesis conditions and calcination temperature, which is varied from 300ºC to 900ºC, and also have strong effect on their magnetic properties. Iron oxide sol with 15% by volume show superparamagnetic behavior while sols prepared with 5% and 10% oleic acid show dia-ferromagnetic and para-ferromagnetic mix behavior. Magnetic properties of sols strongly affect the phases, structural and magnetic properties of iron oxide NPs. XRD results confirm the formation of Fe_3O_4 single phase at a temperature of 500oC with 15% oleic acid. Low oleic acid content resulted in formation of mixed iron oxide phases. Two types of NPs are observed in SEM images one with shell and one without shell with 10% oleic acid. Cubic NPs with size less than 25nm is observed with 15% oleic acid at 500ºC. Highest dielectric constant of ~107.5 (log f = 5.0) was observed for nanoparticles synthesized using 15% oleic acid and calcined at 500ºC due to high grain boundary resistance ~199.8kΩ.These NPs show superparamagnetic behavior thus making them a potential candidate for biomedical applications.

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Oral

Immunology **T056**

The Association of Vitamin D level with Smoking

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Tobacco smoking is common in Pakistan. The tobacco smoke contains nicotine, tar and many carcinogens in it. The prevalence of smoking in Pakistan is 4:1 for males and females respectively. Smoking causes Cardiovascular Diseases, COPD and various types of cancers. Smoking is associated with low levels of Vitamin D. Vitamin D is a vital nutrient obtained either from diet or by photochemical conversion of its precursor to its active circulating form. Smoking is associated with Vitamin D Deficiency (VDD) by different ways. In this cross sectional study, 24 smokers and 24 non-smokers were enrolled. The questionnaires were filled by asking about smoking habits, smoking types, number of cigarettes smoked, age, weight, height, profession, sun exposure, vitamin D intake, Vitamin D supplements, and 3-5ml blood sample was drawn from each person. Serum was obtained to measure the concentration of Vitamin D in both groups with the help of ELISA kit. 25 (OH) - Vitamin was the determinant of vitamin D status. Serum level of vitamin D was compared between smokers and non-smokers. A significant difference was found between the concentrations (ng/ml) of vitamin D in smokers and non- smokers ($p<0.05$). T-test was applied to determine this difference. No statistical difference was found between the BMI of smokers and non- smokers ($p<0.05$). All healthy smokers (n=24) were vitamin D deficient (conc. <12ng/ml) and among non- smokers (n=24), 14 were with insufficient vitamin D (12-20ng/ml) and 10 were with sufficient vitamin D (>20-150ng/ml). Many factors like age, BMI, skin tone, sun exposure time, diet and supplementation may constitute to the insufficiency of vitamin D in non-smokers. We conclude that smoking is positively associated with Vitamin D Deficiency (VDD) in healthy smokers.

Oral

Biochemistry **T057**

Adipokine Serum Visfatin Level in Pregnancy Induced Hypertension and Uncomplicated Pregnancy

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Hypertensive disorder in pregnancy is the significant disorder badly affects the maternal and fetal prognosis and lead to higher mortality and morbidity in the prenatal period. Visfatin, potentially a new adipokine emerged having high contribution in pathogenesis of Pre-eclampsia. The objective of the study is to find the level of Visfatin in pregnancy induced hypertension and normal pregnant women. This study was carried out in tertiary care hospitals, Peshawar KP Pakistan from march-october 2014. A total of 234 pregnant women (gestational age >20 weeks) were included in the study. We had patients group, Preeclampsia (PE=86), Eclampsia (E=74) and control (N=74). Blood was taken for measuring Visfatin level by Enzyme linked immunosorbent assay technique. SPSS version 19 was used for statistical analysis. Student's t test was performed to evaluate the mean differences in patients and control. Serum level of visfatin was significantly higher in pregnancy induced hypertension when compared with control (P value<0.001). Comparisons of mean value of visfatin with age group of 21-40 years, body mass index (BMI), primi parous and parity 2-4, gestational age of >36 weeks and both systolic and diastolic blood pressure were highly significant in pregnancy induced hypertension when compared with control (p value<0.001). It has been concluded that Pregnancy induced hypertensive women showed increased level of serum Visfatin than normal pregnant women.

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Oral

Environmental Microbiology **T058**

Characterization of Thermostable Amylase from *Bacillus* Species and its Potential Application in Food Industry

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Amylase is an extracellular microbial enzyme known for its wide industrial applications. In this study, *Bacillus* species was found to be express it at 4th hour till the cell go to dead phase in growth medium supplemented with 1 % starch. The enzyme exhibited optimum activity at 70°C, 1 % starch, pH 7, 0.03 % triton X-100, 0.03 % tween 20, 0.1 % surf excel, 1 % banana peel, 1 % casein and 1 % starch. Increase in starch concentration decreases its expression. Its activity was improved in the presence of Fe and Mg ions but NH_4 ions and EDTA decreased its activity. The enzyme was isolated and purified using gel filtration chromatography and DEAE chromatography. Its potential application in baking industry was checked but it needed to be improved before this isolate can be commercialized.

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Oral

Environmental Microbiology **T059**

Arsenic Bioremediation Through Indigenous Bacterial Isolates in Two Different Modes of Growth

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Uncontrolled release of industrial effluents contaminated with many different toxins, heavy metals and metalloids result in the increased environmental pollution. Consequently, concentration of many toxic metals and metalloids like arsenic is increasing continuously. Therefore, there is an urgent need to minimize toxic levels of carcinogenic arsenic. This study was aimed at reducing elevated levels of arsenic through bioremediation potentials of indigenous bacterial isolates. In this study, arsenic resistant bacteria were isolated from industrial effluents collected from Kasur, Pakistan. These bacterial isolates were screened for arsenic transformation and biofilm formation potential through qualitative assays. Bacterial isolates presenting both characteristics were studied in detail for arsenic transformation and biosorption through quantitative estimations by HPLC-ICP-MS. A total of 11 different arsenic resistant bacterial strains were isolated from industrial effluents. These bacterial isolates were identified through 16S rRNA ribotyping and were found as members of different genera such as *Exiguobacterium, Bacillus, Ochrobactrum, Achromobacter* and *Vibrio.* Among these bacterial isolates, strains PT2 and SW1 were selected for detailed study based on their potential of biofilm formation and arsenic transformation. Bacterial isolate PT2 transformed higher amount of arsenate (3.73 mM) into arsenite than that by the isolate SW1 (2.05 mM). Moreover, these isolates also displayed arsenic sorption potential within cell biomass. Therefore, these two indigenous bacterial isolates presented an important candidate for potential application in the bioremediation of arsenic.

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Plant Microbe Interaction **T060**

Auxin Producing *Rhizobacteria* as Bioremediating Agents to Minimize Chromium Toxicity in Chromium Contaminated Areas

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Heavy metals contamination in the environment is a serious threat causing health risks to the living organisms. Hence, it is very necessary to get rid of these chemical contamination to have a safe environment. In this regard, bioremediation is considered to be an economic and sustainable agricultural technique rather than use of other chemical methods to mitigate chromium toxicity from agricultural lands together with improvement in plant growth. Present work is focused on the use of plant growth promoting, chromium-resistant bacteria for enhancing the growth of *Zea mays L*. under stress conditions by using six efficient auxin-producing, chromium-resistant bacteria *Bacillus pumulis* (ALa), *Bacillus atrophaeus* (BL2), *Bacillus cereus* (AR), *Staphyllococcus lentus* (E3), T2aii and W6ii grown under various concentrations of chromium stress i.e., 0, 200, 400 and 600µg/ml by giving inoculation treatments to *Zea mays L*. seeds and different growth and biochemical parameters were recorded. Analysis of the experiment demonstrated that application of auxin-producing rhizobacterial inoculation treatment enhances the growth of plants prominently by alleviating chromium toxicity.

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Oral

Nanobiotechnology **T061**

Silver Nanoparticles Synthesis By Bacterial Metabolites and their Significant Effect against Human Pathogens

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The current study aims to provide the insight of usage of bacterial secondary metabolites in silver nanoparticle (AgNPs) synthesis and their elucidation against human pathogens. The strains *Bacillus anthracis, Escherichia coli, Exiguobacterium aurantiacumm* and *Brevundimonas diminuta* (accession number MF754137 MF754138 MF754139 and MF754140) were grown for secondary metabolite production. The nanoparticles, from secondary metabolites, were confirmed and characterized by UV-Vis spectroscopy and Transmission Electron Microscopy (TEM). The optimization study was also carried out to obtain the maximum production of silver nanoparticles from bacterial secondary metabolites. These strains exhibited the great potential as antimicrobial agents against MRSA and several other MDR bacteria (*S.aureus, E.coli, Klebsiella, Enterobacter, Salmonella, P. aeruginosa*) with minimum 10 mm to maximum 21 mm zone of inhibition. The eco-friendly approach of AgNPs synthesis will be beneficial to control antibiotics resistant bacteria at large scale.

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Oral

Metagenomics **T062**

Understanding the Effects of Soil Salinity on Rhizosphere Microbiome Assemblage

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Rhizosphere microbiome plays a significant role in the functioning of plants and provides information regarding plant physiology (osmoregulation) and development. Very limited information has been available on the microbial diversity from different saline environments. Hence in the current study, we used pyrosequencing analysis of the 16S rRNA gene to compare the composition of rhizosphere microbiome of halophytes (*Urochloa, Kochia, Salsola* and *Atriplex*) from moderate and highly saline environments (Khewra Salt Mines) with that of a non-halophyte (*Triticum*). The rhizosphere microbiome of halophytes and non-halophyte were also compared on the basis of alpha and beta diversity. Metagenomic analysis of soil indicated that *Actinobacteria* was the most dominant phylum from saline soil samples and Proteobacteria from non-saline soil samples. *Firmicutes, Acidobacteria, Bacteriodetes, Planctomycetes* and *Thaumarchaeota* were the more dominant phyla while *Cyanobacteria, Verrucomicrobia, Choroflexi, Gemmatimonadetes* and *WPS-2* were less abundant in saline and non-saline soil samples. Sequences from *Euryarchaeota, WPS-1, Ignavibacteriae, Chlamydiae* and *Nanohaloarchaeota* were identified only from the rhizosphere of halophytes. Dominant halophilic bacteria and archaea identified in this study included *Agrococcus, Armatimonadetes gp4, Halomonas, Nocardioides, Solirubrobacter, Halalkalicoccus, Haloferula and Halobacterium*. The results showed that increase in soil salinity correlated with significant differences in the alpha and beta diversity of microbial communities across saline and non-saline soil samples. This study revealed that metagenomic analysis can be used to study how changes in abiotic factors in soil (salinity) affect the microbial diversity across different soil samples.

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Environmental Microbiology **T063**

Acute and Sub-acute Toxicity of Chloroform and Idoform (DBPs) on Common Carp

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Chlorination is developing as most prevalent and adaptable process for the water and waste water treatments in Pakistan. It can improve the quality of water supplying systems and human health as well. Along with these advantages, the immediate result of potabilization is the production of disinfection by-products (DBPs), which poses serious threats to aquatic life as they were branded as carcinogenic and mutagenic. Trihalomethanes were identified in water and wastewater stream, where chlorine is used as a reactive agent. Current study was designed to investigate the potential toxic effects of two selected DBPs Chloroform (CHI_3) and Idoform ($CHCl_3$) on Common Carp (*Cyprinus carpio*) 30-60 g weighted as a model. Idohalomethanes {Idoform ($CHCl_3$)} were recently identified as disinfection by-products and were unregulated in many countries like Pakistan. Therefore, toxicity of Idoform (CHI_3) along with highly abundant trihalomethane, {Chloroform ($CHCl_3$)} was determined towards Common Carp juvenile of Rawal Lake, Islamabad. Rawal Lake is main source of drinking water supplying system for the residents of Rawalpindi and Capital Territory. Young specimens (five fishes per batch) were exposed to control and experimental basin and mortality was observed against applied doses to identified LD50 for (24, 48, 72 and 96 hrs.) of exposure duration. The effect of acute and sub-acute toxicity was measured by applying single-cell gel electrophoresis (Comet Assay) under Fluorescent Microscope (Optika- B353FL). The exposure to Chloroform and Idoform disinfectants showed clear toxic effects in fish behaviors (restlessness, dizziness and abnormal swimming). Weight of exposed specimen showed asymmetrical results after exposure as fish in some batches gain in normal weight and vice versa. Significant genotoxic effects were observed in erythrocytes of common carp after exposure to disinfectants. Substantial decrease was also observed in DNA migration under Florescent Microscope.

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Oral

Diabetes & Molecular Biology **T064**

Extra Virgin Olive Oil as a Lipid Lowering Therapy in Diabetic Patients of Lahore Region

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Studies reported that Extra virgin Olive Oil (EVOO) improves the glycemic index and lipid profile in diabetic patients but never been studied in the diabetic population of Pakistan. So the aim of this study was to check the health benefits of EVOO in the diabetic population of Pakistan. 100 Diabetic patients were given 10ml of EVVO on daily basis and blood samples were taken before the start of the therapy to check the level of Glucose, lipid profile, total cholesterol, HDL-Cholesterol, triglycerides, LFT's, HbA1c and Serum creatinine. After 3 months of therapy blood samples were taken to check the effect of EVOO on these parameters. The results are promising as EVOO containing meal lowers the level of triglycerides and overall lipid profile as compared to control group without EVOO. This is the first study of EVOO on the diabetic patients of Pakistani population which shows the improvement in the glucose level and lipid profile.

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Oral

Medical Sciences **T065**

In-silico and Proteomic Analysis for Lipid Lowering Therapy by Using *Olea europea* in the Patients of Diabetes Mellitus

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Diabetes mellitus is one of the most lethal chronic diseases which could be inherited or acquired by the deficiency in the production and release of hormone insulin by the pancreas beta cells. Among all, the Type 2 diabetes is 90% cases in Pakistan and need to be check out for its therapy. Diabetes mellitus is a disease commonly known by high level of blood sugar in the body. It is the second leading cause of blindness and kidney failure globally. Diabetics have two or four times more cardiovascular disorders and stroke than other non-diabetics persons. The use of olive oil is considered good for the health of cardiovascular system. It has been proposed that prolonged use of minor amount of the polyphenol oleocanthal obtained from olive oil along with mediterranean diet could reduce the incidence of heart diseases.According to epidemiological data the increase quantity of monounsaturated fats in the olive oil could be associated with the lower in the risk of coronary heart disease (CHD). It is suggested that the daily use of olive oil could reduce the risk of all type of causes of mortality and many other chronic diseases.Olive oil use could reduce the lipid profile in diabetes due to the beneficial effect of monounsaturated fatty acid contents or its antioxidants present in it. It reduced the inflammation by acting on the COX enzyme system in a manner that mimics to non-steroidal anti-inflammatory drugs. An excessive use of olive oil decrease the risk of mortality, Heart diseases and stroke, while monounsaturated fatty acids from the origin of animal and plant has no these kind of effects. The findings of this study would provide us the cheapest way to lower the lipid profile in the diabetes mellitus type 2.

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Oral

Medical Microbiology **T066**

Molecular Epidemiology of Extended Spectrum β-Lactamase producing Clinical Isolates from Tertiary Care Hospital, Lahore, Pakistan

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The emergence of extended-spectrum-β-lactamase (ESBLs) producing pathogens has important clinical consequence in community and especially hospital settings.This study was conducted to evaluate distribution of OXA, TEM, SHV and CTX-M producing Enterobacteriacae. From Sep 2014 to July 2017 *Enterobacteriacae* isolated from tertiary care hospital were investigated using phenotypic and molecular techniques. Antimicrobial Sensitivity Testing (AST), Double Disc Synergy Testing (DDST), Combination Drug Synergy Testing (CDST) and Epsilometric Testing (E-test) were used for the phenotypic detection. Multiplex PCR assays were used for the detection of OXA, TEM, SHV and CTX-M genes in ESBL positive strains. Sequencing results were aligned and matched to the reference sequences from NCBI BLAST.497 (49.9%) isolates were confirmed as ESBLs. A total of 995 strains of *Enterobacteriaceae* were selected on the basis of AST screening tests, out of which 497 strains were ESBLs positive. 119 Klebsiella spp and 323 strains were *Escherichia coli*. Among the phenotypic techniques used, CDST was proved to be the most sensitive compared to DDST and E-test. PCR results revealed that 7.2 % strains carry all CTX-M/OXA/TEM/SHV genes. ATCC no KX789530 and KX789531 were obtained for two *Klebsiella spp* and ATCC KX 789532 were obtained for *E. coli*. Eight TEM and six OXA amplicons were sequenced for further analysis.We report a useful multiplex PCR composed of blaCTX-M, blaSHV, blaTEM and blaOXA genes most frequently isolated from our hospitals. There is an urgent need to employ effective methods for the detection of ESBL infections in our diagnostic laboratories.

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Oral

Diagnostic Techniques/Medical Microbiology **T067**

Improved Immunoassay Using tb16.3-Echa1 Fusion Protein for Serodiagnosis of Tuberculosis

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This study focus on developing and assessing the fusion proteins with enhanced sensitivity to detect antibodies in plasma as a diagnostic method for tuberculosis. DNA fragments encoding TB16.3 and echA1 gene regions corresponding to proteins TB16.3 and echA1 from *Mycobacterium tuberculosis* were amplified through PCR. Through a series of restrictions and ligations two novel fusion constructs TB16.3- echA1 and TB16.3-tnPstS1 were produced and expressed in *Escherichia coli*. These were screened for detection of antibodies in human plasma. The individual antigens TB16.3, echA1 and tnPstS1 and the fusion protein TB16.3-tnPstS1 and TB16.3-echA1 showed sensitivities of 29%, 25.5%, 42.8%, 40.0% and 47.2%, respectively. Lower sensitivity in case of TB16.3-tnPstS1 seems to be due to the structural arrangement between the two proteins, which is likely to mask several of their epitopes. The higher sensitivity of TB16.3-echA1 appears to be due to lesser interaction between the two proteins thus allowing free availability of epitopes for binding antibodies. 64% of TB patients were found positive for either one of the two fusion proteins TB16.3-echA1 and TB16.3-tnPstS1. This study indicates that the novel fusion protein TB16.3-echA1 has a potential in serodiagnosis of TB with improved sensitivity and reliability.

MMG 2018
7-9 February 2018

Oral

Human Genetics **T068**

Effect of 50bp Ins/Del Polymorphism in Superoxide Dismutase 1 Gene on its Expressional Variability in Diabetic Cataractogenesis

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Cataract accounts for 51% prevalence throughout the world and serve as a leading cause of blindness. Epidemiological studies have revealed a higher prevalence of cataract in diabetic patients due to oxidative stress, which alters lens protein architecture and leads to the cataract formation. Superoxide dismutase1 (SOD1) is a key primary antioxidant enzyme which is involve in body's first line of defense mechanism against oxidative damage caused by reactive oxygen species. This study will investigate the role of 50bp Ins/Del polymorphism in SOD1 gene promoter region and its effect on differential expression in the pathogenesis of diabetic cataract. 50bp Ins/Del polymorphism in SOD1 gene will be genotyped by insertion/deletion polymerase chain reaction (PCR) followed by DNA sequencing. Impact of this genetic variation on SOD1 enzyme expression will be measured by enzyme linked immunosorbent assay (ELISA) and western blotting on serum and lens tissue samples respectively. Analysis of experimental data and association of genotypic analysis with SOD1 enzyme expression will be analyzed through statistical and bioinformatic tools. Expressional variability of SOD1 enzyme might use as a potential biomarker for the early detection and prognosis of cataract in diabetes mellitus. Significant genetic mutation at promoter region may be responsible for the alterations in transcriptional activity of SOD1 enzyme may serve as an easy target of pharmacogenetics in future studies of research and development.

MMG 2018
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Oral

Biofuels **T069**

Cyanobacteria and Microalgae - Role in Development of Biofuels (Lipids)

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Equilibrium between financial, agriculture and ecological progress could be made by using substitute fuel which should be economically inexpensive, technically feasible, readily obtainable and environmentally satisfactory. Biodiesel obtained from biomass of renewable raw material has all the above mentioned properties. Biodiesel is monoalkyl esters of long chain fatty acids. *Cyanobacteria* (prokaryote) and *microalgae* (eukaryote) are the photosynthetic organisms having diverse types of lipids. Different types of unicellular and filamentous cyanobacteria and microalgae lipids plays significant role in the production of biofuels. First generation fuels (from food crops) and second generation biofuels (from non-food crops) have some limitations and disadvantages due to which researchers shifted to third generation biofuels (*Cyanobacteria* and *microalgae*). 63 different *cyanobacteria* and *microalgae* isolates were obtained from different geographic regions of Pakistan including Northern areas as well. Among 63 strains, 19 were filamentous while remaining were unicellular strains on the basis of molecular methods, physiological and morphological characters. Lipids were extracted by transesterification from above mentioned strains and then quantified. Light, Fluorescent and Confocal microscope was used for the observation and detection of lipids with the help of following lipid staining dyes such as Sudan Black, Nile Red and BODIPY. FTIR analysis was also performed for the detection of lipids. Gas Chromatography also performed for the lipids analysis. The biodiesel obtained from Unicellular MFU-16, MFU-21, MFU-23, MFU-25 and MFU-39 showed higher Diesel/Biomass (%) such as 30.8%, 30.5%, 28.8%, 29.4% and 29.1% respectively. While the filamentous isolates showed MFF-1 (18.6%), MFF-12 (19%) and MFF-19 (35.6%) Diesel/Biomass (%).

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Oral

Genomics **T070**

Antimicrobial Resistance in *Staphylococus aureus* Recovered from Bovine Mastitis

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Mastitis is an important intramammary infectious disease that is usually caused by bacteria. Among them the *S. aureus* is a common pathogenic bacterium that is considered as challenge in both human and animals. Unfortunately this havoc plying bacterium acquired antibiotic resistance against many antibiotics. Development of this resistance has led to difficulty in treating mastitis. In order to control this disease it is primarily important to understand the molecular mechanisms involved in resistance development. These periodic genetic investigations will elucidate the phenotypic and genotypic relatedness of resistant strains of *S.aureus*. These findings further should consider improving overall control measures and accurate treatment strategies against mastitis.

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Antimicrobials **T071**

Isolation and Characterization of Bioactive Compounds from *Calotropis procera L.* for Treating Infections caused by Multi-Drug Resistant Pathogenic Bacteria

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Antibiotic resistance is a matter of great concern and it has finally attracted the attention of mass-media and scientists worldwide. Plant-based bioactive compounds have been known to combat infectious diseases for years. *Calotropisprocera Linn.* (Akk) is such an indigenous plant species of Pakistan having significant antimicrobial potential.To isolate and characterize bioactive compounds of *C. procera* having significant antimicrobial potential against multidrug resistant bacteria.Crude ethanolic extracts of *C. procera* were determined for their antibacterial activity against five multi-drug resistant test bacterial strains viz. *P. aeruginosa, E. coli, S. haemolyticus, Enterobacter sp.* and *S. typhi*. The bioactive compounds were identified and characterized in ethanolic extracts using the Fourier Transform Infrared (FTIR) and Electrospray Ionization Tandem Mass (ESI-MS/MS) spectrometry. The antibacterial activity of ethanolic extracts of root, fruit and leave *C. procera* was maximum against *P. aeruginosa*, however, *C. procera* displayed significant activity against other Gram negative bacteria. Comparatively, the leaves and roots extracts displayed significantly higher antibacterial activity against *P. aeruginosa* than fruit extracts of *C. procera*. The FTIR and ESI-MS/MS spectra of ethanolic plant extracts confirmed the presence of Epidigitoxigenin [m/z 404], Phytyl-iso-octyl-ether [m/z 408], Proceroleanenol-B [m/z 424], Quercetagetin-6-methyl ether 3-O-β-D-4C1-galacturonopyranoside [m/z 508], (E)-3-(4-Methoxyphenyl-2-O-β-D-4C1-glucopyranoside)-methyl propenoate [m/z 370]. Ergosterol peroxide [m/z 429.33], and 9,11-dehydroergosterol peroxide [m/z 427.32]. It was thus concluded that indigenous medicinal plants of Khairpur district could be a potential repository for lead bioactive compounds to meet the alarming situation of increasing multidrug resistance among Pseudomonads.

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Microbial Genetics **T072**

Preliminary Analysis of the Role of Lipase and Kinase in Lipid Metabolism in the *Mucor circinelloides*

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Mucor circinelloides, the oleaginous filamentous fungus has been widely used as a model organism for lipid accumulation studies. It is rich in GLA and was the first microorganism to be used to commercially produced a single cell oil: a GLA-rich oil. Although a lot of work has been done to analyze and explore many essential genes/enzymes involved in lipid accumulation but the function of lipase and kinase has not been studied at all. In this study fermentation of Mucor circinelloides was done including the sampling at different time intervals, cell dry weight determination, lipid extraction and analysis using GC. Glucose and ammonium concentration of the culture was also determined. After extraction of total RNA, transcriptional analysis of gene was done using qRT-PCR. Bioinformatical analysis of these new genes was also done to predict some important characteristics of these genes. Transcriptional analysis revealed that some genes expression was increased significantly during 24 h to 72 h of lipid accumulation while some genes were down-regulated. The results suggested that these genes are key regulators involved in lipid metabolism and may play vital role to increase oleaginicity of strain. However further studies will be done to confirm the findings and elucidate more roles of the genes in lipid metabolism.

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Plant-Microbe Interaction **T073**

Screening of Rhizospheric *Actinomycetes* for Various In-vitro and In-vivo Plant Growth Promoting (PGP) Traits and for Agroactive Compounds

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In this study 98 rhizospheric *actinomycetes* were isolated from different wheat and tomato fields, Punjab, Pakistan. About 30% of the isolates screened were found to be the promising PGP rhizobacteria (PGPRs), which exhibited maximum genetic similarity (up to 98-99%) with different species of the genus *Streptomyces* by using16S rRNA gene sequencing. The most active indole acetic acid (IAA) producer *Streptomyces nobilis* WA-3, *Streptomyces kunmingenesis* WC-3, and Streptomyces enissocaesilis TA-3 produce 79.5, 79.23, and 69.26 µg/ml IAA respectively at 500 µg/ml L-tryptophan. The highest concentration of soluble phosphate was produced by Streptomyces sp. WA-1 (72.13 mg/100 ml) and S. djakartensis TB-4 (70.36 mg/100 ml). All rhizobacterial isolates were positive for siderophore, ammonia, and hydrogen cyanide production. Strain S. mutabilis WD-3 showed highest concentration of ACC-deaminase (1.9 mmol /l). For in-vivo screening, seed germination, and plant growth experiment were conducted by inoculating wheat (Triticum aestivum) seeds with the six selected isolates. Significant increases in shoot length were observed with *S. nobilis* WA-3 (65%), increased root length was recorded in case of *S. nobilis* WA-3 (81%) as compared to water treated control plants. Maximum increases in plant fresh weight were recorded with *S. nobilis* WA-3 (84%), increased plant dry weight was recorded in case of *S. nobilis* WA-3 (85%) as compared to water treated control plants. In case of number of leaves, significant increase was recorded with *S. nobilis* WA-3 (27%) and significant increase in case of number of roots were recorded in case of strain *S. nobilis* WA-3 (30%) as compared to control plants. Over all the study revealed that these rhizospheric PGP *Streptomyces* are good candidates to be developed as bioferlizers for growth promotion and yield enhancement in wheat crop and can be exploited for the commercial production of different agro-active compounds.

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Human Genetics (Forensics) **T074**

Variable Number of Tandem Repeats (VNTR) of Criminal Gene Monoamine Oxidase A (MAO-A) in Pakistani Population

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Behavioral geneticists are showing interest in genes to reveal secrets of human behavior. Aggression is one of the most important traits of the human behavior.Monoamine oxidases are the mitochondrial enzymes that catalyze the oxidative deamination of several biogenic amines. Monoamine oxidase A (MAO-A), that has earned the name "warrior gene". Within the Promoter of monoamine oxidase A gene there is region of DNA that contain variable number of tandem repeats (VNTR). The purpose of the current study was to investigate the association of VNTR aggressive samples and controls. The study population was according to Hardy Weinberg Equilibrium.We collected 282 samples (aggressive samples, diabetics, coronary artery disease CAD patients and controls), isolated their DNA and genotyped for VNTR. Results showed the highest frequency of 3.5 repeat in promoter region. In conclusion, variable number of tandem repeats can serves as potential genetic marker for determining the tendency of aggression in population.

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Environmental Biotechnology & Bioenergy **T075**

Comparative Study of Mutagenic and Non-Mutagenic Bacteria in Methanogenisis to Yield Methane from Solid Waste

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Currently safe disposal of solid waste is a burning iusse throught the globe and millions of tons of solid waste accumulation and its disposal has become a tough task due to paucity of space and technology and the public also opposing to dump in their open places. Western countries are practis(ed)ing incineration and sanitary landfilling methods to produce energy by utilizing this solid waste and recent past they came to realize that these methods are also not ecofriendly. The method biomethanation is a promising technoogy to solve solid waste problem in a meaningful way without alterning the health of the environment. Several researchers using wildtype mathanogenes in production of gas which is less in quantity and quality and also incomplete. In this direction an effort has been made for complete digestion of solidwaste to produce maximum amout of methane with high calorific value by using mutagenic bacteria. In this method antibiotics are used as mutagens. During anaerobic digestion parameters like temperature, pH and nutrient availability are also altered for maximum production of methane gas. The findings are very encouraging with mutagenic bactieria when compared with non-mutagenic bacteria.

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Human Genetics **T076**

Genetic Etiology of Coronary Artery Disease Considering EDN1 Gene Variant rs5370

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The present study aimed to investigate the influence of rs5370 polymorphism on the risk of coronary artery disease in a small group of the Pakistani population. A case-control study, involving 320 blood samples, was completed using the allele specific PCR based strategy. Groups were defined on the basis of presence or absence of coronary artery disease (CAD). The analysis was based on the Hardy-Weinberg equilibrium estimation, chi square test and the estimation of odds ratio.The SNP rs5370 was strongly associated with CAD in the population ($p<0.01$). T allele was found to enhance the risk value of CAD. GG genotype showed the protective role (OR: 0.146, 95% CI 0.088-0.247). Discordantly, TT genotype increased the risk for 8.818 times for onset of CAD (OR: 8.818, 95% CI 5.270-14.933). On the basis of the present finding, it can be concluded that SNP rs5370 is associated with CAD in the local population of Pakistan. It indicates the importance of EDN1 gene in association with the on-set of coronary artery disease.

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Bioinformatics & Pharmaceutical Microbiology **T077**

Antibacterial Screening and *In Silico* Validation of 1, 2, 4-Triazole-3-Thione Derivatives against MDR *Escherichia coli*

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The emergence of severe forms of drug resistant bacteria calls for constant efforts to find alternate, broadly effective chemical entities. In the current study, a series of glucopyranosyl-conjugated 1, 2, 4-triazole-3-thione derivatives (n=20) were tested against the clinical isolates of multidrug resistant (MDR) strains of *Escherichia coli*. After the in vitro screening the chemicals were subjected to molecular docking analysis to determine their probable site of action within the drug resistance machinery. Of all the tested analogs, it was found that pyridyl substituted 1, 2, 4-triazole-3-thione derivatives were effective in decreasing the bacterial growth at the lowest possible concentrations (MIC, 30µg/ml). Moreover, it was found that all the derivatives had the maximum binding affinity with the Outer membrane protein (OmpC), the Gram- negative porin majorly involved in drug efflux. The study, therefore, helps in signifying the potential of the 1, 2, 4-triazole-3-thione nucleus to serve as a lead compound for drug development against MDR strains of the bacteria, targeting specifically the OmpC protein.

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Industrial Microbiology **T078**

A Mono Designed Medium for the Simultaneous Production of Carboxy Methyl Cellulase (CMCase) and Amylase by an Indigenously Isolated Thermophilic Strain *Bacillus licheniformis* TLW-3

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With global population predicted to hit 9 billion people by 2050, the need for additional requirements of agriculture (and food) is eminent. Also, the unpredicted increase in world population has resulted in generation of million tons of agricultural wastes. Biotechnological process for production of green chemicals, namely enzymes that may provided the best utilization of these otherwise unutilized wastes. Microbial enzymes have wide-based applications in industrial to household sectors, biotechnological, medicinal and basic research fields and thus, hold the major share in the global enzyme market. Production of multi enzymes out of a single fermentation process helps in reduce the overall production cost (with particular reference to industrial application of the enzymes. For efficient-simultaneous production of multi-enzymes in a single fermentation, bioprocesses with well-established bioengineering are needed to be developed. In this regard *Bacillus licheniformis* TLW-3 strain (isolated from local hot oven ash) was screened for its ability to produce two industrially important enzymes i.e. CMCase and amylase. This strain was grown in plain production media (PM), PM containing 1% CMC, PM supplemented with 1% starch and production medium containing 1% CMC + 1% starch. After fermentation broth (at different interval) was assayed separately for cellulase and amylase activity by dinitrosalicylic acid method. Growth of the isolate was also estimated. Accordingly, the medium containing both the substrate supports the simultaneous production of CMCase and amylase was observed in the same fermentation unit. Previously (elsewhere), work was done on the co-production of CMCase and amylase by fungi. However, to the best of our knowledge and research data this is the first ever report of its kind reporting the single medium for co-production of CMCase and amylase by bacteria (indigenously isolated).

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Food Microbiology **T079**

Bio-Degradation of Aflatoxin B1 and B2 in Stored Maize using Plant Extracts

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Maize is world's leading cereal crop with added importance for countries like Pakistan where rapidly increasing population has already out stripped the available food supplies. Maize is a good substrate for mold infection and production of dangerous mycotoxins. This study was designed to develop cost effective and an eco-friendly strategy for detoxification of mycotoxins by using medicinal plants extracts. During a survey of maize store houses in 15 districts belonging to three agro-ecological zones of Punjab, Pakistan a total of six fungal genera were isolated from collected samples with highest percentage frequency of occurrence for genera *Aspergillus*, followed by *Penicillium* and *Fusarium*. Aflatoxin B1 and B2 was found 97.3% and 78.9% respectively in the samples. Whereas aflatoxin G1, G2 and ochratoxin A were not detected in any of the sample. In Vitro and In Vivo detoxification assays were performed under optimized temperature, pH and incubation period using aqueous extracts of medicinal plants. Among ten tested plants, *E. citriodora* leaves, T. ammi seeds, *O. basilicum*, A. nilotica and Mentha arvensis leaves extract significantly degraded AFB1 and AFB2 under optimized conditions. Data recovered from TLC and HPLC revealed that treated aflatoxins were degraded into a number of other compounds with properties different from parent toxins. So, the presence of these degraded products was further confirmed by LCMS/MS studies. Furthermore, biological toxicity of degraded products were tested by brine shrimps (*Artemia salina*) bioassay in which least shrimp's mortality was recorded with toxins treated with *E. citriodora* leaves followed by *T. ammi* seeds extract.

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Environmental Microbiology **T080**

New Record of Rust Fungi from Northern Areas of Pakistan and Azad Jammu & Kashmir

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The *Urediniomycotina* are a very large and diverse group of fungi making one of the three subphyla of basidiomycota. More than 8000 species of this group have been described but the majority of these belong to the order Uredinales. Approximately 400 species in 23 genera on app 350 plant host species have been reported from Pakistan. The main objective of this research work is to characterize, enlist and upgrade existing descriptions and illustrations of Uredinales of Pakistan. Plants infected with rust fungi were collected from northern areas of Pakistan and Azad Kashmir.Host plants were identified. Infected portions were observed under stereomicroscope. Handmade sections were cut of infected portions. Scratched material was mounted in Lactophenol to prepare semi-permanent slides. Slides were observed under microscope. Illustrations of spores were made under Lucida camera while transverse sections of sori weremicro photographed. Rust species were identified and characterized using available data from published papers and books. Two species were found as new records from Pakistan: *Puccinia cuneata* on *Geranium essence* and *Puccinia nysalindica*on *Brachiaria ramosa*. Both these species have been previously reported from Japan but they are newly reported from Pakistan.As rust fungi are host specific and have narrow host ranges so this knowledge could be utilized as biocontrol agents on certain noxious weeds on economically important plants in future rather than using hazardous herbicides.

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Human Genetics **T081**

Gene Polymorphisms of DISC1 are Associated with Schizophrenia: A Family Based Study

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Schizophrenia is a chronic mental condition with worldwide prevalence of 1% and no gender differentiation. Schizophrenia is a polygenic multifactorial disorder in which heritability content is estimated up to 80% and several genes involved in neuronal development and function are found to be altered in this condition. DISC1 is a gene present on chromosome 1 that plays important role in neurogenesis. Single nucleotide polymorphisms in DISC1 are found associated with schizophrenia. For the present study 10 families having at least one patient of schizophrenia were identified and a total of 76 individuals were evaluated for four DISC1 polymorphisms (rs1417584, rs1954175, rs821616 and rs113012343). Allele specific PCR and PCR-RFLP were done to analyze the four SNPs. CTAG haplotype for DISC1 polymorphisms spanning 353Kb on human genome was found associated with disease onset Transmission / non transmission ratio of 2.519. While the co inheritance of 80% was found in case of first three SNPs (325kb region) based on linkage disequilibrium plot. It is concluded that in Pakistani population DISC1 plays important role in development of schizophrenia because of its interactions in neuronal development, stability, regulation and maintenance.

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Biochemistry **T082**

Role of RING Finger Protein 10 (Rnf10) during RA Induced Neuronal Differentiation of Stem Cells

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Ring finger protein10 is recently reported to be involved in neuronal differentiation, development and proliferation. In current study, mRNA and protein expression level of Rnf10 was found to be increased significantly upon the retinoic acid-induced neuronal differentiation of P19 cells. Silencing of Rnf10 gene by RNA interference has significantly impaired neuronal differentiation of P19 cells as indicated by attenuated expression of neuronal markers. Cell cycle profiling showed that Rnf10-knockdown cells were unable to establish cell cycle arrest after RA treatment. In agreement with flow cytometry analysis, elevated cell proliferation was observed after RA induction in Rnf10 knockdown cells as determined by a BrdU incorporation assay. Moreover, the mRNA levels and protein expression of p21 and p27 were also increased upon RA induction. However, Rnf10 silencing only resulted in a reduction of p21 expression, while p27 and p57 expression remained the same, indicating that Rnf10 may regulate cell cycle exit through the p21 pathway. Ectopic expression of p21 partially rescued the effect of Rnf10 depletion on the neuronal differentiation of P19 cells. In conclusion, these results indicated that elevated Rnf10 expression upon RA induction is necessary for the positive regulation of cyclin kinase inhibitor p21 expression, which leads to cell cycle arrest and is critical for neuronal differentiation.

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Environmental Microbiology/Biochemistry **T083**

Heavy Metal Resistant *Pichia hampshirensis* 4Aer, Isolated from Industrial Effluent and Its Potential Use in Cadmium Removal from Wastewater

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Pichia hampshirensis 4Aer is first ever used yeast for the bioremediation of environmental cadmium (Cd^{+2}) which could maximally remove 22 mM/g and 28 mM/g Cd^{+2} from aqueous medium at lab and large scales, respectively. The biosorption was found to be the function of temperature, pH of solution, initial Cd^{+2} concentration and biomass dosage. Competitive biosorption was investigated in binary and multi-metal system which indicated the decrease in Cd^{+2} biosorption with increasing the competitive metal ions attributed to their higher electronegativity and larger radius. FTIR analysis revealed the active participation of amide and carbonyl moieties in Cd^{+2} adsorption confirmed by EDX analysis. Electron micrographs summoned further surface adsorption and increased cell size due to intracellular Cd^{+2} accumulation. Cd^{+2} was the causative agent of some metal binding proteins as well as an prodigious increase in glutathione and other non-protein thiols levels which is the crucial for the yeast to thrive oxidative stress generated by Cd^{+2}. Our experimental data was consistent with Langmuir as well as Freundlich isotherm models. Thermodynamic and kinetic studies revealed the Cd^{+2} biosorption as exothermic, spontaneous and feasible process. The yeast obeyed pseudo second order kinetic model which make it an effective biosorbent for Cd^{+2}. High bioremediation potential and spontaneity and feasibility of the process make *P. hampshirensis* 4Aer an impending foundation for green chemistry to exterminate environmental Cd^{+2}.

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Public Health **T084**

Public Health Significance and Vaccination of Poultry against *Salmonella*

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Salmonellosis is the most prevalent food-borne zoonotic disease throughout the world. Food animals serve as reservoirs for *Salmonella* infections (Nontyphoid). In poultry, salmonella is host-specific causing fowl typhoid and pullorum diseases that result in greater economic losses throughout the world. Contaminated foods are the most important source for human infection with non-host adapted *Salmonella*. Food items including raw, under and over cooked meat and egg products pose a great threat to public health. Regarding critical analysis in detecting the *Salmonella* serotypes in Humans (infected) and food, it was made easy in concluding that 20% *Salmonella* cases in humans are being caused by *Salmonella* from swine origin and 60 % to 65 % of *Salmonella* from poultry meat and eggs (*S. enteritidis*). Efficient management at the farm regarding sanitary measures is a key to control *Salmonella* infection. These measures include avoiding the hatching eggs, day-old chicks, feed, water, rodents and environmental contamination being the sources of infection in the healthy flock. For that purpose, careful identification of *Salmonella* survival sites, efficiency of disinfectants to reduce the *Salmonella* count are mandatory. Several precautionary measures are now being used but vaccination id the most common because it avoids contamination of poultry products and by-products and also prevents diseases in humans. Vaccines against *Salmonella* can reduce public health risk to greater extent by reducing the *Salmonella* to colonize and disruption of organs including disruption of reproductive tissues, and by reducing the fecal shedding and environmental contamination.

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Oral

Biofilms **T085**

Microbiologically Influenced Corrosion (MIC) Behaviour of Cu-Ni (70-30) Alloy in the Presence of *Bacillus subtilis* and *Pseudomonas aeruginosa*

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Microbiologically influenced corrosion (MIC) behavior of copper-nickel 70-30 (Cu-Ni 70-30) in the presence of bacterial biofilms produced by *Bacillus subtilis* strain S1X and *Pseudomonas aeruginosa* strain ZK was studied in minimal salt medium containing NaCl (1.5%) as corrosive agent. MIC was investigated using Tafel polarization, electrochemical impedance spectroscopy (EIS), scanning electron microscopy-energy dispersive spectrum analysis (SEM-EDAX), atomic force microscopy (AFM), Fourier transform infrared spectroscopy (FTIR) and weight loss studies. Compared to control (uninoculated) system, the bacterial inoculated systems showed decreased values for corrosion rate and corrosion current. Electrochemical data showed corrosion inhibition of Cu-Ni 70-30 in the presence of bacteria through the formation of a protective layer on metal surface. SEM-EDAX and weight loss studies also supported this observation. AFM and FTIR showed the formation of thick biofilm on Cu-Ni 70-30. A decrease in pH values was measured for bacterial inoculated systems which is an indication for the production of some acidic compounds

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Oral

Biotechnology **T086**

Qualitative Screening of Different Fungal Strains Potentially Strong Toward Laccase Biosynthesis, Isolated from Different Locations of Pakistan

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In present study, *Pleurotus ostreatus* (jacq.) *P. Kumm.*, *Ganoderma lucidum* (Curtis) *P. Karst*., *Ganoderma ahmadii Steyaert*, *Ganoderma applanatum Conk*., *Ganoderma australe* (Fr.) *Pat*, *Ganoderma colossus* (Fr.) *C. F. Baker*, *Ganoderma flexipes Pat.*, *Ganoderma resinaceum Bourd*., *Ganoderma tornatum* (Persoon) *Bresadola*, *Coriolus hirsutus* (Wulfen) *Pilat*, *Coriolus proteus* (Berk.) *Dutta Roy*, *Coriolus pubescens* (Trametes pubescens (Schum: Fr.) Pil), *Coriolus tephroleucus* (Trametes tephroleuca Berk.), *Coriolus versicolor* (Fr. ex Fr.) *Quel*, *Trametes insularis Murr*., *Coriolus zonatus* (Nees) QuÃ©l, *Fomes fomentarius* (L. ex. Fr.) *Fr.*, *Fomes scruposus* (Fr.) *G. H. Cunn*., *Fomitopsis semitostus* (Berk.) *Ryv*., *Fomes lividus* (Kalchbr.) *Sacc.*, *Fomes linteus* (Berk. and Curt.), *Phellinus allardii* (Bres.) *Ahmad*, *Phellinus badius* (Berk. Cke.) *Cunn*., *Phellinus callimorphus* (Leveille) *Ryvarden*, *Phellinus caryophylli* (Racib.) *G. Cunn.*, *Phellinus pini* (Thore: Fr.) *Ames*, *Phellinus torulosus* (Pers.) *Boud. Galz*., *Poria ravenalae* (Berk. and Br.) *Cooke*, *Poria versipora* (Pers.) *Rom.*, *Poria paradoxa* (Schizopora paradoxa (Schrad.:Fr.) Donk,), *Poria latemarginata* (Durieu & Mont.) *Cooke*, *Heterobasidion insulare* (Murrill) *Ryvarden sensu lato*, *Schizophyllum commune* (Fr.), *Schizophyllum radiatum* (Sw.) *Fr. Daldinia sp*. (Ces.) De not., *Xylaria sp.* (Pres.) *Grev*., were collected, isolated, identified and then screened qualitatively for their laccase activity.

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Microbiology **T087**

Relationship of Cell Surface Hydrophobicity with Biofilm Formation and Growth Rate -A study on *P. aeruginosa, S. aureus* and *E. coli*

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The present study describes the association of these bacteria in planktonic and biofilm lifestyles. A total of seventeen (17) isolates each of *P. aeruginosa, S. aureus* and *E. coli* were studied. These isolates were recovered from different food items and identified on the basis of growth characters on differential and selective media and biochemical characteristics. The results showed that *E. coli* dominated the pre-biofilm stage, at which *E. coli* population was much higher than *P. aeruginosa* and *S. aureus*. At this stage, *S. aureus* also dominated *P. aeruginosa*. Moreover, *E. coli* adopted biofilm life much before *S. aureus* and *P. aeruginosa*. However, after adopting biofilm life style, slowly and gradually, *P. aeruginosa* dominated the consortia and dispersed other stake holders. It was noticed that subject isolates of *P. aeruginosa* produces cis-2-decanoic acid to disperse or inhibit *S. aureus* and *E. coli* biofilms. Gas-chromatography and Mass spectrometry results showed that cis-2-decanoic was higher in co-culture condition and increased at late log-phase or at stationary phase. Although a majority of the *S. aureus* were unable to compete with *P. aeruginosa*, however, a minor population competed, survived and persisted in biofilm consortia as small colony variants. The survivors showed higher expression of sigB and sarA genes. *P. aeruginosa* showed comparatively higher hydrophobic surface properties. Comparative analysis showed that cell surface hydrophobicity, growth rate, biofilm formation and small colony variants (SCVs) are correlated. Subject isolates of *P. aeruginosa* and *S. aureus* with hydrophobic surface properties were comparatively slow growers, showed late but strong biofilm formation, and were difficult to disperse and switch to SCVs.

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Food Microbiology **T088**

Occurrence of Gram Negative Pathogenic Bacteria in Ready to Eat Leafy Vegetables from Lahore

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Leafy green vegetables are good source of nutrition, for this reason they are extensively used in variety of food. Due to nutritional contents, leafy greens are prone to be contaminated with bacterial pathogens during cultivation, harvesting and storage. Many outbreaks that has caused a vast morbidity and mortality have been reported yet in which leafy greens were the source of pathogen. For this reason, three extensively used leafy greens coriander, mint and lettuce were selected and sampled form 16 sites from Lahore in 4 groups urban, suburban, suburb and supermarkets. A sum of 75 samples of these three leafy greens were analysed for aerobic plate count and coliform count. Based on colony morphology and lactose fermentation gram negative bacteria were isolated and purified for identification. Emerging trend of antibiotic resistance was also taken into count and antibiotic resistance was analysed by Kirby Bauer Method. The APC was ranged in 2.0×107-1.1×109 CFU/g. Coliform count was 66.0×105 to 4.8×107. Sixteen types of gram negative bacteria were identified after purification. All the selected strains were Multiple antibiotic resistant. The highest multi drug resistant were observed in case of *Proteus* and *Weeksella* species.

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Nanobiotechnology **T089**

In vitro Antibiofilm and Antiadhesion Effects of Magnesium Oxide Nanoparticles against Antibiotic Resistant Bacteria

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The aim of the current investigation was to determine antibacterial and antibiofilm potential of magnesium oxide (MgO) nanoparticles against antibiotic resistant clinical strains of bacteria. MgO nanoparticles were synthesized by wet chemical method, and were further characterized by scanning electron microscopy (SEM) and energy dispersive X-ray (EDX). Antibacterial activity was determined by broth microdilution and agar diffusion method. Bradford method was used to assess cellular protein leakage as a result of loss of membrane integrity. Microtiter plate assay following crystal violet staining was employed to determine the effect of MgO nanoparticles on biofilm formation and established biofilms removal. The results indicated that MIC values ranged between 125-500 µgml-1. Moreover, MgO nanoparticles treatment accelerated rate of membrane disruption, measured as a function of leakage of cellular proteins. Leakage of cellular protein content was more among Gram negative bacteria. Cell adherence assay indicated 25.3-49.8 % inhibition of bacterial attachment to plastic surfaces. According to static biofilm method MgO nanoparticles reduced biofilm formation potential up to 31 to 82.9 % in a time dependent manner. Moreover, nanoparticles also significantly ($p < 0.05$) reduced the biofilm biomass of 48, 72, 96 and 120 h old biofilms. Cytotoxicity experiments using neutral red assay revealed that MgO nanoparticles were non-toxic to HeLa cells at the concentrations of 15-120 μgml^{-1}. The data provides in vitro scientific evidence to use MgO nanoparticles effectively and safely as antibiofilm agent to inhibit adhesion, biofilm formation and removal of established biofilms of multidrug resistant bacteria.

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Virology **T090**

A Bacteriophage RS with Segmented Double Stranded DNA Genome RS against Multi Drug Resistant *Pseudomonas aeruginosa* has very Unique Characteristics

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Antimicrobial resistance is a global threat, so exploring new and alternate antimicrobials is the need of the day. Among different alternatives, bacteriophages are a potential candidate. The bacteriophage RS is isolated from River Ravi, Lahore, Pakistan against a well characterized strain PA-1 of *Pseudomonas aeruginosa* (Accession Number MG763232). The phage isolation and characterization is performed through well-established methods. The genome is extracted by modified Phenol Chloroform Iso-amyl Alcohol (PCI) method. The genome is sequenced by Illumina Next Generation sequencing. Assembly performed by Abyss, Velvet & CLC workbench, while annotation was done by RAST server and Phaster online databases. RS produce circular transparent plaques having a size of 3-4mm in diameter, while showed stability at 4, 25, 37, 45 & 60^{o}C temperature and 5-9 pH. A unique feature of plaque size enhancement is observed by incubating at 45 and 60^{o}C. The RS inhibited the complete growth of its host till 10 hours, while very few cells started growing after 10 hours till 24 hours. The RS showed tendency to infect majority of tested clinical *P. aeruginosa* isolates. The RS phage has a unique segmented DNA genome of 93 kbps and 63kpbs respectively, which is not reported for DNA viruses yet. The gene annotation identified a number of proteins with known and unknown functions in the genome of RS phage. The traditional research combined with advance molecular biology tools might lead to utilize the potential of RS phage for treating infections with antibiotic resistant superbugs.

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Public Health **T091**

Health, Safety and Environmental Status at a Small Scale Knitting Industry in Pakistan

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Antimicrobials **T092**

Comparative Efficacy of Fungicides, Botanical Extracts and Antagonistic Microorganisms against *Phytophthora Palmivora* causing Leaf Spots of Chinese Fan Palm

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Effectiveness of fungicides, methanolic botanical extracts and antagonists againstinvasive *P. palmivora* inciting leaf spots of Chinese fan Palm was carried out. Best fungicide inrestraining the development of the organism, in plunging request were Range, Difenconazole,Acrobat MZ, Precure and Thiophenate methyle as they caused 100, 100, 97.33, 92 and 90.88percent decrease over the control in mycelial development of *P. palmivora*. While, Propmeb,Sulfur and Mancozeb were slightest powerful in hindering the mycelial development of parasiteas they caused 40.73, 34.78 and 33.22 percent lessening in mycelial development. Bestmethanolic plant extricates in restraining the development of the organism, in plummeting request were Moringa Leaf Concentrate, Neem Leaf Concentrate, Akk Leaf Concentrate, Garlic Concentrate and Datura Leaf Concentrate as they caused 87.2, 85.6, 76.4, 76 and 69.8 percentdiminishment over the control in mycelial development *P. palmivora*, separately. There wascontinuous trend of reduction in mycellial growth with increase in fungicide and methanollicplant extract concentration. All the antagonistic microorganisms reduced the mycelial growth of*P. palmivora* significantly. *Pseudomonas* fluorescence has produced largest inhibition zone (5.5 mm) followed by *Penicillium* spp.which has produced (5 mm) inhibition zone. *Basillus fortis*was the third most effective antagonist against *P. palmivora* with 4.5 mm inhibition zone. *Aspergilus nigar* proved to be least effective antagonistic microorganisms which has produced 2 mm inhibition zone as compared to other antagonists.

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Plant-Microbe Interaction **T093**

Halotolerant Rhizobacteria: Beneficial Plant Metabolites and Growth Enhancement of *Triticum aestivum L.* in Salt Amended Soils

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Salt-tolerant strains of *Enterobacter asburiae*, *Bacillus thuringiensis*, *Moraxella pluranimalium* and *Pseudomonas stutzeri* were evaluated for their ability to alleviate salt stress of wheat (*Triticum aestivum L.*) seedlings. 1-Aminocyclopropane-1-carboxylate deaminase activity of *P. stutzeri* S-80 and *B. thuringiensis* S-26 was 190 and 183 nmol h^{-1}, respectively. Maximum levels of auxin were recorded with *P. stutzeri* S-80 (107 µg ml^{-1}) and *E. asburiae* S-24 (143 µg ml^{-1}) under normal and salt-stressed conditions (0.25M NaCl), respectively, with 500 µg ml^{-1} L-tryptophan. Auxin response mediated by rhizobacteria was also demonstrated by microscopically assaying the transgenic auxin-responsive reporter DR5::GUS expression tomato (*Solanum lycopersicum* L. cv. MicroTom). In pot trials, seedlings fresh and dry biomass witnessed highly significant improvements of 1- and 2.2-folds, respectively, with *M. pluranimalium* S-29 (at 100 mM NaCl) and *E. asburiae* S-24 (150 mM NaCl), over control. At final harvest, maximum increase in number of tillers (up to 94%) and seed weight (up to 40%) was recorded with E. asburiae S-24 and *M. pluranimalium* S-29 at 200 mM salt stress. In conclusion, newly isolated strains of *M. pluranimalium* S-29, *E. asburiae* S-24 and *P. stutzeri* S-80 enhanced the growth of *T. aestivum* L. by mitigating the salt stress of plants.

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Environmental Microbiology **T094**

Impact of Climate Change Plant Diseases Distribution in Punjab, Pakistan

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The review of investigation on distribution of some economically important diseases on sunflower, bitter gourd, sesame seed, and peanut is compiled and revisited. Shifting of weather and changing intensity of environmental gradient is creating serious threats for farm economy in Pakistan. Very little information is available on effect of change of temperature, rain intensity and frequency resulting in weather shift. A study on epidemiology of major diseases caused by Macrophominaphaseolina charcoal rot, Myrothecium leaf spot of Bitter gourd, fussarium rot of Gladioulus as test diseases. The detailed surveys were conducted in various agro ecological zones of Punjab province at different intervals from 2000 to 2014. Field survey data was collected on prescribed visual rating scale and socio economic information from stakeholders was collected on a structured questionnaire. The collected information was compared with previous crop and disease statistics of Punjab agriculture department and our Phd thesis projects. The impact of climate change was observed not only in the change in disease index but also as variations in morphology and physiology of pathogenic fungi. Areas of Dera Ghazi Khan observed as new areas for cultivation of crops and vegetables but it needs effective farm market network.

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Antimicrobials **T095**

Actinomycetes Flora of Pakistan: A Valuable Resource of Novel Bioactive Molecules

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The antibiotics resistance especially the emergence of multidrug resistant (MDR) and extensively drug resistant (XDR) bacterial pathogens necessitates the continuous screening and search for new members from various sources. Most of the commercially available antibiotics, antifungal, and anticancer agents have their origin in microbial natural products and are the secondary metabolites of various bacterial and fungal species. Pakistan is rich in biodiversity and there are diverse ecosystems in different regions of the country, including forests, saline lands, deserts, lakes and marine coasts. The interesting fact is most of these ecological niches are still unexplored and their microbial diversity is untapped with reference to the screening for new antibiotics and other chemotherapeutics. The actinomycetes are gram positive filamentous bacteria having high GC content in their genome and are the leading producers of most of the antibiotics and chemotherapeutics. In our search for novel bioactive molecules we have isolated a large number of actinomycetes strains from different ecological niches in Pakistan. The isolated strains have been identified by microbiological, biochemical and genetic approaches (16S rRNA gene sequencing) etc. The laboratory scale cultivation of the selected strains and subsequent solvent extraction, purification and structure elucidation of the active molecules by mass spectrometry and NMR spectroscopy, yielded commercially useful known and new antibiotics and anticancer agents. Overall the study revealed that the actinomycetes flora of Pakistan is an untapped source and harbors the immense potential to produce novel bioactive molecules, and should continuously be explored to combat the emerging antibiotics resistance and to fulfill the increasing demand of new chemotherapeutics.

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Plant-Microbe Interaction **T096**

Indole-3-acetic acid (IAA) Production by Rhizobacteria was Associated with Improved Growth and Yield of *Triticum aestivum* L. under Drought Stress

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Drought tolerant rhizobacteria were evaluated for their potential to mitigate the water stress of wheat (*Triticum aestivum* L.). Taxonomic status of the strains isolated from semi-arid areas was confirmed through 16S rRNA gene sequencing. Ultra High Performance Liquid Chromatography (UPLC) showed the presence of three different types of auxins in bacterial crude extracts. UPLC analysis detected the indole-3-acetic acid (IAA), indole-3-carboxylic acid (ICA) and indole-3-lactic acid (ILA). Highest auxin production of 25.9 µg ml^{-1} was observed with *Bacillus amyloliquefaciens* S-134. In bacterial-plant experiments, *B. thuringiensis* (*in vitro*) witnessed 76% and 74% root and shoot growth, overall control. In pot trials, at highest water stress i.e. 10% field capacity (FC), significant improvement of shoot length was observed with *B. amyloliquefaciens* S-134 (36%) and mixed culture M-3 (28%), over respective control. For yield parameters, 126%, 34%, 200% and 100% increases were observed, respectively, for tillers (M-3), spike length (*B. muralis* D-5), number of spikelets (M-2) and seed weight (*Enterobacter aerogenes* S-10). Rhizobacterial inoculations stimulated plant peroxidase (6 folds), acid phosphatase (2 folds) and proline content (7.4 folds) under water stressed conditions. Findings of this study suggested that application of drought tolerant rhizobacteria can help to overcome productivity losses in drought prone areas.

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Medical Microbiology **T097**

Phenotypic and Molecular Characterization of CTX-M Encoding Beta-Lactamases from Clinical *Enterobacteriaceae*

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CTX-M extended-spectrum beta-lactamases (ESBLs) are increasingly prevalent among *Enterobacteriaceae* throughout the world. CTX-M-type ESBLs exhibit powerful activity against cefotaxime and ceftriaxone. The present study was conducted at the Department of Microbiology and Molecular Genetics (University of the Punjab), Punjab Institute of Cardiology (PIC), and CitiLab & Research Center Lahore. Distribution of CTX-M group 1 and 3 extended spectrum beta-lactamases (ESBLs) producing clinical isolates was characterized by phenotypic and molecular detection techniques. Out of 285 clinical isolates 112 (39%) were ESBLs producers. 79% isolates were *Enterobacteriaceae* and 21% were Non-*Enterobacteriaceae*. Phenotypic analysis revealed that *E. coli* (36 %), *Klebsiella* spp (28 %) and *Pseudomonas* spp (21 %) were dominant pathogens in both community and nosocomial settings. Multiple antibiotic resistance (MAR) indices were calculated ranging from 0.2 to 0.9 in all bacterial isolates. PCR revealed 76% of CTX-M group1 among the selected isolates. Prevalence of CTX-M-1 was maximum among the *Klebsiella* spp (100 %), *E.coli* (97.5 %) and *Pseudomonas* spp (4.5 %). The results of another set of PCR showed that 40 % isolates of *E .coli* and 40% *Klebsiella*s spp were positive for CTXM-3. Carbapenems, tazocine and sulzones are still a good choice for treating such infections.

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Medical Microbiology **T098**

Current Extended Spectrum beta-lactamases production in *Salmonella typhi*

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Typhoid represents the 4th most common cause of death in Pakistan. Emergence of ESBL has been identified in *MDR S. typhi* after fluoroquinolone resistance. Re-emerging sensitivity of 1st line anti typhoid and emergence of ESBLS has transpired the need for genetic analysis. Four thousands seven hundred and forty eight (n = 4748) *S. typhi* isolates were collected from blood culture samples, Identified by AP1-20E, susceptibility by Kirby-Baur and Production of ESBL using cephalosporin indicator discs in combination with co-amoxiclav. *S. typhi* DNA and drug resistant genes isolated by molecular method. Susceptibility of chloramphenicol, ampicillin and trimethoprim/ Sulfamethoxazole 87.3 %, 81.1 % and 78.2 %. Ciprofloxacin 45.1 %, ceftriaxone, cefotaxime and ceftazidime was found 95.7 %, 94.8 % and 94.6 % respectively. Only three isolates appeared ESBL producers 0.69% Imipenem and meropenem were 97.7% and 97.8% effective. PCR products sequencing showed Ser 83 to Phe (TCC to TTC) mutation in all the cases. No mutation a codon 87 was seen in any case. *S. typhi* should be continuously monitored for the presence of plasmid carrying resistant markers against antimicrobials.

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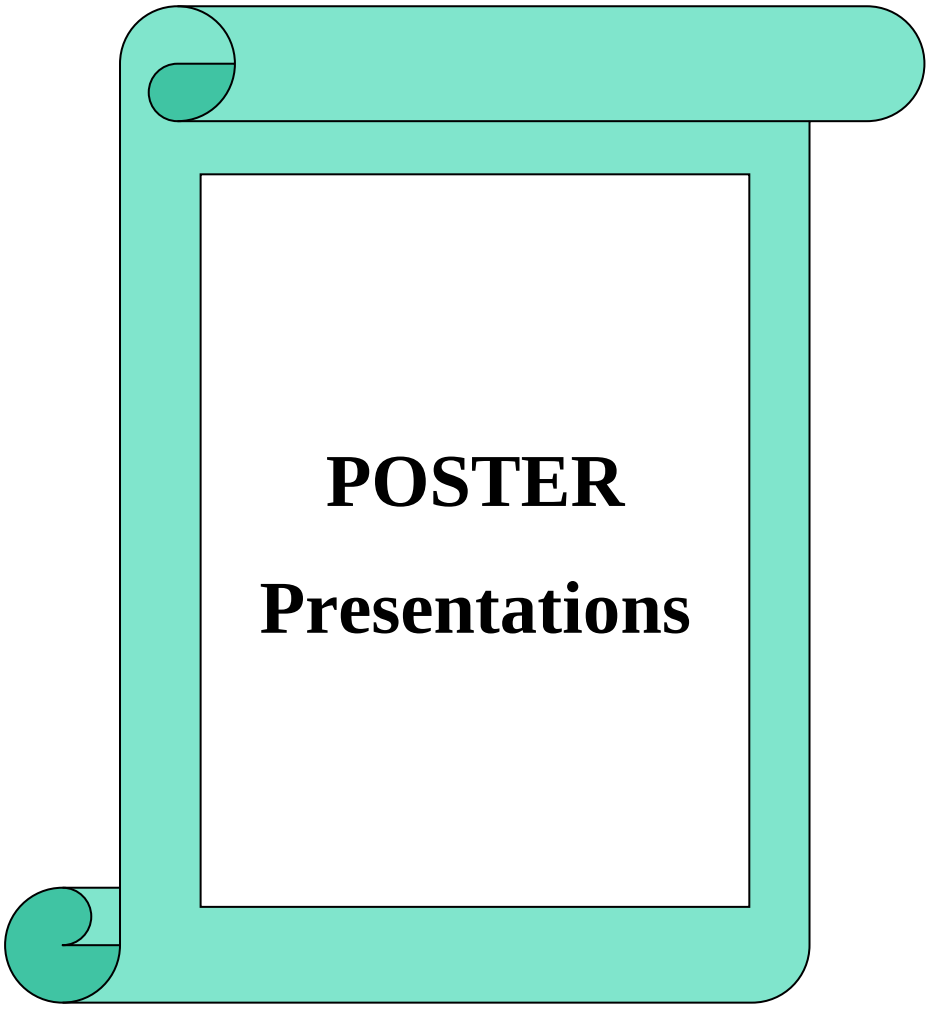

POSTER

Presentations

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Poster

Medical Microbiology **P001**

Horizontal Gene Transfer and Antibacterial Effect of *Allium sativum* (Garlic) on Methicillin-Resistant *Staphylococcus aureus*

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To evaluate the horizontal gene transfer of antibiotic resistance and antibacterial effect of Allium sativum on MRSA isolates from a tertiary care hospital, Lahore. Place and Duration: The cross-sectional study was conducted in Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore from January 2016 to July 2016. 310 MRSA isolates were processed for antibiotic susceptibility testing by using Disc Diffusion Assay. Using plasmid isolation and plasmid transfer conjugation experiments horizontal gene transfer by conjugation was studied. Competent MSSA cultures were transformed using MRSA donor DNA. Agar well diffusion assay and the effect of SDS, high temperature and Proteinase k on antibacterial activity of crude garlic extract was determined. The MRSA strains were resistant to oxacillin, penicillin, amoxicillin, ampicillin, cephradine, cefuroxime, ciprofloxacin, gentamycin, erythromycin, fusidic acid, and co-trimoxazole. Almost all stains were susceptible to vancomycin. Most strains were susceptible to ceftriaxone, novobiocin, teicoplanin, and tazobactam. Plasmids ranging in size from 25kb to 42kb were observed in selected strains. Conjugation and transformation assays showed acquisition of multidrug resistance in MSSA. Crude extracts of garlic demonstrated potent antibacterial activity against selected MRSA strains. Treatment of crude garlic extracts with physical and chemical agents (SDS, heat, Proteinase k) almost totally abolished the antibacterial activity. Here, we observed the horizontal gene transfer of multidrug resistance in Staphylococcus aureus and began to study the antibacterial activity of garlic against antibiotic-resistant bacteria.

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Poster

Antimicrobials **P002**

Phytochemical Screening, Antimicrobial and Antioxidant Activity of *Butea monosperma*

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The plant *Butea monosperma* is used as medicinal plant, it is also known as flame of forest due to color of its flower. Plant contains high amount of phytochemical compounds like alkaloids, flavonoids, carbohydrates and many others. Previous studies shows that this plant can be used to treat many diseases like diarrhea, Leucorrhoea, genital disease, diabetes, to treat intestinal worm, sore throat and many others.The main objectives of present project were to analyze the phytochemical compounds of *Butea monosperma* and checked the antimicrobial and antioxidant activity in ethanolic extracts from different parts of plant.The extract of leaves, bark and stem were prepared by the maceration method. Phytochemical analysis was also done to check the presences of many metabolites like alkaloids, steroids, phenol, flavonoids and others. Obtained extracts were also used to examine the antibacterial and antifungal activity by disc diffusion method. Antioxidant activity was also determined by using DPPH and ascorbic acid as a positive control. In phytochmical screening, extracts (leaf, bark and stem) showed the presences of saponins, terpenoids, tannis, phenol, steroids, glycoside, carbohydrates, further the ethanolic extract showed negative results to inhibit the bacteria and fungus. *Butea monosperma* has high antioxidant activity.
The extract of *Butea monosperma* has many important metabolites like alkaloids, phenol, steroids and it do not have any visible effect on microbes. Stem and bark extract has more antioxidant activity as compare to leaf.

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Poster

Environmental Microbiology **P003**

Contaminated Environments: Problems and Solutions at One Place

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Nature is a gift of God to human beings, but it is continuously disturbed by extensive use of man-made things known as xenobiotics. The xenobiotics are nowadays a global issue that have been associated with most of the health problems. The most common xenobiotic pollutants include petrochemicals, pesticides and paints. Fortunately, nature possesses its solutions within itself, we just have to explore them.To explore contaminated environments for better natural remedial measures to revive them back to normal conditions. The sites contaminated with used engine oil/diesel oil, pesticides and oil-based paints were selected and explored for enumeration of indigenous pollutant-degrading microorganisms. The bioremediation experiments were carried out under laboratory settings. The outcomes of remediation experiments were evaluated using standard analytical methods.The indigenous bacterial isolates of genera *Stenotrophomonas* and *Bacillus* could remove up to 95-99% petrochemical hydrocarbons from liquid media under laboratory settings. In addition, the indigenous bacterial isolate *Serratia marcescens* strain S1A completely biodegraded pesticide (Lambda-cyhalothrin) during 20 days of incubation. Similarly, 78-83% of oil-based paints were removed by using a novel indigenous bacterial isolate of *Brevibacillus parabrevis* strain NAP3, however three indigenous *Bacillus subtilis* strains isolated from contaminated soil of paint-warehouses displayed remarkable removal of oil-based paints from aqueous media up to 55-67%. Evidently, it was concluded that although contaminated environments pose serious health risk but, at the same time, are also a source of indigenous pollutant-degrading microorganisms.

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Environmental Microbiology **P004**

Bioremediation of Water-Based Paint (WBP) by *Pseudomonas aeruginosa* using Soil-Slury Mehtod

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Environmental pollution with aqueous paint effluents need attention due to its vast application and accidental spillage into the environment. The bioremediation is a safe, simple and reliable method for the treatment of pollutants to render them non-toxic.To explore bacterial strain Pseudomonas aeruginosa for efficient removal of water based paints (WBPs) using soil-slurry method. The bacterium was isolated from the soil sediment polluted with paints using enrichment culture conditions (12days, 160 rpm, 37°C) in mineral salt medium (MSM) containing WBPs (Conc. 300ppm). The isolate was screened using conventional molecular techniques followed by identification through 16S rRNA gene sequence homology. Afterward, the isolate was used for the treatment of WBP at different concentration (200, 400, 600, 800 and 1000 ppm) by soil slurry method in MSM broth (w/v) at environmental conditions (37°C, 180 rpm for 21st days) and percent removal was monitored using UV-Vis spectrophotometer at 365nm (λ max) against the standards curve prepared at different concentration of paints.The isolated bacterium was confirmed as *P. aeruginosa* exhibiting 99% similarity with other closely related taxa in GenBank database. The isolate exhibited maximum % removal of 90%, 85%, 77.3%, 71% and 67.1% during 3 weeks incubation from the paint concentration of 200, 400, 600, 800 and 1000ppm, respectively.The isolated bacterial strain of *a* could be further explored for molecular studies and genetic recombination in order to synthesize efficient non-pathogenic mutants for environmental cleanup of xenobiotics.

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Environmental Microbiology **P005**

Bioremediation of Oil-Based Paints (Obp) By *Pseudomonas aeruginosa* using Soil-Slurry Technique

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Worldwide health hazard from the environmental pollutants has been recognized as one of the major concern nowadays due to pollutants containing volatile organic compounds (VOCs). Oil-based paints (OBPs) consisting VOCs and other hazardous compounds also pose serious threat to the environment. Bioremediation methods offer eco-friendly treatment of such pollutants.The present study was aimed at isolation of *Pseudomonas* for the efficient removal of oil based paint effluents using soil slurry method.The bacterium was isolated using culture enrichment conditions (12days, 160 rpm, 37°C) in mineral salt medium (MSM) containing OBPs (Conc. 100ppm). The isolate was screened using conventional molecular techniques followed by the 16S rRNA gene sequence homology. Afterward, the isolate was used for the treatment of OBP at different concentration (200, 400, 600, 800 and 1000 ppm) by soil slurry method in MSM broth (w/v) at environmental conditions (37°C, 180 rpm for 21st days) and percent removal was monitored using UV-Vis spectrophotometer at 380nm (ƛ max) against the standards curve prepared at different concentration of paints.The isolated bacterium (SSP4) was confirmed as *Pseudomonas aeruginosa* exhibiting 100% similarity with *Pseudomonas aeruginosa* strain. The isolate achieved maximum % removal of 89.15%, 73.38%, 58.02%, 54.58%, and 42.08% of 200, 400, 600, 800 and 1000ppm respectively, during 21 days of incubation.The isolated strain of present study could be explored for the removal of xenobiotics for safe and hazard free environment

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Poster

Bioinformatics **P006**

Exploiting Bioinformatics Tools for Bacterial Genes Expressing Pollutant Degrading Enzymes

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Enzymatic remediation of polluted environments is considered to be the cheap, environmental friendly method. Bioinformatics tools have been considered as reliable and time-saving approaches to identify and characterize target genes available in genomic databases. To explore genomic databases for target genes expressing pollutant-degrading enzymes and to design set of specific primers for molecular characterization of such target genes.The pollutant-degrading bacterial strains and respective enzymes were selected based on previous literature. Subsequently, bioinformatics tools were used for retrieving genetic sequences against each of the genes encoding pollutant-degrading enzymes viz. phosphatase (phoD), dehydrogenase (adh), and β-glucosidase (bgl). Moreover, two different primer designing tools, i.e. Pimer3 plus and Pick Primers from NCBI, were used designing gene-specific primers for each gene. The PCR-based amplification of all three genes and gele-electrophoresis were achieved to confirm their amplicon sizes and quality.The targeted genes were successfully retrieved from bacterial species including *Pseudomonas, Bacillus, Eschrichia, Bradyrhizobium, Xanthomonas* using GenBank database of NCBI. Finally, more than 3 pairs of specific primers for each gene were designed keeping view of the size of amplicon (at least 500bp), annealing temperature and gene loci. The target genes phoD, adh, and bgl were successfully amplified using gradient PCR having amplicon sizes of 794 bp, 967 bp and 978 bp, respectively, were resolved on gel-electrophoresis.The use of bioinformatics tools can save cost and time. Moreover, the current approach can be helpful for identifying pollutant-degrading genes in environmental metagenomes

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Poster

P007

Antibiotic Susceptibility Profile of *Salmonella enterica* Isolated From Poultry Eggs, Khairpur, Sindh

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Multidrug resistant bacteria in food and food related items are one of foremost significant health and socio-economic problem, most often for the developing countries like, Pakistan.Thus present study sought to explore the isolation and identification of multidrug resistant *Salmonella enterica* from the chicken eggs.Total 300 eggs samples (150 eggs shell and 150 egg interior) were collected randomly for the isolation of pathogenic bacteria by using enrichment technique and all the isolates were identified using routine microbiological assays followed by the 16S rRNA gene sequence homology. The antimicrobial resistance was determined using Kirby-Bauer disc diffusion assay against panel of 18 different antibiotics pertaining to 09 different antimicrobial classes. The 46% (15 egg interior and 123 egg shell) samples were growth positive, while 54% (135 egg interior and 27 eggs shell) samples were growth negative at similar experimental conditions. The antibiotics sensitivity profiling results revealed that 50% of the isolated species of *Salmonella* were resistant to amoxicillin (30µg), urixin (20 µg), fosfomycin (30µg), whereas all *Salmonellas* pp were completely resistant to cephalothin (30µg), cefuroxime sodium (30µg), erythromycin (10µg), vancomycin (30µg), moxifloxacin (5µg), and azomax (15µg). Keeping in view the resistance profile, selected isolate was confirmed as *S. enteric* by gene sequence homology.Findings of current study raise a question for the quality of chicken eggs being utilized, while it is suggested that eggs should not be utilized without proper cooking in order to minimize the risk associated with eggs.

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Environmental Microbiology **P008**

Bioaerosols of The Composting Sites and Landfill Areas as a Source of Microbiological Air Pollution and Health Hazard

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The objective of this study was to document the bioaerosol levels in the air of landfill and composting sites of Lahore. Evaluation of air pollution was made during winter season. A total of 6 samples were collected from dumping Transfer stations of Valancia Town, Saghian Pull Katar Band Road and landfill sites of Lakhodair and Mehmood Booti. The mixed cellulose filter paper with 47mm diameter was used for sampling. Different media were used for total selective isolates count in the samples. The media were Trypticase soy agar for total cfu count, Voilet Red Bile (VRB) agar for total coliforms, Mannitol Salt agar (MSA) for total staphylococcus, Salmonella Shigella agar (SS) for Salmonella and Shigella, MacConkey agar for all gram negative bacteria and Starch Caesin Potassium Nitrate agar (SCPN) for Actinomycetes. 64 isolates of bacteria were isolated using Tryticase Soy Agar medium. Air samples were collected in November at about 37-43ºC for 8 min and in December at about 16-28 ºC for 8 min. Predominant identified bacterial strains were mostly *Bacillus, Brevibacillus Staphylococcus, Streptococcus* and *Micrococcus* and *Klebseilla sp*. These results suggest that the open-solid waste dumping sites are a major source of bioaerosols, and residents living in the nearby areas of landfills are at high health risks.

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Medical Microbiology **P009**

Potential Role of *Staphlococcal penicillin* Binding proteins (PBPs) in Cross Reactivity with Murine Cytokine Assay

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Methicilin resistance *Staphylococcus aureus* (MRSA) is a well-known super bug. This pathogen is notorious because of nososomial and community acquired diseases in humans. Formerly *Staphylococcus aureus* was known as opportunistic pathogen. It is estimated that approximately 30% humans have *S. aureus* as a normal nasal flora. *S. aureus* is certainly vulnerable to almost every antibiotic that has ever been established. Resistance is often developed by horizontal transfer of genes from some outside sources. The infections caused via antibiotic resistant strains of S. aureus have been reached widespread proportions globally. The global burden of staphylococcal illness mainly that is caused by MRSA strains has been increased in several countries. MRSA infection results in high temperature, cough, chills. It can also infect urinary tract, heart, or blood stream. It also causes necrotizing fasciitis. MRSA is identified by the presence of low affinity penicillin binding protein 2A (PBP-2a). This protein is encoded by the mecA gene. The resistant strains characteristically produced an enzyme, named as β-lactamase, which cause the inactivation of the β-lactam antibiotics. In response to the MRSA infection, immune response generates. Large number of macrophages and neutrophils reach at the site of infection. There occurs the production of inflammatory mediators, cytokines such as interleukin 1β, IL-12 and IL-6. in monocytes. IL-1β is the master mediator of inflammatory process. This can be quantified through sandwich ELISA. In recent study, cross reactivity has been shown by sandwich ELISA by some unknown epitopes from several strains of *S. aureus*. This work would revealed the potential role of PBP-2a in cross reactivity with murine cytokine assays.

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Medical Microbiology **P010**

Prevalence of Pediatric Infections Related MRSA Among Population of Lahore

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Staphylococcus aureus is a major pathogen causing both nosocomial and community-acquired infections. Community-associated MRSA (CA-MRSA) has caused increased MRSA infections in the general population, including children. Infact, MRSA infection chances are more common in poor hygenic conditions and in people with less developed immue system. Pediatric age group is closer to open playgrounds, floors and mostly less aware of hygenic practices in daily routine activities. Also, their immune system is yet in developing stage at such young age. A three year surveillance of CA-*Staphylococcus aureus* infections in children has shown a striking increase in frequency of MRSA infections in pediatrics. Present study is based on statistical analysis of proportion and prevalence of MRSA-associated infections in pediatric age group relating to their social economic status. Populations with a low social economic status might be at higher risk to such infections due to false diagnoses of diseases, improper treatments and also unhealthy and low-quality lifestyles. Laboratory records of MRSA isolates, antibiotic susceptibilities and information from patient medical records are reviewed from hospitals of both government and private sectors. These isolates are characterized through techniques approaching genomic level under stringent conditions in laboratory. This characterization and screening is significant because false recognition of an infection to be an MRSA-associated one calls for their treatment with higher antibiotic i.e vancomycin leading to development of more antibiotic resistant bacterial strains. The study will highlight the prevalence and potential risk factors about pediatric associated MRSA infections.

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Comparative Study of Medicinal Plant Antibiotics **P011**

Prevalence of *Pseudomonas aeruginosa* in Burn Infection and Comparative Study of Antibiotics and Medicianal Plants against Isolated Specie

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The aims of our study were association of *Pseudomonas aeruginosa* with burn infection and comparative study of antibiotics sensitivity of available antibiotics and traditional herbs medicine against isolated Pseudononas aeruginosa isolates from different ecological zones of District swat. A total 90 swab samples were collected from burn infected patients. Out of these 80.68 % were positive for *Pseudomonas aeruginosa*. Study the effect of environmental conditions Mingora region have high (52 %) prevalence when compare with Islampur region (14 %). Age group of 1 to 15 years have high prevalence with 89. 18 percent. in gender wise distribution female have more (61.97 %) infection than male (38.02 %). Antibiotics Piperacilin/Tazobactum recorded high growth inhibition with average inhibition zone of 28mm among the 9 examined antibiotics. Ciprofloxacine and cefoperazone/Sulbactumhave intermediate response with 21mm and 19mm average zones of inhibition respectively. While Polymixin showed no response against *Pseudomonas aeruginosa*. *Berberis lyceum* and *Polygonum aviculare* have remarkable activities with 27.33 mm and 23.33mm growth inhibiotn zones, respectively. Similarly, *Trachspermum ammi* (19.88mm), *Menthalon gifolia* (17.88mm) and *Sapindus mukrossi* (17.88mm) have intermideaate response against isolates of the given pathogen from different ecological zones while *Feoniculum vulgaris* and *Ocimum basilicum* have no effect on growth of isolates.

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Cancer Genetics **P012**

Detection of Inherited Mutations for Breast Cancer in Pakistani Patients

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Breast cancer is the second most common type of cancer in Pakistan with approximately 40,000 new cases every year. Mutations in BRCA1 and BRCA2 genes are considered to be the main cause of breast cancer and are extensively studied now a day. The aim of the study is to check some common inherited mutations like BRCA1 5382insC, BRCA1 MS, BRCA1 4184del4, BRCA1 1294Del40, BRCA2 2157delG in the population of Pakistan. After DNA isolation, genotyping of 100 samples (50 breast cancer patients and 50 control) is being carried out using Allele specific PCR. Mutations are detected by running the samples on polyacrylamide gel electrophoresis and further downstream by RFLP (Restriction Fragment Length Polymorphism). Whole procedure is under process. As allele specific PCR and RFLP are highly sensitive for detecting mutations in the genes of DNA fragment and the mutation rate is 57% for BRCA1 gene and 33% for BRCA2 gene in Pakistan. So, it is expected that the selected mutations will be inherited in the patients of breast cancer in Pakistan.

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Antimicrobial Agents **P013**

Phytochemical Investigation, Antioxidant and Synergistic Activity of Leaf Extracts of *Piper betle* and *Cymbopogon citratus* with Some Antibiotics against Clinical Pathogens

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The resistance against antibiotics in the microorganisms is increasing day by day. Synergism, i.e. the combination of two or more agents, is a new approach in combating the resistant bacteria. *P. betle* and *C. citratus* have been used traditionally in various medicine systems of the world to treat many diseases. So, this study is designed to investigate the chemical constituents of these medicinal plants and also to check the potential of these plants with antibiotics to be used as a powerful agent against microorganisms exerting a greater inhibitory effect. The leaf extracts of *P. betle* and *C. citratus* were prepared by using water, methanol and acetone. These extracts were subjected to phytochemical analysis. Antioxidant activity was determined by reducing power assay. Antibacterial activity and synergistic activity was determined by using agar well-diffusion method against three clinical pathogens, i.e. *E. coli*, *S. aureus* and *P. aeruginosa*. Micro-tube dilution method was used to determine the MIC of the extracts. The phytochemical screening revealed the presence of carbohydrates, proteins, phenols, flavonoids, terpenoids taninns and saponins in *P. betle* and carbohydrates, proteins, phenols, alkaloids, tannins, glycosides, quinines and saponins in *C.citratus*. The highest antioxidant activity was found to be 86% by acetone extract of *P. betle* and the lowest antioxidant activity was exhibited by aqueous extract of *C. citratus* to be 30% in comparison with the ascorbic acid. Antibacterial activity was found to be highest for *E. coli* by acetone extract of P.betle with 14 mm zone of inhibition, and 9mm for *S. aureus* and *P. aeruginosa*. All the plant extracts when combined with Streptomycin showed comparatively greater synergistic activity against *E. coli*, when combined with Chloramphenicol showed greater synergistic activity for *S. aureus* and when combied with Ciprofloxacin showed best results for *P. aeruginosa*.

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Punjab Institute of Cardiology Lahore **P014**

Antimicrobial Susceptibility Profiles of Bacterial Pathogens Associated with Endocarditis

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This prospective study was performed at Punjab Institute of Cardiology to determine the bacterial pathogens causing endocarditis along with in vitro estimation of antimicrobial susceptibility profiles followed by measuring the in vivo effect of empirical antibiotic therapy in endocarditis patients. For the purpose, a total of 110 blood samples from endocarditis patients aged 20-40 years were collected. The antibiotics already given to the patients as empirical therapy were noted before sampling. A total of 60 samples were found positive including 53 (88.3%) Gram positive and 7 (11.7%) Gram negative bacteria. In case of Gram positive isolates, 39 (65%) were Methicillin-sensitive *Staphylococcus aureus*, 2 (3.3%) were Methicillin-resistant Staphylococcus aureus and 12 (20%) were *Streptococcus*. Among Gram negative bacteria 5 (8.4%) isolates were of *Escherichia coli*, 2 (3.3%) isolates of *Pseudomonas aeruginosa*. The antimicrobial susceptibility of the isolates against various antibiotics was determined in vitro by using Kirby Bauer disc diffusion method. In empirical therapy, the combination of Benzyl Penicillin-Gentamicin was given to group-I patients, combination of Vancomycin-Gentamicin was given to group-II patients and Vancomycin to group-III patients. In vitro antimicrobial susceptibility data showed that all *S. aureus* isolates were susceptible to Vancomycin, and all *P. aeruginosa* showed were susceptible to Ciprofloxacin, Ceftazidime, Piperacillin-Tazobactam and Tigecycline while *Escherichia coli* showed 60% susceptibility against Amikacin and Co-Amoxiclav. Results of empirical therapy showed fast recovery in Group-II individuals treated with combination of Vancomycin-Gentamicin as compared to Group I and Group-II.

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Medical Microbiology **P015**

Genetic Sequence of *Clostridium tetani* by using Multilocus Sequence Typing

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The most theatrical and generally widespread diseases of humans is tetanus. It has been reported for over 24 centuries and is the main issue in developing countries. The expression of the disease is spastic paralysis and muscle contraction mainly lock jaws that is caused by poisonous material named is tetanus toxin. Fortunately, World Health Organization is successfully controlled this disease through immunization with tetanus toxoid, it is widely spread disease in developing countries however mostly cause neonatal tetanus that is major issue. *Clostridium tetani* is an anaerobic spore-forming bacterium, whose natural habitat is soil, dust, and intestinal tracts of various animals. Here Report the cmplete squence of *C. tetani* by the use of multilucos. Tetani E88, a variant of strain Massachusetts. The genome consists of a 2,799,250-bp chromosome encoding 2,372 ORFs. The tetanus toxin and a collagenase are encoded on a 74,082-bp plasmid. It is an anaerobic spore-forming bacteri containing 61 ORFs. Additional virulence-related factors could be identified, such as an array of surface-layer and adhesion proteins (35 ORFs), some of them unique to *C. tetani*. Comparative genomics with the genomes of *Clostridium perfringens*, the causative agent of gas gangrene, and *Clostridium acetobutylicum*, a nonpathogenic solvent producer, revealed a remarkable capacity of *C. tetani*: The organism can rely on an extensive sodium ion bioenergetics. Additional candidate genes involved in the establishment and maintenance of a pathogenic lifestyle of *C. tetani* are presented.

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Environmental Microbiology **P016**

Microbial Degradation of Sulfamethoxazole

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Ongoing industrial development, urbanization and agricultural trends have given rise to a series of environmental problems among which degradation of water bodies is an alarming area of great concern. Various types of pharmaceuticals detected probably enter the water bodies through runoff water from poultry sheds, sewage and aqua culture. Sulfamethoxazole (SMX) is amongst those antibiotics which are extensively used as human and veterinary medicine. The aim of this study was to isolate and identify potential environmental bacteria to degrade SMX and to assess the effects of the antibiotic towards non target organisms such as fish. Since pharmaceuticals are designed to be biological active even at low concentration, these pose a risk to aquatic wildlife. In this study different strains of bacteria were isolated from the five tributaries (Ratahutar, Nupur, Jinnah, Quaid-e-Azam, Shahdara and Korang) that converge to form Rawal Lake the main source of drinking water for the inhabitants of Islamabad and Rawalpindi. Samples were collected from upstream and downstream points. Physicochemical parameters and microbial analyses of the samples was done. Total viable count was performed by spread plate technique. All samples showed values under the permissible limits for TDS, salinity, pH, turbidity and electrical conductivity except Korang River samples which depicted higher values for turbidity. The microorganisms isolated were used to degrade sulfamethoxazole after enrichment was performed in MSM media where SMX was used as sole carbon source. Degradation analysis was performed using HPLC revealed potential strains that may degrade SMX, identification of the strains was performed by PCR and 16SrRNA gene analysis.

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Environmental Microbiology **P017**

Isolation and Molecular Characterization of Potential Lambda Cyhalothrin Degrading Bacteria

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Synthetic Pyrethroids are now extensively used for the improvement of crop production and quality in Pakistan. Along with the advantages, pyrethroids also pose serious threats to organisms and environment. The present study is designed to isolate and identify different bacterial strains isolated from water samples to degrade the pesticide Lambda Cyhalothrin, a known synthetic pyrethroid. The water samples were collected from different streams of Rawal Lake. Physico-chemical characterization of water samples was performed. LC degrading bacterial strains were isolated from water samples and were taxonomically characterized on the basis of their morphology, Gram staining, biochemical tests and phylogenetic similarity index of 16S rRNA gene sequence. Potential discrete colonies were isolated and purified by streak plate method. The strains were acclimatized by inoculating them into MSM (minimal salt medium) containing various concentrations of lambda cyhalothrin (100-500 mg/L) as a sole source of carbon. Optical cell density was also monitored with the help of UV-visible spectrophotometer. A bench scale reactor system was fabricated that contained MSM having optimum pesticide concentration and isolated bacterial cultures. The sample was collected at different time intervals (0, 24, 48, 72, and 96 hrs.) and percentage of residual lambda cyhalothrin was determined using High performance liquid chromatography. The residual level of pesticide decreased in the sample collected after prolong period of biodegradation and simultaneously the percentage of degradation also increased gradually indicating the capability of the selected microorganisms in degrading the pesticide, lambda cyhalothrin.

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Public Health **P018**

Evaluation of NUST's Constructed Wetland and Determination of Parasite Presence in Raw Wastewater by Bailenger Method

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Constructed wetland is a biological wastewater treatment method, which has great potential for improving treatability of wastewater. The main aim of this study was to evaluate performance efficiency of constructed wetland developed in the NUST and identify and enumerate parasites presence in raw and treated wastewater. In this study, weekly sample were collected from both Inlet and outlet of constructed wetland for a period of 6 months. Physicochemical and biological analysis i.e. Temperature, Chemical Oxygen Demand, Total Dissolved Solids, Turbidity, Total Coliforms and, Electric Conductivity were carried out according to standard methods of APHA, 2012. Helminthes eggs were identified by microscope using modified Bailenger method using Mc master slides. The results showed significant performance efficiency (%) of COD, EC, Turbidity and TDS of wastewater up to 58 %, 77%, 85% and 67% and effective Total Coliforms and nutrient removal up to 82% by constructed wetland. Total Coliforms concentration were significantly higher in raw wastewater than treated wastewater Parasitological analysis of raw wastewater showed presence of increased number of helminthes eggs per liter as compared to treated wastewater. Mainly *Ascaris lumbricoides* eggs, *Hymenolepis nana* eggs, and *Trichuris trichirua* were present. The study revealed the effectiveness of constructed wetland for wastewater treatment and acceptable nutrient and parasites removal. To improve public health there's a need to reduce parasitic contamination, encourage public for proper use of disinfectants.

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Antimicrobial Drugs **P019**

Probiotics as a Living Drugs for Urinary Tract Infections

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The consumption of probiotics increases due to the progression in probiotics use. In previous history, probiotics use to treat UTI by their potentiality; mostly this has concern due to their antibiotics resistance. *Lactobacillus* and *Bifidobacterium* mostly use on large scale now a days. They are gut beneficial bacteria, these organisms have enzymes, H_2O_2, lactic acid, other acids which are utilize as an antimicrobial agent to treat digestive pain, eczema, tumors, ulceration. Dairy-based products are mostly that contain probiotics like; milk, yogurt, cheese, ice cream, milk powder, dark chocolate, fermented milk etc. samples were collected from different market enriched all samples for 24 hours after that check morphology by gram staining, biochemical test, catalase test, and check antimicrobial activity against uropathogens (*Klebsiella* specie and *E.coli*) to measure the zone of inhibition of tested sample. Current study showed results that were probable to promote rate of UTI treatment in women who are above 50 years. It was exposed that natural products contain probiotics which participate main function in consumer for their beneficial health effect. These probiotics colonize in intestine and execute as a barrier to prevent the urinary tract infection.

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Bioinformatics **P020**

Lung Segmentation in CT Scans for Lung Nodule and Cancer Detection

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Cancer is difficult to diagnose because of its complexity. It is a heterogeneous disease which adds to the difficulty of diagnosis and prognosis. Lung cancer is among the most inflicting type of cancer. It has high incident rate and high mortality rate as it is often diagnosed at the later stages when it is challenging to treat it. Therefore, a significant research effort is being done to help the oncologists in early lung cancer diagnosis and treatment. Computed Tomography Scan (CT Scans) are widely used to detect the disease; it helps to visualize small nodules or tumors which cannot be seen with a plain film X-ray. Computer Aided Devices (CAD) are being developed to diagnose the disease at earlier stages efficiently. The preliminary stage of lung cancer diagnosis via CAD is lung segmentation from the chest CT scans. The lung segmentation is considered a fundamental activity in such image analysis systems as the performance of the later stages in such analysis largely depends on the segmentation accuracy. In this paper, we propose a lung segmentation algorithm. The algorithm utilizes the morphological image processing techniques to efficiently segment the lungs from the chest CT scans. The propose algorithm works in four steps: in the first step, the preprocessing the CT scan images is performed, which includes the conversion of DICOM images into a loss aversion format i.e. PNG. In the second step, a histogram of the gray scale image is constructed to automatically estimate the threshold to separate the lung region from the background. In the third step, the connected components are computed to remove the any remaining background. In the final step, the morphological operator dilation is used to improve the segmentation.

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Agricultural Microbiology **P021**

Biological Control of Leaf Spot causing Fungal Pathogens in Red Edge *Dracaena*and *Sow Thistle*

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Leaf necrosis is a common fungal problem of plants. During a survey of Lahore, *Dracaena* Red Edge (*Dracaena mariginata Lam*.) and *Sow thistle* (*Sonchus oleraceus L.*) were found to be infected with leaf spots. Pathogens isolation from the infected leaves of both plants was done followed by confirmation of Koch's pathogenicity postulates. Based on morphological and genetic characterization, *Alternaria arborescens Simmons.* and *Phyllosticta aristolochiicola R.G. Shivas*. were isolated from *D. mariginata* and *S. oleraceus*, respectively. In addition to isolation and identification, biological control of both pathogens was conducted using methanol extract of Cinnamon (*Cinnamomum verum J. Presl.*). All concentrations of extract suppressed the growth of both pathogens to variable extent. In case of *A. arborescens*, 0.5% concentration induced approximately 90% suppression in fungal growth. Contrastingly *P. aristolochiicola* was least affected at lower concentrations of extract as only 4-10% arrest in fungal biomass production was recorded at 0.5-1.5% concentrations. However 2.0% or more of the extract was most potent as fungal pathogens failed to grow at these concentrations. Therefore, Cinnamon extract was considered more effective in controlling *A. arborescens* than *P. aristolochiicola*. Further studies are required to identify active antifungal compounds in Cinnamon crude extract against the target pathogens.

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Poster

Agricultural Microbiology **P022**

Identification and Management of Alternaria ochroleuca -A Cause of Leaf Necrosis in Money Plant

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Diseases caused by fungal pathogens are very common to occur worldwide. Biological control is an approach that provides safe fungal management program and a substitute for reliance on chemical treatments. A survey was conducted in vicinity of Institute of Agricultural Sciences, University of the Punjab, Lahore and Money plant (*Epipremnum aureum)* was found to be infected with fungal leaf spot. The infected samples were collected for isolation, purification, and identification of the pathogen. The identification was carried out microscopically for morphological characterization and genetically from nucleotide sequencing of amplified ITS1-5.8S-ITS4 region of rDNA. *Alternaria ochroleuca* was identified as a leaf spot causing pathogen of money plant. Afterwards, pathogenicity aptitude of identified pathogen was confirmed by re-isolation of same pathogen from the artificially innoculated leaves of host plant using detached leaf method. Further in this study, the biological control of A. ochroleuca was carried out using methanol extract of *Piper nigrum L*. (Black pepper) and *Amomum subulatum Roxb* (Cardamom). Both types of extracts presented varied results. However, all the employed concentrations of methanolic Cardamom extract suppressed the fungal growth except 1.5% concentration. Contrastingly, Black pepper extract didn't show any inhibition in fungal biomass production. Therefore, Cardamom extract was considered more effective in controlling *A. ochroleuca*. Further studies will be carried out to fractionate different compounds from Cardamom and to determine the efficacy of these compounds against target pathogen.

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Poster

Antimicrobial Agents **P023**

Mining of Marine Biowaste for Biotechnological Approach: Extraction of Chitin for Potential Antimicrobial Activity

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Marine biota is constituted by materials with a vast range of properties and characteristics. The sustainable exploitation of natural marine resources and the valorization of residues from marine origin, constitutes a highly interesting platform for the development of novel biomaterials. Within the seafood industries, the waste management of crustacean waste is a huge environmental issue due to its lack of cost-effective utilization. There has been considerable interest in the development and commercialization of usable products derived from marine biowaste materials. Crustacean shells comprise of chitin, a polysaccharide which could be recovered from the crustacean exoskeletal structure. Chitin is eco-friendly biopolymer with a wide range of unique applications. The present research demonstrates that how our natural marine resources could be utilized for the extraction of promising antimicrobial chitin based polymers. In order to minimize marine biowaste, fishery food processing waste samples were collected from Pakistan Marine Fisheries to recover chitin by using effective green technology. To determine the biotechnological potential of chitin, their antimicrobial activity will be conducted which will govern the vast array of applications for these commercially valuable chitin-based products. The recycling of crustacean shell biowaste which is usually disposed of in landfills will be beneficial to the environment as it can contribute to the bioeconomy as value-added biomaterials.

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Poster

Environmental Microbiology **P024**

Analysis of Arsenic Biotransformation by Purple Non-Sulfur Bacteria

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Purple non-sulfur bacteria (PNSB) are gram negative anaerobic or microaerophilic rods belonging to Phylum *Proteobacteria*. They can grow anaerobically by using different carbon sources as electron donors and by respiring different compounds as electron acceptors in their electron transport chain. PNSB were isolated from industrial effluent and paddy fields. The bacteria were found to resist nickel, cobalt, copper, selenium etc. Most of the isolates were able to utilize succinate, propionate, acetate, oxalate, citrate and lactate. Some of PNSB were found to respire arsenic in the presence of oxalate and lactate. Production of photopigments was estimated. All the isolates were found to contain bacteriochlorophyll a. Qualitative and quantitative arsenic estimation assay was done to determine their ability to biotransform arsenic. All the isolates were found to reduce arsenic. These isolates were identified as *Rhodospirillaceae bacterium*, *Enterobacter cloacae* and *Rhodospirillum* by 16S rRNA gene sequencing. Further studies are needed to upgrade this lab scale project to pilot scale, for bioremediation of arsenic.

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Poster

Plant-Microbe Interaction **P025**

Study of Modulating Interactions of Rhizobacteria Using *Zea Mays* as Host Plant

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Plant rhizosphere is a soil region closest to plant roots inhabiting different types of microorganisms including rhizobacteria. The rhizobacteria which enhance the plant growth, are known as plant growth promoting rhizobacteria (PGPR). The chemical fertilizers which have been conventionally used for enhanced crop production are dangerous in terms of minimizing nutritional value of crops and may also be perilous for biological agents such as wasps, so the use of PGPR is favourable for improved crop production over chemical fertilizers. In this study, twenty five rhizospheric bacterial strains were isolated from Baluchistan and tested for plant growth promoting traits i.e., HCN production, ammonification and auxin production. Most of the bacterial strains gave positive results. To study the beneficial effects of these bacteria on plants, plant-microbial interaction assay was done using *Zea mays*. Results revealed that these PGPR enhanced the growth as compared to control plants. Bacterial isolates N2, T6, Cn6, Cn7 and PP3 proved as strong ammonia producers. The isolates Cn5, PP2 and PP5 showed strong potential of HCN production whereas only Cn6 and PP3 were auxin producers. Increase in fresh weight of plants was observed in treatment with N5 showing 96.67% increase over control. Pl2 showed increase (32.14%) in shoot length while Cn5 showed prominent increase (64.95%) in root length compared to control plant. The isolates Cn5 and Cn4 showed improvement in total chlorophyll content of treated plants with percentage increase of 100% and 99.82% respectively. In conclusion, these PGPR may further be used in agriculture research for growth improvement.

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Poster

Medical Microbiology **P026**

ESBL and MBL producing *P.aeruginosa*; A potent Enemy

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Pseudomonas aeruginosa is an important bacterial pathogen most frequently responsible for nosocomial infections, especially in immunocompromised patients. The aim of our study is to determine the emergence of ESBL & MBL in P.aeruginosa. Early detection of these threatening β-Lactamase producing organisms is essential to aid infection control and to prevent the dissemination of these organisms. Eighty eight strains of *P.aeruginosa* have been collected from Chughtais Lahore Lab, CMH Hospital and Children Hospital Lahore. Identification of these strains was done by their morphology, cultural characteristics and biochemical profile. Susceptibility to various antibiotics and production of Extended-Spectrum β-Lactamase (ESBL) and Metallo-Beta-Lactamase (MBL) were determined using modified Kirby Bauer disk diffusion method, double disk synergy test and disk potentiation test / inhibitor-potentiated disk diffusion test (IPD) respectively. Out of eighty eight strains tested three were ESBL (3.9%) producers and a total of eleven strains (12.5%) were found to be resistant to carbapenems and of these eight (72.7%) were MBL producers. All these β-Lactamase producing strains (14 strains) were multidrug-resistant (MDR). Piperacillin and Piperacillin/tazobactam proved to be the most effective antibiotics in both types of β-Lactamase producing strains.

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Poster

Food Micrbiology **P027**

Food Allergens

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Food allergy is defined as the immune response of the body against the food substances especially the protein enrich diets. Food allergy is affecting 8% of young ones and 2% of adult people in the western countries. The occurrence of food allergy give rise to allergic reactions leading to all the allergic diseases in the body. Anaphylaxis increases the immune response mediated by IgE in case of skin allergy (urticaria) and as a result disorders regarding cell mediation are well recognized such as enterocolitis induced by food protein and eosinophilic esophagitis. The IgE molecules circulate in the body and attach to specialized cells called basophils and mast cells. Mast cells being present all over the body and are abundantly found in skin, mucosa of the lungs and GIT (gastro intestinal track), mouth, eyes and nose. In a sensitized individual, the food proteins bind to the IgE attached to the mast cells stimulating the release of chemical mediators such as histamine. There is an interaction of mediators with specific receptors present mainly in the skin, throat, airways, intestines, and heart ultimately leading to several allergic reactions. New technologies regarding pathogenesis of various diseases that are either IgE and non-IgE mediated have been developed. These techniques have protective mechanisms against the allergic reactions. Food allergy is primarily managed in order to avoid the consumption of the food causing allergic reactions. Ingredient labels of food products should be carefully checked regarding food composition of the product. Recently, management associated with food allergens is to create awareness among the patients and all the people to avoid consuming the food and other substances that are the major cause of food allergy. In case of ingestion, supportive therapy (prophylaxis) should be provided. However, Unique strategies are being studied, including immunotherapy through sublingual /oral ways and are believed to have a bright future.

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Poster

Nanobiotechnology **P028**

Food Safety and Nanobiotechnology

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Food safety is an area of emerging interest because the people now a days are more conscious about their health as well as the nutritive value of food. Food safety is essential in protecting individuals from pathogens that can be passed through food consumption. Proper hygiene during production, storage & packaging are important to alleviate illnesses and fatalities thus improving the safety and wholesomeness of food products. The food industry is under intense pressure to ensure food safety and at the same time to increase profit margin. Nano-biotechnology is the understanding and control of matter at dimensions of coarsely 1 to 100 nano-meters, where distinctive phenomena assist novel applications. Nanotechnologies may beneficially contribute to food safety and also expected to bring a range of advances in food sector including better tastes, textures and sensation, nutrient absorption, improved packaging, and traceability of food products. The innovative lightweight, stronger, functional packaging may extend shelf life of food, improve food safety, alert consumer that the food is spoiled or contaminated and food products with less or no preservatives. Nano-biotechnology holds great promise to provide benefits not just within food products but also around food products. The use of nano-biotechnology in the food safety industry is here and if estimates are accurate, it will continue to develop rapidly over the next few years. Its potential for providing safer and more nutritious foods is vital; however, there are major knowledge gaps in our understanding of the properties, behavior and effects of the Nano-materials that are (or may be) used for food applications. There is a need for a practical approach to a case-by-case pre-market safety evaluation of the nano-biotechnology-derived food products to ensure maximum food safety and personal health protection.

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Environmental Microbiology **P029**

Escherichia coli from Waste Water: Antibiotic Sensitivity, Biofilm Formation and Screening for Shiga Toxin Producing Genes

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Contamination of Potable water is the leading cause of death worldwide. Waste water if not treated properly poses health related hazards. Infectious disease haemorrhagic uremic syndrome is common worldwide due to presence of pathogenic strains of *Escherichia coli* in the environment. This study was conducted to isolate the pathogenic strains of *E. coli* O157 from the sewage waste water of Lahore, Pakistan. Selective media Hichrome EC O157 agar was used for isolation of *E. coli*, taxonomic status of strain was confirmed by 16S rRNA gene sequencing. Serotyping of bacterial strains by ProlexTME. coli O157 Latex Test Reagent Kit results for O157. Bacterial strains were also evaluated for biofilm formation and toxin genes (stx1, stx2, stx2c, stx2d) amplification. All the strains were catalase positive, oxidase negative, MR positive and VP negative. %. Bacterial strains showed resistant pattern against different antibiotics. *E. coli* strains were screened for shiga toxin producing genes and biofilm formation. PCR amplification recorded negative results for shiga toxin genes Maximum biofilm was produced by strain E124 when used as monoculture while in cocultures, strains E35 & E101 were efficient biofilm formers. Shiga toxin genes were not present in any of the *E. coli* strain, from which we can suggest that our environment is free of shiga toxin genes. *E. coli* was present in sewage water, its cross contamination with drinking water may affect the community. So, waste water should be treated properly before discarding it into the common water bodies.

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Poster

Infectious Diseases **P030**

Antibiotic Sensitivity Testing of *Staphylococcus aureus* Strains from Urinary Catheters at A Tertiary Care Hospital in Haripur

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Staphylococcus aureus is a medically significant bacterial species that can cause a range of infections including superficial skin infections, catheter associated infections, urinary tract infections, food poisoning, necrotic pneumonia etc. A large number of *S. aureus* may be present in hospital environment as well as on patient body that may cause catheter associated infections among catheterized patients. Recent studies from different cities of Pakistan report a high level of antibiotic resistance to commonly used antibiotics against *S. aureus*. Further, nosocomial transmission of MRSA among catheterized patients may complicate their treatment. The aim of the present study was to isolate *S. aureus* from urinary catheters of hospitalized patients and determine antibiotic resistance profiles of these strains. One hundred thirty seven swab samples were collected from gynae ward of DHQ hospital Haripur, KPK Pakistan. The swab sample was taken by rubbing the sterile swab on the used urinary catheters by the patient. Strains were identified by using different microbiological and biochemical assays. Antibiotic sensitivity testing was performed by disc diffusion assay. The isolated strains of *S. aureus* show resistance against a number of antibiotics used in our study. The overall resistance profiles of the strains are as following, 83% (n=33) resistance to lincomycin, 75% (n=30) to bacitracin, 65% (n=26) to cefoxitin, 60% (n=24) to ceftriaxone and rifampin, 45% (n=18) to erythromycin, 38% (n=15) to doxycycline and ciprofloxacin, 35% (n=14) to gentamycin and 30% (n=12) to sulpamethoxazole. The percentages of MRSA and MSSA are 65% and 35% respectively. The results of this study reveal that strains isolated from urinary catheters are highly resistant to all tested antibiotics and there is a high risk of nosocomial infection among those receiving urinary catheters.

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Poster

Biodegradation **P031**

Biodegradation of Petroleum Hydrocarbon by *Micrococcus* Sp. in Form of Wax Ball

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Hydrocarbon containing compounds like diesel, crude oil and petrol cause environmental pollution. Hydrocarbon contaminated sites contain large amount of microbial community adapted for utilizing these compound as a source of carbon and other metabolic activities. A study was conducted in order to isolate the micro-organism from hydrocarbon contaminated sites for petroleum degradation. The soil sample was collected from the bus terminal of Punjab University, Lahore. The isolate (*Micrococcus* sp.) was characterized microbiologically by using microbiological techniques such as staining and biochemical testing based on Bergey's manual. Isolated organism has an ability to degrade the petrol as well as paraffin wax as a sole carbon source. This ability shows clear evidence that genome of this isolate harbors gene for degradation. By combined both properties, we can reduce petroleum product from hydrocarbon impacted environment.

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Poster

Public Health **P032**

Awareness of Iron Deficiency Anemia among the Women of Punjab, Pakistan

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The empirical objective of conducting the survey inquest is appraising the awareness level about the iron deficiency anemia among the general population (females having age greater than 18 years) of Punjab, Pakistan. A structured questionnaire having discrete segments elucidating iron deficiency anemia's causes, symptoms, treatments and transmission is accordingly filled by the women with their consent. All the data obtained was interrogated and scrutinized by Chi-square test and percentage analysis centered on the age, education and marital status of the women.

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Poster

Medical Microbiology **P033**

Distribution of Carbapenem Resistant Metallo-Beta-Lactamase (MBL) Producing Gram Negative Bacteria in a Tertiary Care Hospital, Lahore

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Carbapenems are beta-lactam drugs and only treatment option against extended spectrum beta-lactamases (ESBLs). However, the appearance of carbapenemases, showing high hydrolysis profile towards all drugs, has compromised their efficacy over the last decade. Recent studies have testified global appearance of highly resistant variants of *Klebsiella pneumoniae* carbapenemases (KPCs), New Delhi Metallo-beta-lactamases (NDMs) and IMP carbapenemases belonging to *Enterobacteriaceae*. These agents can only be treated using inhibitors like EDTA, supplied in combination with imipenem or meropenem. In the current study, the selected imipenem resistant isolates were characterized and metallo-beta-lactamase (MBL) activity was confirmed using combination disc test and modified-Hodge test. The isolates were genotypically analyzed for the presence of blaIMP, blaOXA and blaSHV resistance genes by means of colony PCR.The study revealed highest frequency of MBLs among *Klebsiella spp*. and *Pseudomonas spp*., and highest age related drug resistance in age-group 31-45 years, with greater ratio in males. The isolates showed highest resistance against carbapenems, among which 70% were confirmed to be metallo-beta-lactamase producers, along with monabactams and cephalosporins. Colony PCR confirmed the presence genes blaSHV and blaOXA (ESBL activity) in few isolates confirming them to be MDRs. In addition, PCR for detection of MBL gene blaIMP-1 gene was optimized. In conclusion, the study demonstrates recent emergence and prevalence of multi-drug resistant MBLs among various clinical isolates. MBLs are being referred to as the beginners of post-antibiotic era. There is a need for the development of effective and sensitive phenotypic diagnostic tests and easy molecular detection techniques for the screening and monitoring of MBLs.

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Poster

Environmental Microbiology **P034**

Anoxic Growth Optimization of Arsenic Resistant Purple Non Sulfur Bacteria

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Purple nonsulfur bacteria are anaerobic photosynthesizing organisms capable of growth by multiple metabolic pathways. Fourteen isolates were enriched with arsenic from two sources; industrial drainage and fish pond. Growth optimization tests were performed using different carbon sources with metals as combinations for electron donor and acceptor, respectively. Lactate and acetate proved to be good electron donors while nickel and selenium were good electron acceptors. MIC was determined for arsenate and arsenite, maximum resistance at aresenate 1 mM and arsenite 0.5 mM for all isolates. Bacteriochlorophyll detection test revealed presence of bacteriochlorophyll a. Carotenoid estimation was done and a maximum of 4.2 mg g^{-1} was calculated. After growth optimization, gas production was checked for selected isolates with best activity in all assays. Results revealed that lactate and acetate were efficiently utilized with arsenic for gas production. This study forms the basis of important biotechnological procedures which employ the uniqueness and versatility of PNSBs.

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Poster

Environmental Microbiology **P035**

Anoxic Growth Optimization for Photobiological Gas Production by Arsenic Enriched *Rhodopseudomonas plaustris* Isolated from Fish Pond

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Purple nonsulfur bacteria are anaerobic photosynthesizing organisms capable of growth by multiple metabolic pathways. Seven isolates were enriched with arsenic from fish pond out of which one strain (PI5) was homologically identified as *Rhodopseudomonas plaustris* with properties of applicable bio-remedial versatility in its growth optimization profile. Profiling done for all isolates revealed lactate and acetate as good electron donors while lead, cobalt and selenium as good electron acceptors. For selected isolates with best activity in all assays, gas production was detected. Rhodopseudomonas plaustris is capable of oxidizing complex carbon source benzoate while respiring metals arsenic and selenium and is capable of photofermentative gas production. The study reveals importance of PNSBs with reference to heavy metal resistance as a maximum of 10 mM arsenate and 2.5 mM arsenite were observed, the underlying mechanisms of which seemingly unaffected by aerobic conditions, observable in the case of Rhodopseudomonas plaustris as well. Switchable modes of metabolism of PNSBs lead us to study that anaerobic profile is better suited to the efficiency of PNSBs when being employed for industrial processes. Carotenoid harvesting is among industrial uses of PNSBs, to which our study contributes a recorded maximum of 2.04 mg g^{-1} carotenoid content.

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Medical Microbiology **P036**

Antimicrobial Susceptibility Profile of Urinary Tract Pathogens from a Tertiary Care Hospital, Faisalabad

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Poster

Proteomics **P037**

Upregulation of Aroma Related Proteins in Heat Shocked Rice using Foliar Application

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Rice is the second major cash crop of Pakistan after wheat, where aromatic rice is preferred over non-aromatic rice as an important source of foreign exchange earning. Numerous aroma compounds have been identified amongst which, 2-acetyl-pyrroline (2 AP) is known to be the potent compound imparting fragrance in aromatic rice. Genetics reveal fgr gene on chromosome 8 codes for the enzyme betaine aldehyde dehydrogenase 2 (BADH2). Mutation in this gene truncates to produce non-functional BADH2 leading to accumulation of 2AP in rice. Besides BADH2 enzyme, other enzymes are also identified in the 2AP biosynthetic pathway for aroma such as glyceraldehyde-3-phosphate dehydrogenase (GAPDH), aspartate aminotransferase (AAT) and glutamine synthetase (GS). The 2 AP biosynthetic pathway also identifies proline as an important precursor of 2 AP. It is also observed to act as an osmolyte against various environmental stresses amongst which, high temperatures may compromise fragrance trait in rice. The current study aims to use the dual role of proline to mitigate damaging effects of heat stress while retaining aroma in rice. Rice will be cultivated, and leaf samples will be collected at heading, flowering and maturity stages followed by proline spray application and heat shock treatments. Total proteins will be quantified followed by 2-dimensional gel electrophoresis. Proteins will be identified by MALDI-TOF/TOF-MS followed by GC-MS for 2 AP quantification. The study proposes the idea of utilizing foliar proline approach to ameliorate high temperature stress for selection of better thermotolerant varieties which in turn decrease annual loss to agriculture.

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Poster

Infectious Diseases **P038**

Antibiotic Resistance Profiling and PVL Gene Detection among *Staphylococcus aureus* from Nasal Passage of Hospital Staff in Haripur and Abbottabad Cities of Pakistan

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A cross-sectional study was carried out on health care staff of different hospitals of Hazara Division in the year of 2015 to determine the nasal carriage of MRSA. The swab samples were collected from nasal passage of the healthcare professionals (including physicians, nurses, ward boys, operation theater staff etc.) who are working in the hospitals for one or more than one year duration. The samples were taken to microbiology lab of University of Haripur and *Staphylococcus aureus* was identified on the basis of various microbiological and biochemical assays as well as PCR. Antibiotic sensitivity testing against selected antibiotics was carried out using disc diffusion assay. Furthermore, multiplex PCR was performed to detect antibiotic resistant and virulent genes. Of all collected samples, 167 (81%) were found positive for *S. aureus*. When tested against eight selected antibiotics, large number of isolates showed resistance against them [Me= 96%, (n= 160), OFX= 34% (n= 57), E=77% (n= 128), DO=21% (n=36), MY=70% (n= 117), AMC= 59% (n= 99), CIP= 37% (n= 62), CRO=53% (n=88)]. Ninety six percent isolates were found resistant against methicillin. After performing PCR, 29% of isolates were found positive for pvl gene. Results of the present study reveal higher resistance against all antibiotics used, especially against methicillin. It may be concluded that due to improper use of different antibiotics, the resistant strains are emerging. Health care workers may be a source of infection for patients.

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Poster

Pharmaceutical Microbiology **P039**

Genetic Transformation of *Moringa oleifera* by *Agrobacterium rhizogenes* for the Enhanced Production of Valuable Anticancer Agents

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Agrobacterium rhizogenes is a gram-negative soil borne bacteria have its place in a group of phytopathogenic bacteria within order Rhizobiales and the causative agent of hairy root disease by Agrobacterium-mediated transformation. This transformation is carried out in plant cell by the integration of plasmid derived DNA of bacteria which cause hairy root disease. The bacterium infect at wounded site of the plant by transferring of a particular DNA segment (T-DNA) from the root-inducing (Ri) plasmid (pRi) of the bacteria. This T DNA carries a set of genes (rol genes) that encode enzymes for the phytohormone auxin control and cytokinin biosynthesis. *Moringa oleifera* (Moringaceae) which is also known as a ”miracle tree” contains functional bioactive compounds, so the aim of this study is to enhance its secondary metabolites by hairy root culture which is ideal biotechnological system to produce phytochemicals on large scale. Plant tissue culture of *Moringa oleifera* is established by using its seed as explant, the leaves grown *in vitro* will be wounded with *Agrobacterium rhizogenes* for the establishment of hairy root culture. Transgenic hairy root lines will be analyzed by polymerase chain reaction (PCR) for the conformation of tumor causing genes. Quantification of compounds will be determined by High performance liquid chromatography (HPLC), and determine its cytotoxicity activity. The result will be analyzed by SPSS by comparing the secondary metabolites production in transformed roots against non-transformed roots. The evaluation of cytotoxic effects in *Moringa oleifera* hairy roots will be carried out to validate its use as an anti-cancer drug and served as chemical factories.

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Poster

Medical Microbiology **P040**

Distribution of Metallo-beta-lactamase (MBL) producing Gram negative Bacteria in a Tertiary Care Hospital, Lahore

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Carabapnems are beta-lactam drugs and only treatment option against extended spectrum beta-lactamases (ESBLs). However, the appearance of carbapenemases, showing high hydrolysis profile towards all drugs, has compromised their efficacy over the last decade. Recent studies have testified global appearance of highly resistant variants of *Klebsiella pneumoniae* carbapenemases (KPCs), New Delhi Metallo-beta-lactamases (NDMs) and IMP carbapenemases belonging to Enterobacteriaceae. These agents can only be treated using inhibitors like EDTA, supplied in combination with imipenem or meropenem. In the current study, the selected imipenem resistant isolates were characterized and metallo-beta-lactamase (MBL) activity was confirmed using combination disc test and modified-Hodge test. The isolates were genotypically analyzed for the presence of blaIMP, blaOXA and blaSHV resistance genes by means of colony PCR. The study revealed highest frequency of MBLs among *Klebsiella spp*. and *Pseudomonas spp*., and highest age related drug resistance in age-group 31-45 years, with greater ratio in males. The isolates showed highest resistance against carbapenems, among which 70% were confirmed to be metallo-beta-lactamase producers, along with monabactams and cephalosporins. Colony PCR confirmed the presence genes blaSHV and blaOXA (ESBL activity) in few isolates confirming them to be MDRs. In addition, PCR for detection of MBL gene blaIMP-1 gene was optimized. In conclusion, the study demonstrates recent emergence and prevalence of multi-drug resistant MBLs among various clinical isolates. MBLs are being referred to as the beginners of post-antibiotic era. There is a need for the development of effective and sensitive phenotypic diagnostic tests and easy molecular detection techniques for the screening and monitoring of MBLs.

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Poster

Public Health **P041**

Hashish: A Silent Killer of Young Generation of Pakistan

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In Pakistani population ratio of Hashish addiction has been increased drastically since last few years. The most effected individuals are youngster especially from educational institutions. It is an alarming situation for higher authorities also.The present study project was designed to investigate the risk factors associated to hashish addiction. It was a questionnaire based descriptive cross sectional study. Different educational institutes (both private and public) of Lahore were visited for 1 year (Oct 2016-- Oct 2017) to collect required data of addicted individuals. A well-defined inclusion and exclusion criteria was applied during sampling. 300 samples from different educational institutes were collected. The age group for present study was (18-36). The significant outcomes of study were showing that addicted individuals at high risk of short term memory loss.High dose for long duration can affect the memory and responsible for impaired memory and neuropsychological decline, irritable behaviour, decreases concentration in daily tasks, decreases the capacity of learning and thinking and poor in education performance. High Potency with long term history, can also decrease the BMI and increase blood pressure causes hypertension ultimately lead to heart diseases. The addicted person was at the high risk to abuse other drugs (Alcohol, Cocaine). The addicted persons can be at high risk of road accidents during motor vehicle drive. The addicted individual at the high risk of relations clashes, withdraw from friends who do not use hashish. It is suggested that possible preventive measures should be adopted at personal as well as public levels, from both sides as parents and higher authorities'. In this way we can protect our young generation against this killer.

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Poster

Plant Microbe Interaction **P042**

Effect of *Bacillus* on the Growth of *Cicer arietinum L.* (Black Chickpea)

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Most of the global human population lives on a diet based on staple crops. In order to meet the challenges of providing food to the ever increasing population there is an urgent need to boost the production of staple crops. *Cicer arientinum L.* or black chickpea is one of the main staple crop. In recent years scientists have diverted their attention towards the ways by which they produce good quality maximum yield in shorter period of time. One of the best ways is by using microorganisms which act as biofertilizer. This research was conducted to check the effect of plant growth promoting rhizobacteria (PGPR) on the growth of black chick pea. The microorganism used was isolated from the rhizosphere of black chickpea. It was characterized as *Bacillus* sp. On provision of bacterial inoculum, all the parameters i.e. root length, shoot length, number of roots and leaves were enhanced. The biochemical testing was applied to both PGPR and the plant. The tests applied to PGPR were siderophore production, indole acetic acid production and phosphate solublization. While the tests applied to the plants include chlorophyll analysis, carbohydrate estimation, test for phenols and flavonoids. The biochemical testing was done in order to check the role of PGPR in promoting the plant growth. It will be helpful to use the bacterial strain *Bacillus* sp. as a biofertilizer.

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Poster

Environmental Microbiology **P043**

Screening and Optimization of Lipase from *Bacillus altitudinis*

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Lipase is an enzyme responsible for breaking lipids. It has vast applications in various industries like pharmaceuticals, textile, food, etc. To isolate lipase producing bacteria, ten samples were collected from different automobile workshops. From these samples, six bacterial isolates were obtained. Lipase screening methods confirmed lipase production by all six isolates. On the basis of lipase quantification, S2 showed maximum lipase activity (0.551 Units/min) and hence was selected. Morphological, biochemical and molecular characterization revealed it as Bacillus altitudinis. Its growth was induced by 1 % olive oil at 80 °C and pH 5. It produced extracellular lipase. The protein content of crude lipase was 350 mg/ ml. Partial purification of lipase by 80 % ammonium sulphate precipitation, dialysis, column chromatography and ion exchange chromatography showed protein content of 211, 98, 88 and 75 mg/ ml respectively. This purified enzyme showed maximum activity at 90°C, pH 5 and Ca ions. Maltose and olive oil were most suitable carbon sources for it. Its optimum activity was observed with wheat husk waste, yeast extract, sodium dodecyl sulphate (SDS) and Tween 20. It showed a protein band of 30 kDa on polyacrylamide gel electrophoresis (PAGE). Based on the properties of purified lipase, it can be speculated to have applications as a detergent.

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Poster

Environmental Microbiology **P044**

Isolation and Molecular Characterization of Lipase Producing Bacteria

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Lipase is an enzyme responsible for breaking lipids. It has vast applications in various industries like pharmaceuticals, textile, food, etc. To isolate lipase producing bacteria, ten samples were collected from different food stalls. From these samples, six bacterial isolates were obtained. Lipase screening methods confirmed lipase production by all six isolates. On the basis of lipase quantification, M4 showed maximum lipase activity (0.054 cm zone of lipolysis) and hence was selected. Morphological, biochemical and molecular characterization revealed it as *Bacillus stratosphericus*. Its growth was induced by 1 % olive oil at 45°C and pH 5. It produced extracellular lipase. The protein content of crude lipase was 220 mg/ ml. Partial purification of lipase by 80 % ammonium sulphate precipitation, dialysis, column chromatography and ion exchange chromatography showed protein content of 114, 107, 91 and 85 mg/ ml respectively. This purified enzyme showed maximum activity at 90°C, pH 9 and Na ions. Fructose and olive oil were most suitable carbon sources for it. Its optimum activity was observed with corn wastee, yeast extract, casein and Tween-20. Sodium dodecyl sulphate (SDS) and Triton X-100 inhibited its activity. It showed a protein band of 50 kDa on polyacrylamide gel electrophoresis (PAGE). Based on the properties of purified lipase, it can be speculated to have applications as biosurfactant in food industry.

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Poster

Microbiology & Molecular Genetics **P045**

Isolation and Characterization of Biosurfactant Producing Bacteria and their Potential Role in Oil Biodegradation

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Biosurfactants are heterogeneous groups having surface active molecules which are produced by the help of mico-organisms. They have potential to solubilize hydrocarbon contaminants and make them available for microbial degradation. From environmental point of view biosurfactants has role in bioremediation of oil contaminated sites, pollutants in sea and on land. They reduce surface tension of liquids. Biosurfactants find applications in different industries, the most promising among which is metal removal from contaminated wastewater/soil. They also helpful in oil spill removal in soil and aquifers which have harmful effect on environment. MEOR is a powerful technique to recover oil from reservoirs. Present proposal focus on isolation, screening and characterization of biosurfactant producing bacterial isolates and their potential application for the oil biodegradation. A variety of quantitative and qualitative analytical techniques will be applied for biosurfactant production. Oil degradation study will be carried out at field and lab scale. Plant microbe interaction experiments will also be conducted both in laboratory and at field scale under stress conditions. Besides this biosurfactant will be isolated from the isolated strains will be characterized. The strain which will give better results will be identified through 16S rRNA gene sequencing.The strains will be analyzed by checking their resistance against different metals, antibiotics and their antimicrobial activity against the pathogenic strains. Oil degradation will also be checked by decolorization of redox indicator of biosurfactant producing strains. The strains will also be introduced in the environment to remove oil contamination from industries as well as from soil and aquifers.

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Poster

Environmental Microbiology **P046**

Survey of Physical, Chemical and Microbiological Properties of Drinking Water from Various Areas of Lahore

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Water is vital to all life forms. There is no life without water. Human body is composed of two-third water. In order to maintain good health, proper proportion of drinking water is very important. Drinking water should be clear of any type of contamination. The biggest source of contamination in drinking water is pathogens. Water-borne pathogens results in water borne diseases. The purpose of this survey was to make finding of quality of drinking water in areas of central Lahore. It was observed that few areas including Choungi Amar Sadhu, Township and Greentown were highly contaminated with *T. coliforms*, *E. coli* and *Pseudomonas*. Although physical properties of water samples from these areas were satisfied showing no turbidity or odor. The chemical properties of all water samples were in accordance with the standards. The quality of drinking water should be checked in order to save the public from waterborne diseases.

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Poster

Environmental Microbiology **P047**

Growth Promotion of *Zea mays* by Exopolysaccharides Producing Bacteria from Azad Kashmir

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This study aim is isolation of Exopolysaccharides (EPS) producing bacteria from extreme environment and check their effect on growth enhancement of *Zea mays*. Seven bacterial strains were isolated from Tattapani hot spring, Azad Kashmir, Pakistan. All strains were EPS producers with high carbohydrate and protein content in their EPS but three strains (BE1, BN1 and BN3) exhibited high EPS production (14 g-15 g /100 ml) when screened on P-media. These strains had ability to solubilize phosphate and produce HCN when analyzed for plant growth promoting characteristics. Plant microbe interaction experiment was performed in field under natural conditions. Bacterial strains were used to inoculate *Zea mays* seeds because it is an important cereal globally. All inoculated seedlings displayed significant betterment in germination and growth parameters as compare to non-inoculated seedling. Inoculated *Zea mays* seedling's roots showed good colonization as compared to non-inoculated seedling when Alcian staining was performed. Auxin and soluble protein content of inoculated *Zea mays* seedlings were also significantly increased as compare to control. EPS production estimation and growth optimization of bacterial strains were evaluated by varying different parameters i.e. pH, temperature, carbon and nitrogen source. Phylogenetic analysis based on 16S rRNA sequencing revealed that bacterial strains BE1, BN1, BN3 were closely related to *Ochrobactrum intermedium, Bacillus pumilus* and *Enterobacter ludwigii*, respectively. This study revealed that all isolated bacterial strains are excellent EPS producers and have excellent capability to enhance yield of *Zea mays* and make the crop resistant from unsuitable environment.

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Poster

Biochemistry, Biofilms **P048**

Antimicrobial Significance and Bioactive Agent in *Areca* Nuts

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The effect of *Areca catechu* (nut) on oral microflora was studied and analyzed in this research work. A total of 50 samples were collected from people who were in the habit of chewing paan on a daily basis. From these samples, 3 bacterial isolates were obtained which were labelled as ML-1, ML-2 and ML-3 respectively. After Gram staining, the strains were visualized as Gram positive cocci and rods and biochemical characterization revealed these strains belonging to *Streptococcus sp., Staphylococcus sp*. and *Bacillus spp.* Three extracts (ethanol, methanol, distilled water) were made to check the antimicrobial activity which was affirmed by well diffusion and disc diffusion methods. Thin layer chromatography identified several Rf values in ethanolic and methanolic extracts, one of which was quercetin. Its detection was further affirmed by High Performance Liquid Chromatography (HPLC). Quercetin was the only compound that exhibited antimicrobial activity. Biofilm of the bacterial isolate ML-1 which was previously cultured and grown in a microtiter plate was inhibited in the presence of all extracts. Results demonstrated significant demolishing activity by indicating the effectiveness of the ethanolic, methanolic and aqueous extracts. These extracts can be used in formulating mouth washes but further research work is needed to check the antimicrobial effect of Areca nut on different microorganisms, including pathogenic and non-pathogenic bacteria.

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Poster

Food Microbiology **P049**

Microbiological Quality and Antibiotic Resistance Studies of Bacteria Isolated from Raw, Pasteurized and UHT Milk

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Milk spoliation with antibiotic resistance microscopic organisms can be a noteworthy risk to general well-being, as the antibiotic resistance elements can be exchanged to other pathogenic microorganisms perhaps compromising the treatment of extreme bacterial diseases. This study was directed to examine the microbial quality and anti-microbial resistance of bacterial isolates. Total 100 milk samples containing 60 raw and 30 UHT and 10 pasteurized were arbitrarily collected from milk sellers, milk shops, and markets sold in different regions of Lahore. The microbiological characteristics of raw milk test were poor when contrasted with Pasteurized milk while the vast majority of the UHT milk tests show amazing results. The high level of isolate numbers and antibiotic resistant of bacterial isolates especially in raw and pasteurized milk represents a poor keeping quality of milk, indiscriminate and regular use of antibiotics which has now put the users of milk at risk of being infected. This recommends the requirement for enhanced hygienic practice at all levels of milk generation and the regular utilization of anti-infection agents ought to be prohibited. On the other UHT milk which sees as a promptly drinkable beverage must not be acquired following three months from creation because of microbial substance in milk test expanded by significant sum.

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Environmental Microbiology **P050**

Isolation and Molecular Characterization of Protease Producing Bacteria

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Proteases refer to a group of enzymes whose catalytic function is to hydrolyze proteins. Protease has various and huge applications in commercial and industrial areas like detergent industry, cosmetic etc. For isolation purpose, 10 samples were collected from various dairy farms. From these soil samples, 6 isolates were obtained. Confirmation of protease producing bacteria was done by different screening methodologies. For quantification purpose, FA1 showed maximum proteolytic activity (0.4 cm zone of proteolysis), hence was selected. Bacillus cereus was identified by following morphological, molecular and biochemical characterization. 1 % casein at 50°C and pH 9 enhanced the production of extracellular protease enzyme. 0.346 mg/ ml was protein content of crude protease. Partial purification of protease enzyme by following methods, ammonium sulphate precipitation, dialysis, column chromatography and ion exchange chromatography showed 114, 107, 91, and 85 mg/ ml protein content respectively. Purified protease enzyme indicated maximum activity at 90°C and pH 9. Skim milk and casein were most suitable carbon source for protease. Its optimum activity was observed with corn waste, yeast extract, casein and Tween-20. Proteolytic activity was inhibited and stopped by sodium dodecyl sulphate (SDS) and Triton X-100. On polyacrylamide gel electrophoresis (PAGE) it showed protein band of 50 kDa. On the basis of its properties like stability and reliability it would be selected and considered as most applicable in leather processing, detergent industry and food industries.

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Poster

Environmental Microbiology **P051**

Screening and Optimization of Lipase from *Bacillus* sp.

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Proteolytic enzymes are ubiquitous in occurrence, being found in all living organisms, and are essential for cell breaking proteins. It has vast applications in various industries like pharmaceutical, detergent cosmetic, and food etc. To isolate protease producing bacteria, ten samples were collected from different meat stalls, selling raw meat. From these samples, six bacterial isolates were obtained. Protease screening methods confirmed protease production by all six isolates. On the basis of protease quantification A4 showed maximum protease activity (0.6 cm zone of proteolysis) and hence was selected. Morphological, biochemical and molecular characterization revealed it is *Bacillus amyloliquifaciens*. Its growth was induced by 1 % casein at 30 ÂºC and pH 6. It produced extracellular protease. The protein content of crude protease was 0.182 mg/ ml. Partial purification of protease by 80 % ammonium sulphate precipitation, dialysis, column chromatography and ion exchange chromatography showed protein content of 211, 98, 88, and 75 mg/ml respectively. This purified enzyme showed maximum activity at 90 ºC, pH 5 and Ca ions. Casein and skim milk were most suitable carbon sources for it. Casein, yeast extract, sodium dodecyl sulphate (SDS) and Tween 20 were used to determine its optimum activity. A protein band of 50 kDa was showed by it on polyacrylamide gel electrophoresis (PAGE). According to properties of purified protease, it can be speculated to have applications in food industry.

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Poster

Plant-Microbe Interactions **P052**

Effect of Plant Growth Promoting Rhizobacteria on the Growth of *Cicer arietinum*

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Cicer arietinum or chickpea is a staple food crop which is considered as an important food legume plant in sustinable agriculture system. In this research work, the effect of plant growth promoting rhizobacteria (PGPR) on the growth of chickpea was investigated. For this, microorganisms were isolated from rhizosphere of chickpea plant. The cultural, biochemical and molecular characterization revealed it as *Bacillus velezensis*. The chickpea seeds were grown in the presence and absence of *B. velezensis* culture. All the parameters of the plant i.e. root length, shoot length, number of roots and number of leaves were enhanced in the presence of bacterial inoculation. The biochemical tests for PGPR including siderophore production, indole acetic acid production and phosphate solubilization tests and for plants including chlorophyll analysis, carbohydrate estimation, test for phenols and flavonoids will be performed to have an insight into role of PGPR in promoting the growth of plant. It will help to use *B. velezensis* as a biofertilizer.

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Poster

Nanobiotechnology **P053**

Comparative Cytotoxicity Study of Selenium Nanoparticles Synthesized by Utilizing Bacterial Cells and Plant Extracts

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Conventional Nanoparticles synthesis through physical compound and physical combination strategies may bring about the synthesis of nanoparticles in mixture form not in pure form with poor morphology and low product yield and these methods are environment un-friendly. Recently, ecofriendly synthesis of nanoparticles has developed as an attractive option over conventional strategies for nanoparticle synthesis This biosynthesis includes green science based approach that utilizes unicellular and multicellular biological entities. In this study three different bacterial strains Vibrio cincinnatiensis (W-2) obtained from Waste water of tannery treatment plant; Kasur, Pakistan, *Bacillus subtilis* (W-5) isolated from Waste water of Sugar mill:Shakar Ganj and Bacillus licheniformis were used for nanoparticle synthesis. For green synthesis orange peel extract, banana peel extract , fenugreek seeds, cardamom, cinnamon, clove and black cumin were used. Nanoparticles were obtained in all cases except clove and fenugreek seeds. These selenium nanoparticles were characterized by UV vis spectrometry, Scanning Electron Microscopy and Energy dispersive X-ray analysis. Toxicity level of SeNP were compared with sodium selenite and it was observed that SeNP were less toxic than its original salts at respectively same concentrations. Effect of NPs on growth features of *Solanum tuberosum* were observed. In general, all types of nanoparticles have positive effects on growth features over respective control ((Na_2SeO_3 100 µg/mL) except SeNP synthesized by cinnamon extract.

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Food Microbiology **P054**

Characterization of Bacterial Strains from Rotten Fruits Treated with Harmful Preservatives

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This study was designed to isolate bacterial strains from fruits treated with different preservatives which are actually harmful for the human health. Rotten fruit samples were collected from different shops of Moon Market and Neelam block, Allama Iqbal Town, Lahore and used to purify bacterial strains by growing on simple N-agar medium. Biochemical characterization was performed by different tests including gram staining, catalase, mannitol salt agar, glucose fermentation, fructose fermentation and nitrate reduction test and were characterized as *Bacillus sp, Staphylococcus aureus, Micrococcus varians, Staphylococcus epidermidis* and *Staphylococcus* saprophyticus. Bacterial strains were further subjected to additional tests like HCN, H_2S production, metal resistance and antibiotic sensitivity tests and most were positive for these tests. Many bacterial strains were resistant to antibiotics and high doses of most metals specially mercury. In conclusion, due to the use of high doses of mercury for the storage of fruits, microorganisms evolved resistance. It is an urgent need to take other safe measures for the storage of fruits.

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Poster

Environmental Microbiology **P055**

Optimization of Bioplastic Production by *Bacillus* sp.

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Bioplastics or biodegradable plastics are biodegradable polyesters that are eco-friendly. They are produced and deposited as cytoplasmic inclusions inside bacteria. In this research work, an attempt was made to isolate and characterize bioplastic producing microorganism from the contaminated environment and to optimize the growth of bioplastic. For this, soil samples were collected from industrial effluent contaminated environment. Out of thirteen soil samples, 6 bacterial strains were isolated. They were screened for their bioplastic producing ability by using chloroform and SDS methods. Out of six strains, one strain showed maximum bioplastic production named as Z1. The cultural, morphological, biochemical and molecular characterization revealed it as Bacillus sp. It produced 10 mg/ml bioplastic after 18 hours incubation at 37°C which can be observed by Sudan black staining. Its bioplastic production was optimized using wastes as carbon sources including vegetable peels and agricultural wastes. The bioplastic produced by Z1 showed stability at 90°C and pH 7. The thermostability render its use in packaging industry.

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Poster

Biofilms **P056**

Effect of Paan Leaf Extracts on *Bacillus subtilis* Biofilm

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Piper betel (paan leaf) has been used as mouth freshener in many cultures of the world along with *Areca nuts* (nuts), *Areca catechu* (katha), lime and cardamom. In this research study, the effect of paan leaf on biofilm formation of *Bacillus subtilis* was studied. For this, the dental swabs were collected from 100 individuals having healthy teeth. Three bacterial isolates were initially isolated, but only one isolate labelled as SN-1 showed intact biofilm formation. It was selected. The morphological, biochemical and molecular characterization revealed it as *Bacillus subtilis*. Paan leaf extracts were prepared in ethanol, methanol, water and chloroform. The antimicrobial activities of all extracts were studied by well diffusion and agar diffusion methods. Only 50 % ethanolic and methanolic extracts of paan leaf exhibited antimicrobial activity by showing large zones of inhibition in ethanolic extract followed by methanolic extract i.e. 0.9 cm and 0.5 cm respectively. Thin layer chromatography (TLC) of ethanolic and methanolic extracts showed the presence of quercetin, which has Rf value 0.53. The presence of quercetin was confirmed by High Performance Liquid Chromatography (HPLC). HPLC further showed the presence of other two phenolic acids, coumaric acid and benzoic acid in ethanol and methanol extract respectively. Formation of SN-1 biofilm was 50 % inhibited by both the extracts. These extracts also helped in demolishing 63 % established biofilm which showed the importance of the antibiofilm property of the paan leaf due to presence of quercetin, coumaric acid and benzoic acid. Paan leaf being the cheaper source of antibiofilm agent can be used in mouthwash formulation to get rid of plaque / biofilm forming pathogenic and non-pathogenic bacteria.

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Poster

Environmental Microbiology **P057**

Isolation and Screening of Copper Resistant Bacteria from Industrial Effluents

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Industrialization has introduced a huge amount of heavy metals in our environment. These metals are toxic for all life forms. Microorganisms surviving in such toxic environments have the ability to uptake metal and convert them into less toxic forms. In this research work, 5 soil samples were collected from drainages of Rohi-Nala Lahore and industrial areas of Kotlakhpat, Faisalabad and Gujranwala. Out of thirteen bacterial isolates obtained from five samples, only one SSR-1 was selected as it showed resistance to multiple msetals including copper sulphate ($CuSO_4$), nickel sulphate ($NiSO_4$), zinc sulphate ($ZnSO_4$), mercuric chloride ($HgCl_2$) and potassium chromate ($K_2Cr_2O_7$). The optimum growth conditions of SSR-1 were 37°C, pH 7 and 1 mM phosphate. Copper (Cu) enhanced the growth of SSR-1 up to 8 hours. The maximum Cu uptake (6 mg/ L) was observed in first 5 hours whereas intracellular amount was 4.8 mg/ L. The remaining 1.2 mg /L remained attached to outer cell wall. In pilot scale studies, about 67 % Cu was removed at 4^{th} day which was decreased to 62 % at 8^{th} day. Presence of Cu induced metallothioneins of 25 kDa on polyacrylamide gel electrophoresis (PAGE). Molecular characterization done by 16S rRNA sequencing showed SSR-1 was *Bacillus velezensis*. It was found to be potential candidate for heavy metal bioremediation.

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Poster

Medical Microbiology **P058**

Microbial Flora Analysis of Urinary Tract Infection in Patients Suffering from Nephrotic Syndrome in Lahore, Pakistan

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Nephrotic syndrome (NS) is a common renal disorder. If left untreated, results in urinary tract infection (UTI) by bacteria. In this study, 500 samples were collected from nephrotic syndrome patients suffered from UTI. Midstream urine was collected in sterile container and processed by standard microbiological methods. Females were found to be more affected (61 %) as compared to males (39 %). *E. coli* showed the highest frequency (71 %) followed by *Klebsiella* (10 %), *S. aureus* (10 %), *Enterococcus* (3 %), *Streptococcus* (3 %) and *P. aeruginosa* (3 %). Gram positive bacteria showed sensitivity to vancomycin, linzolid, ampicillin, nitrofurantoin, cephradin and penicillin. Gram negative bacteria showed sensitivity to imipenem, meropenem, gentamicin, amikacin, sulfzone, tazocin, cefruxime polymyxin B, colistin and nitrofurantoin. Precautionary measures should be strictly followed to avoid infections and complications in urinary system by proper cleanliness and awareness of sex education in adults.

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Poster

Antimicrobial Agents & Chemotherapy **P059**

Evaluation of Phytochemical Constituents, Antioxidant and Antibacterial Activities of Few Medicinal Plants, AJK

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Use of plants to cure different ailments is in practice from centuries. In Pakistan, Tatta Pani region of Azad Jammu and Kashmir (AJK) has great biodiversity of flora due to its ideal climatic conditions and diverse topographical features. In current study, phytochemical constituents, antioxidant and antibacterial activity of *Achillea millefolium* (Kangi), *Sisymbrium irio* (Khoob Kalan) and *Viola canescens* (Banafsha) was performed. Presence of carbohydrates, coumarines, flavonoids, phenols, phlobatonins and saponins was detected in all ethanolic extracts of plants. DPPH, reducing power and phosphomolybdate assay were conducted to find out antioxidant capacity of the plant extracts. Antioxidant activity of *V. canescens* was found 86% which is maximum among selected plants. Antibacterial activity of plant extracts was determined by agar well diffuion assay against *Bacillus* and *Pseudomonas* strains. Each of the three selected extracts showed antibacterial activity by agar well diffusion assay, but *V. canescens* also showed 7 mm zone of inhibition against *Bacillus* by disc diffusion assay. Minimum inhibitory concentration of *V. canescens* against *Bacillus* came out 1.25 mg/ml. Thin layer chromatography of *V. canescens* showed 15 spots. Eight out of these fifteen spots showed antibacterial activity against *Bacillus*. Hence these extracts can be used by pharmaceutical industries to develop drugs to cure inflammatory diseases.

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Poster

Environmenatal Microbiology **P060**

Growth Optimization of Chromium Resistant Bacteria from Tannery Effluent

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The major use of chromium in industry tend to be major cause of environmental pollution. Chromium-resistant bacteria are those microorganisms that could reduce toxic Cr (VI) to less toxic Cr (III). Twenty isolates able to grow on LB agar containing 500 µg/mL of Cr (VI) were isolated from tannery effluent of Kala Shah Kaku. Ten bacterial isolates MAK01, MAK02, MAK03, MAK04, MAK05, MAK06, MAK07, MAK08, MAK09, MAK10 were selected due to their high resistivity of Cr (VI) which was found to be 2000 µG/ML. These strains could remove about 90% of Cr (VI) after 24 hours of incubation at an initial concentration of hexavalent chromium of 100 µg/mL. Further, the optimization of various environmental factors like pH, temperature, and incubation time on Cr (VI) removal and bacterial growth were studied.

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Poster

Environmental Microbiology **P061**

Characterization of Cr and Ni Resistant Bacteria for Plant Growth Promotion

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Hexavalent chromium and nickel are carcinogenic, mutagenic heavy metals that cause many health hazards to all living beings include humans, plants and animals. Some bacteria are capable of resisting high concentrations of hexavalent Cr by converting it into trivalent form, which is non-toxic; similarly some bacteria can remove toxic nickel from the environment. The objective of present study was to isolate bacterial strains, resistant to chromium and nickel as well as proficient to promote plant growth. The bacteria isolated from metal contaminated soil, where industrial effluents are discharged, selected on the basis of their metal resistance and plant growth promoting properties and characterized phenotypically and genetically. The extent of metal resistance and plant growth promotion, phosphate solubilization, production of IAA, siderophore and ACC Deaminase of selected isolates was determined. The DNA of selected strains was isolated, amplified and sequenced for 16s ribosomal identification. Seeds of *Triticum aestivum* treated with bacterial strains were grown in pot having chromium (k_2CrO_4) and Nickel ($NiCl_2$) concentrations 0, 200, 500 µg/kg of soil. The Strains resisted Cr and Ni metal stress in media 2500-4000 µg/ml and 500 µg/kg in pot soil. The strains showed growth at alkaline environment up to pH 11, hypothermophillic, grew at 55°C temperature and also halophillic, tolerated salt concentration upto 5 % (w/v). The Bacterial strains enhanced the growth of Triticum aestivum by increasing its shoot length, number of tillers, spike length, number of spikelets, number of grains and seed weight. The effect of inoculum of bacterial strains on the growth of Spinacia oleracea was also tested via pot trials. The isolated bacterial strains can be used for plant growth promotion as well as environment cleanliness from toxic heavy metals specifically Cr and Ni.

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Poster

Environmental Biology, Biofuels, Photosynthetic Prokaryotes

P062

Arsenic Tolerance and Redox Potential of Purple Non-Sulfur Bacteria Isolated from Industrial Effluents and Paddy Field

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Due to alarming rise in arsenic concentration in water bodies, there should be eco-friendly methods to detoxify the toxic metal into less toxic form. Purple non-sulfur bacteria (PNSB) are one of those eco-friendly candidates; they can utilize different organic and inorganic sources to grow and can employ different strategies to detoxify certain toxic metals. For this purpose, five PNSB were isolated from industrial effluents and paddy fields. They were checked for arsenic resistance as well as for arsenic reduction. The selected bacteria were identified as *Rhodospirillum rubrum* strain code Q3B, *Rhodospirillaceae sp.* strain code Q3C and *Rhodospirillum rubrum* strain code P.F.2 (i) through 16S rRNA gene sequencing. Highest arsenic biosorption and reduction was shown in anaerobic conditions. The carotenoid content was also estimated as it serves as a nutritional source for aquatic organisms. PNSB are also known to be potential source for bio-hydrogen production which can be used as fuel. Among these isolates, Rhodospirillaceae sp. strain code Q3C and Q1B were found to produce gas (hydrogen) which was detected by displacement method. These bacteria can further be optimized for hydrogen production along with As-bioremediation.

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Poster

Medical Microbiology **P063**

Prevalence of Multi-Resistant Bacteria and their Antibiotic Resistance Pattern Isolated from Hospital Wastewater Samples

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Antibiotics are natural or synthetic compounds used to treat infections caused by pathogenic bacteria. However, antimicrobial resistance has become global threat negatively affecting the health and economics of a country. In present study, prevalence of multi-resistant bacterial strains from hospital waste water samples was determined. Samples were collected from different waste water hospital premises across Multan. Bacterial resistant pattern against a panel of fourteen antibiotics were recorded as zone of inhibition in millimeters using standard disc diffusion method. Later, the isolates were biochemically characterized. A total of 45 bacterial isolates were isolated from five waste water samples using plate dilution method. Out of 45, 13 (29%) were resistance against more than one class of antibiotics. Majority of the bacterial pathogens were highly resistance against trimethoprim and highly sensitive against linezolid. Twenty five bacterial strains were *Bacillus species* (55.5%) and other were *Staphylococcus species* (17%), *Streptococcus species* (8%), *Micrococcus species* (11%) and other species (8.5%). Hospital based resistant bacterial isolates demands effective treatment plants to treat hospital wastes before their disposal into general waste lines. Besides these, public awareness is highly demanded at personal and community level.

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Poster

Environmental Microbiology **P064**

Prevalence and Antibiogram of Bacteria Isolated from Hospital Environmental Soil Samples

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Misuse along with overuse of antibiotics has increased the antibiotic resistance. This study was performed to determine the prevalence of resistant bacterial isolates in hospital soils and their antimicrobial resistant profiles. Random soil sampling technique was used to isolate bacterial strains by plate dilution method. Isolates were screened against a panel of 14 known antibiotics using Kirby Bauer Disc Diffusion method. Furthermore, strains were biochemically identified following Bergeys Manual of Determinative Bacteriology. Bacterial isolates were highly resistant against ampicillin with a frequency of 73% in contrary to ciprofloxacin against which 2% isolates were resistant. Other notable antibiotics against which bacterial isolates were resistant were fusidic acid (64%), trimethoprim (55%) and oxacillin (50%). Interestingly, all isolates were sensitive to chloramphenicol, gentamicin and streptomycin. It was observed that resistant pattern varies from one location to another location i.e. among 3/4 samples, most resistant isolates were against ampicillin (88%). Only in 1/4 sample, oxacillin (14%) was highly resistant. *Bacillus sp*. being the most common bacterial isolates, were highly resistant against ampicillin (91%) while *Staphylococcus sp*. were against tetracycline (78%). Presence of drug residue in the hospital waste leads to the development of drug resistance. A significant rise of MDR in hospital waste demands effective management against them.

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Poster

Chemotherapy **P065**

Screening of Antioxidant Properties of Various Solvent Extracts of Edible Plants

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Plants have been used to cure or lessen symptoms of illness in humans throughout the history. Eighty percent of the total population depends on restorative plants for their essential medicinal services. The aim of this study was to demonstrate the bioactivities of edible fruits along with their preliminary phytochemical analysis. Three edible plants; *Mangifera indica*, *Malus domestica* and *Triticum aestivum* were examined using methanol, ethanol, acetone, dimethyl sulfoxide and water as extraction solvent. The preliminary qualitative and quantitative phytochemical screening was performed using standard procedures. In *Mangifera indica*, extracts in DMSO, in *Malus domestica*, extract in acetone and in *Triticum aestivum*, extracts in ethanol showed presence of comparatively high phytoconstituents contents. Furthermore, peel and pulp extracts of *Mangifera indica* and *Malus domestica* did not varied compared to whole plant extracts. In *Triticum aestivum*, the methanolic extract and ethanolic extract showed the highest reducing activities. Potential scavenging activity was also found by DPPH scavenging activity test. It was observed that extracts in methanol and ethanol showed the highest inhibition activity while extracts in acetone, water and DMSO did not show good scavenging activities. A strong correlation was observed between phytochemical constituents and antioxidant activity. Better understanding of nutritional value of edible plants increase popularization among their consumers.

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Poster

Chemotherapy **P066**

Analytical Profiling of Phytochemicals and Antioxidant Activities of Different Edible Plants

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Presence of bioactive compounds in medicinal plants has open new insights into the green technology. The present study aims to analyze the potential of edible plants. In the present study qualitative and quantitative phytochemical analysis of methanol, ethanol, DMSO, acetone and water extracts of three edible plants- *Momordica charantia* (Karela), *Brassica oleracea* (Cabbage) and *Abelmoschus esculentus* (Lady Finger) were performed according to the standard methods. The highest activities were observed in extracts with methanol followed by ethanol, dimethyl sulfoxide, acetone and water. Among the plant species, *Momordica charantia* showed the promising phytochemical activities while *Abelmoschus esculentus* showed the highest antioxidant activity of 2.58 ±0.17 nm at 100 µg/ml. Total phenolic contents and flavonoid contents varies from 4.31-2.13 mg GA/g and 2.24 to 0.71 mg RU/g respectively. The results suggests that studied plant extracts are potent and can be further utilized.

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Poster

Human Genetics **P067**

Effect of ALDH2 RS671 Polymorphism on Coronary Artery Disease and *In-Silico* Study of ALDH2 Gene

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Coronary artery disease (CAD) is the leading cause of disability and death worldwide particularly in South Asians due to high prevalence of smoking, diabetes, fat rich diet and other factors especially in Pakistan. Narrowing of coronary arteries by any above factor, enough oxygenated blood is not supplied to heart. The aim of the current study was to analyze ALDH2 Glu504Lys polymorphism in the local subjects, to determine their genotype and allele frequency and in-silico study of ALDH2 gene. For in-silico study many tools e.g. Oligoanalyzer, Neb cutter, Gene card, PDBsum, and VMD were used to check various features of primers, enzyme and selected gene before practical work. Blood samples were collected and genomic DNA was isolated, quantified and amplified using specific primers. RFLP (restriction fragment length polymorphism) was used to investigate genotyping of ALDH2 rs671 polymorphism (Glu504Lys) using HincII enzyme (HindII) for the digestion of amplified PCR products. Minor allele frequency for controls was higher (0.283) than for cases (0.130) indicating that possibly the minor allele A is acting as a protective allele in our population. The previous studies have mostly been done in Chinese populations that have genetic makeup distinct from our population therefore another reason for contradictory results of the current study can be the regional differences as to the best of our knowledge we are first to report this polymorphism (rs671 Glu504Lys) in Pakistan. So sampling should be done from different provinces of Pakistan to analyze environmental influence on rs671 polymorphism in ALDH2.

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Poster

Industrial Microbiology **P068**

Comparative Analysis of Antibacterial and Antitumor Activity of the Endophytes and Plant Extracts of *Carica Papaya Linn.*

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Microbes have always been the noteworthy source of antibiotics, enzymes and various other compounds. The emerging issue of multidrug resistance has increased the demand for mining out novel sources of antimicrobial agents. Thus, researchers are now trying to explore the underexplored microbial resources for getting new therapeutics. In this study we isolated 42 endophytic bacteria including; 22 endophytic actinomycetes and 20 *Bacillus* strains from different parts (roots, shoots and leaves) of *Carica papaya Linn.* After performing biochemical and physiological characterization of selected endophytes, screening for antimicrobial activity of crude extracts against multidrug resistant pathogens; *Staphylococcus aureus* (MRSA), *E. coli, Proteus* and *Pseudomonas aeruginosa* was done. *In vitro* antitumor activity at the lowest concentration of 0.1 mg/ml was determined by MTT assay against colorectal carcinoma cell line (HCT 116, ATTC CCL-247). Chemical profiling of the active extract was done by TLC and HPLC. The super active strains were identified up to specie level through 16S rRNA gene analysis. Antimicrobial analysis proposed that the plant extracts were more potent against MRSA and *Bacillus*, in comparison to the extracts of endophytic bacteria. The extracts also exhibited notable in vitro antitumor activity showing maximum inhibition up to 80% by extracts of plant parts comparatively higher than 78% and 70% of endophytic *Bacillus* and endophytic actinomycetes respectively. Our findings suggested that endophytic bacteria and plant tissue extracts of medicinal plant *Carica papaya* are promising producers of antimicrobial and antitumor compounds. The purification and identification of active compounds from these sources may yield some useful drug candidates.

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Poster

Clinical Microbiology **P069**

Enterococcins: Isolation-Purification- Characterization and their Bioactivity against Biofilm Producing Indigenous MDR Bacterial Strains

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Ever since the use of antimicrobial drugs, microorganisms have gained resistance against almost all generations of antibiotics, thereby, resulting the emergence of multidrug resistant (MDR) strains. Biofilm formation is one of the characteristic features of MDRs. Biofilm formation constitutes a crucial challenge in hospital settings and is a nuisance in infectious disease management. Some of the bacteria are able to produce protein (like) substances that inhibit growth of the related microorganisms. In this regard, we pursued a research project involving isolation of clinical Enterococcus strains from urinary tract infection patients. The cell free neutralized supernatant (CFNS) was prepared and which was tested against varied Gram positive and Gram negative clinical bacterial strains. The corresponding bacteriocinogenic activity of Enterococcins was determined by stab-overlay, disc diffusion and agar well assays. Protein was precipitated by acetone, chloroform (separately) and concentrated by 'Rotary evaporator'at 40 ◦C. Estimation of proteins was carried out by Nanodrop 'Protein 280' and Bradford method. Enterococcin was found to be active against Staphylococcus spp, *Streptococcus spp, Listeria spp, Escherichia colispp, Pseudomonas spp, Salmonella spp* and other Enterococcal strains. (Associated with pathogenicity). High concentration of protein was found out in sample precipitated by acetone i.e. 33.580 ng/µl. Many of the representative MDR Gram positive and Gram negative species were susceptible to enterococcins. As efficacy of conventional antibiotics is losing luster by the passage of time. Enterococcins may be presented as a substitute of antibiotics. This will also help eliminate the anticipated adverse side effects caused by classical antibiotics because *Enterococcus spp.* Constitute 'normal flora'of gastrointestinal tract (GIT) and also rated a 'Probiotic bacteria' (GRAS). Therefore, enterococcins can help in minimizing the frequency of infectious diseases by combating biofilm producing MDR strains.

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Poster

Human Genetics **P070**

Long QT Syndrome: Diving in Genetics from Pakistani Perspective

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The long QT syndrome (LQTS) is a cardiac channelopathy which prolong the contraction and relaxation of ventricular part of the heart. This type of abnormality may lead to fainting, syncope, seizures and sudden cardiac death because of ventricular fibrillation. This condition is termed for the appearance of prolong QT interval on electrocardiogram. The molecular biology had made impressive progress in 1990s. The new genetic techniques developed especially association analysis and DNA sequencing have a potential for identification of disease gene(s) and their mutations. Association studies are use case-control design in which controls are family members of cases. This is a familial-based association study which can get much closer to identifying disease variants and help to address issues of population stratification. Currently there are fifteen genetic forms of LQTS from LQT1-LQT15. These gene mainly encode the cardiac ion channel. The most common cause of LQTS is mutation in KCNQ1 (LQT1), KCNH2 (LQT2), and SCN5A (LQT3) genes affect about 75% of patients with LQTS. It arises from mutation(s) that prolong the QT duration of lower chamber (ventricles) of heart due to change in the ion channel protein. The normal QT interval is between 350 to 440 milliseconds. Patients with a clinical or ECG presentation of long QT syndrome need genetic testing to identify the mutation(s). For this study, the samples will be collected from affected families for screening the reported loci through PCR amplification and sequencing. There are two studies about long QT syndrome in Pakistan which are based on ECG, without genetic analysis. Therefore, the aim of the study is familial based association study of long QT syndrome in Pakistani families based on genetics, prevalence and association of SNPs in long QT syndrome.

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Poster

Human Genetics **P071**

Studies on the Role of NS3 And NS5A Non-Structural Genes of HCV Genotype 3A Local Isolates in Apoptosis

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One of the important causes of liver diseases and hepatocellular cancer is hepatitis C virus that belongs to family Flaviviridae that causes both acute and chronic hepatitis. Among non-structural genes of HCV, NS3 and NS5A plays an important role in apoptosis. NS3 and NS5A genes of HCV interact with the p53 tumor suppressor gene differentially. To analyze the interaction of NS3 and NS5A genes of HCV-3A genotype with p53 gene, sub-genomic HCV replicons harboring NS3 and NS5A genes were prepared. Huh-7 cells lines stably expressing NS3 and NS5A genes were generated. The stable cell lines were confirmed by western blot, RT PCR and immunofluorescence assay. Hepatitis C virus NS3 and NS5A expressing cell lines were transfected with p53 expressing clone. Results showed that NS3 and NS5A both interact with p53 by down regulating expression of p53 gene. In HCV sub-genomic harboring cells interaction between NS3, NS5A and p53 was observed consistently. Suppression in expression of p53 gene by NS3 and NS5A was observed significantly as compared with NS3 and NS5A-negative control huh-7 cells. The above results suggest the possibility that both non-structural genes (NS3 and NS5A) of HCV play an important role in the hepatocarcinogenesis of HCV by interacting directly or indirectly in different manners with p53 gene.

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Poster

Plant-Microbe Interaction **P072**

Growth, Physiological and Antioxidative Responses of Tomato to Early Blight Pathogen

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Early blight disease caused by notorious *Alternaria solani* poses a serious threat in tomato growing areas with significant yield losses. The current study was conducted to screen out twenty five tomato genotypes (*Solanum lycopersicum L.*) against *A. solani* and investigate the physiological and biochemical changes with the involvement of total phenolics and antioxidant enzymes in infected plants. Tomato seedlings were transplanted and artificially inoculated with pure culture of *A. solani*. On the basis of disease severity (disease incidence, percent severity index and mortality), twenty five genotypes were categorized into six groups, as highly resistant, resistant, moderately resistant, tolerant, susceptible and highly susceptible. Pathogen infection on tomato genotypes led to alternations in host growth, physiology and biochemistry. Growth attributes were more significantly decreased in highly susceptible genotypes with the substantial increase in growth inhibition index. Total phenolics, total protein content and the activities of antioxidants (catalase, peroxidase and polyphenol oxidase) were highly up-regulated in resistant groups than in susceptible groups linked with the induction of resistance in former group. Screening of tomato genotypes against early blight disease proved an efficient and effective strategy to identify most appropriate genotype for Pakistan agro-climatic conditions to have good tomato yield and could be evolved by incorporating resistant traits into susceptible genotypes by using them in the breeding program.

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Poster

Microbial Genetics **P073**

Isolation and Characterization of Tannase Producing Bacteria from Local Environment

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Tannases are an important group of biotechnologically relevant enzymes which have numerous of industrial applications. Microbial tannases are promising candidates for producing environment friendly, low cost enzymes. In this study, we have isolated tannic acid tolerant bacteria also capable of producing high yield of tannases, from indigenous environment. Bacterial isolates, which tolerate up to 3.5 % tannic acid, were further screened for tannase production. Among tannic acid tolerant bacteria; WK-1 (*Bacillus methylotrophicus*), WK-A2 (*Bacillus tequilensis*), WK-A3 (*Bacillus subtilis*), WK-4 (*Bacillus pumilis*) showed the maximum tannase activity as estimated by their crude enzyme extract. Extracellular enzyme production was observed in some isolates. Biochemical characterization of the partially purified enzyme was determined. Tannase enzyme showed stability with little reduced activity under extreme conditions. Protein concentrations were also estimated in partially purified enzyme extracts. Among all four bacterial isolates designated as WK-1, WK-A2, WK-A3 and WK-4 showed the maximum activity through their crude enzyme extract. All isolates showed ability to grow in a diverse range of pH and temperature. Growth curves of these organisms showed that growth was slower in the presence of tannic acid. In controlled environment (N-broth), organisms showed a shorter lag phase and reached at log phase within 4-12 h.Tannase activity can be cell associated or extracellular. In the present study, it was found that most enzyme production was extracellular in isolated bacteria but they also showed very little cell associated activity too. The protein banding pattern showed an extra protein band in the presence of tannic acid as compared to the control culture. Tannase isolated from the bacterial isolates can find some applications in industry.

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Poster

Biofuels **P074**

Cost Effective Biofuel: Optimizing Conditions for Cost Effective Microalgal Biomass Production and Harvest

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Poster

Human Genetics, Oncology **P075**

Genotypic and Computational Analysis of BRCA1 Gene Single Nucleotide Polymorphism with Benign Breast Tumors

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Benign tumors of breast tissue are believed and scientifically proven to be pre-stages of malignancy. Multiple confounding factors interacting in combinations can lead to benign breast tumor; however, genes play a highly significant role in it. Total of 191 Pakistani women were included in this study. This study presents to be an investigative research of BRCA1SNP i.e. rs35086932 and its association with benign breast tumor of female along with computational confirmation of its mutant isoform structure and visualization of its structural deviation by 3D-superimposition model. The SNP showed deviation from Hardy Weinberg Equilibrium in controls while significant association was found in benign breast tumor cases. Furthermore, increased risk of benign breast tumor was associated with major allele (common allele T). Genotypic frequencies showed significant p value <0.05 with patients, but not with controls. Similarly, results of allele frequency were found to be in accordance to genotypic results, the major allele T showed increased risk with benign breast tumor, with a p value of <0.0001. Bioinformatic analysis of normal and mutant isoforms of selected SNP of BRCA1 gene proposed that polymorphism in this gene occurs due to the premature termination of translation of BRCA1 protein. Substitution of a Glutamine residue with Amber stop codon results in a truncated protein product. Normal protein size is 1863 amino acid while the mutant isoform consisted on 1457 amino acids. 3D- Superimposition Model exhibited structural deviation represented through multi-colored twists due to presence of SNP at that position. These findings suggest that selected BRCA1 variant is involved in breast tumor risk, much proven by computational analysis of SNP. We anticipate that targeting specific genetic variations confined to ethnic groups would be more effective in future therapeutic approaches for prevention and treatment of benign breast tumors.

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Poster

Environmental Microbiology **P076**

Produced Water as a Source of Biosurfactant Producing Bacteria

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Oil and gas reservoirs often contain water produced during oil formation trapped in underground channels which is brought up to surface during drilling, production and treatment processes, thus named as produced water (PW). PW is the largest by-product associated with oil and gas exploration usually having an oily texture and diesel like smell. Chemical constituents of PW vary depending upon the hydrocarbon product extracted and chemical composition of underground channels in contact with PW. Various organic and inorganic chemicals are also found in PW. Certain indigenous hydrocarbon degrading microorganisms thrive in this environmentally stressed hydrocarbon contaminated PW. Most active hydrocarbon degraders are bacteria. In PW, the supply of nitrogen and phosphorous serves as a limiting factor while enhanced availability of carbon selects for bacteria that feed exclusively on hydrocarbon. Biosufactant (BS) producers are a group of hydrocarbon degraders which produce surface active compounds that serve to reduce surface and interfacial tension. Present study focuses on isolation of BS producing bacteria from produced water samples collected from Fim Kassar, Potwar, Pakistan. 13 strains (12 gram positive rods and 1 gram positive cocci) were isolated and screened for BS production by hemolysis test, for which 30 % isolates tested positive. These hemolytic strains were further screened using criteria such as emulsification index test, oil spreading test. Results of this study suggest that selected isolates can be used for bioremediation of hydrocarbon contaminated sites, petroleum up-grading, in crude oil drilling and in food processing industry.

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Poster

Microbial Genetics **P077**

Analysis of rpsL and PncA Gene Mutations in Multi Drug Resistant Strains of *Mycobacterium*

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Streptomycin and Pyrazinamide are important first line drugs for treatment of tuberculosis. rpsL gene of Mycobacterium tuberculosis encodes for 30S ribosomal subunit protein S12 involved in the translation initiation step. Streptomycin binds to the 30S subunit, and inhibits protein synthesis. PncA encodes for pyrazinamidase which hydrolyses pyrazinamide into pyrazinoic acid. Pyrazinoic acid is the active form of drug, which has bactericidal effects due to destabilization of membrane potential and alteration of transport function. Present study aims to find out the mutations in rpsL and PncA in the streptomycin and pyrazinamide resistant strains respectively. Cultures of local clinical isolates of streptomycin and pyrazinamide resistant MDR-tuberculosis strains were collected from Gulab Devi Hospital Lahore. Genomic DNA was extracted using CTAB-NaCl method. Full length rpsL and PncA genes were amplified from 13 MDR-tuberculosis cultures. Samples were submitted for sequencing at commercially available facility. Sequence analysis revealed 38 G>A mutation in rpsL and 195C>T, 195C>A, 389_390insG, 344C>G and 376G>A mutations in PncA. Mutations detected in rpsL and PncA could provide insights for quick diagnosis of drug resistance against streptomycin and pyrazinamide. This knowledge could also be helpful for development of better drugs against tuberculosis.

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Poster

Microbiology **P078**

Isolation and Characterization of Cellulase Producing Bacteria

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Four cellulose degrading bacterial isolates as *Bacillus subtilis*, *Corynebacterium xerosis*, *Bacillus insolitus* and *Bacillus marinus* were isolated by growing them on Carboxymethyl cellulose Na agar medium and showed hallow zones around the growth. These bacterial isolates were further grown in salt medium containing filter paper and showed growth .These bacterial isolates had a shorter lag phase when grown in L.B medium as compare to CMC-Na medium. In enzyme assay *Bacillus subtilis* has shown maximum O.D as compare to other strains. Phylogenetic analysis through neighbor joining method done to analyze phylogenetic relationship of isolated strains. Dark proteins bands with molecular weight of 44 and 55 KDa were observed in case of *Bacillus subtilis* through Polyacrylamide gel electrophoresis. Bacterial enzymes having a high potential to degrade such complex polysaccharides can be exploited for industrial biotechnologies.

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Poster

Immunity **P079**

Newcastle Disease Virus with Particular References to Diagnosis, Vaccination and Immunity Development

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Newcastle disease (ND), is caused by paramyxovirus serotype 1 (APMV-1) virus that. Newcastle disease represents one of the most important threats to the poultry industry worldwide. The clinical signs exist in birds infested with NDV can vary widely but principally depend on virulence of the virus. Other factors regulate the consequence of the disease such as the breed susceptibility, strain, age, immune status, infection with other organisms, nutrition and ecological stress. The clinical signs and occurrence of new stereotypes make it challenging to recognition and diagnosis. Laboratory testing is essential to confirm field suspicion, to distinguish the virus, and to succumb with international reporting requirements. Newcastle disease can be prevented by good biosecurity practices and vaccination. Newcastle disease virus may cause conjunctivitis in human but human-to-human spread has never been reported. A combination of live and killed ND vaccine provides better protection against virulent NDV. Locally isolated vaccine strains offer an attractive approach for immune intrusions by providing long-lasting immunity. However, continuous improvements in ND vaccination requires a superior immunological mechanism. A major function of the humoral immunity is to protect the birds against clinical signs caused by infection with virulent NDV strains, whereas increase in numbers of leukocytes at the place of vaccination could be responsible for uptake, processing of virus antigen and production of antibodies and antiviral cytokines. These local immune responses & body's first line of defense helps to protect the birds against ND infection of the mucosal lining of respiratory tract, which is the primary target site of virus.

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Poster

Environmental Microbiology **P080**

Analysis of Phytochemicals, Antibacterial and Antioxidant Activities in some Medicinal Plants

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Phytochemicals present in medicinal plants are a major source of imparting different characteristics to the plant. Antibacterial and antioxidant activity of these bioactive compounds was tested in four medicinal plants i.e. *Ziziphus jujube* (Unaab), *Fagonia arabica* (Dhamasa Booti), *Mallatus phillipensis* (Kameela) and *Hemidesmus indicus* (Ushba). Ethanol and hexane extracts of these selected medicinal plants were prepared and phytochemical analysis was done. Antibacterial activity of these extracts was performed by qualitative and quantitative methods i.e. agar well diffusion and MIC assay respectively. Plant that exhibited maximum antibacterial activity was tested for its bacterial efflux pump inhibition potential and its components were separated by TLC. Component showing antibacterial potential was subjected to GCMS analysis. Antioxidant activity of these extracts was also estimated using different methods. Phytochemical analysis revealed the presence of various components in selected plants. Maximum antibacterial activity against gram positive strain was shown by ethanol extracts while for gram negative strain no considerable inhibition was observed by either of the extracts. *Mallatus phillipensis* (ethyl acetate) extract showed maximum inhibition potential and GCMS analysis indicated phthalic acid to be the component responsible for this activity. Significant antioxidant activity was also observed from *Mallotus phillepensis* extract. Phathalic acid, responsible for antibacterial activity of plant extract, can be used in medicine industry to treat certain bacterial infections.

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Poster

Plant Microbe Interactions **P081**

Plant Growth Promoting Parameters of Isolated Bacterial Strains from Bovine Manure for Sustainable Vegetable Farming as Biofertilizer

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The diversity of plant growth promoting bacteria found in raw bovine manure exhibit great potential for colonizing rhizosphere and facilitating plant growth. Therefore, exploitation and application of plant growth promoting bacteria should be amplified for sustainable agriculture. The bacterial strains isolated from the raw bovine manure have different genetic and metabolic characteristics. Randomly selected samples were screened out for the isolation of beneficial bacterial strains. Selected bacterial strains were subjected to biochemical tests and identified as; *Bacillus*, *Staphylococcus*, *Streptococcus*, *Micrococcus*, *Corynebacterium* and *Pseudomonas*, predominantly. These bacteria were analyzed for plant growth promoting parameters by performing IAA production and nitrogen fixation tests along with phosphate and potassium solubilization test. In addition further tests such as calcium, zinc, magnesium, manganese solubilization and HCN production were also performed. Production of beneficial enzymes such as cellulase and pectinase were also checked on specific media for these tests. In conclusion, the bacterial strains which exhibit multiple beneficial characteristics could be used as biofertilizer for sustainable agriculture as well as to replace artificial chemical fertilizers increasing environmental pollution day by day to save environment.

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Poster

Microbiology **P082**

Metabolic Fingerprinting of Bacterial Strains for Pectinase and Cellulase Production

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Pectinases and cellulases are industrially important enzymes. These enzymes are produced by a variety of microorganisms. However there are few studies on the production of these enzymes by different species. This research work has been undertaken for the screening, isolation, purification and characterization of pectinase and cellulase producing strains of bacteria. That was carried out from different fruits, vegetables and soil samples. Twenty three bacterial strains were isolated and purified on L- agar medium by these samples and 26 strains were taken from the Lab 1 repository of Department of microbiology and molecular genetics. Biochemical tests including catalase test, oxidase test, starch hydrolysis test, glucose fermentation test, mannitol salt agar test, lactose fermentation test, MR-VP test, indole test and hydrogen sulfide production test were done for the characterization of selected bacterial strains and identified as *Bacillus, Lactobacillus, Citrobacter, Klebseila* and *Crorynebacteria* species. Antibiotic susceptibility was also tested for all the isolates. The screening of the pectinase producing strains was carried out by using well plate method on MS medium supplemented with pectin while for cellulases CMC minimal salt agar medium was used. For the quantification of enzyme activity Dinitrosalicylic acid method was used. Out of 49 strains, 28 were pectinase producer while 27 were cellulase producer. Most of the strains gave significant cellulase and pectinase activity but few of them did not show any zone of inhibition. The maximum pectinase activity was 2.74U/µl that was given by strain N while maximum cellulase activity was 1.39U/µl ($p > 0.05$) given by strain P41.

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Poster

Industrial Microbiology **P083**

Inhibitory Effects of Plant Extracts and Putative Endophytic Actinomycetes from the Selected Members of Meliaceae Family against MDR *Pseudomonas aeruginosa*

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The demands for the new antibiotic are increasing in the pharmaceutical market because of the increasing trends in the resistance pattern among the infectious microbes and increased prevalence of diseases in hospital. The increase in the resistance also leads to the marketing of chemically synthesized drugs that results in high cost and economic burdens. The aim of the present study was to investigate the endophytes from medicinal plants which are relatively untapped natural source that can compensate the need for new antibiotics for pathogens especially against MDRs. So in this study, a total of 20 endophytic actinomycetes strains from the plants of Meliaceae family, *Azadirachta indica* (Neem) and *Melia azidarach* (Dharek) were isolated following the standard surface sterilization protocols. The endophytic actinomycetes were isolated by surface sterilization of the plant tissues. The methanolic crude extracts of the endophytic actinomycetes and extracts of plant tissues were prepared. Antibiotyping of the *Pseudomonas aeruginosa* was done using a panel of 13 antibiotics by disc diffusion assay. The antimicrobial activity of the methanolic extracts was determined by well diffusion assay, while the cytotoxicity of the extracts was determined by brine shrimp microwell cytotoxicity assay. The chemical profiling of the extracts was performed by thin layer chromatography (TLC) and high performance liquid chromatography (HPLC/UV) analysis. Over all the study revealed that the isolated endophytic actinomycetes of Meliaceae family as well as their plant extract contains bioactive metabolites that inhibit the growth of MDR *Pseudomonas aeruginosa*.

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Poster

Pharmaceutical Microbiology **P084**

Evaluvation of Antibacterial and Antioxidant Properties of Ethanolic Extracts of *Althaea officinalis*, *Caesalpinia bonduc* and *Nymphaea alba*

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Medicinal plants are rich source of secondary metabolites that exhibit antimicrobial activity to combat various infections. Three different local medicinal plants *Althaea officinalis* (khatmi), *Caesalpinia bonduc* (karanjwa) and *Nymphaea alba* (gul-e-neelofer) were selected due to their cholesterol lowering ability. Common phytochemicals found in these plant extracts were alkaloids, flavonoids, glycosides and carbohydrates. Antibacterial activity was performed with different concentrations i.e. 250 mg/ml, 100 mg/ml and 75 mg/ml against *Bacillus* and *Pseudomonas* by agar well diffusion assay. Most significant zones of inhibition were revealed by *Nymphaea alba* against gram positive bacteria (30 mm), however, *Althaea officinalis* showed best zones against gram negative bacteria (21 mm). Maximum antioxidant activity (85%) was revealed by *N. alba* as compared to the other extracts. Minimum inhibitory concentration of *Nymphaea alba* extract was not determined before and showed that 1.562 mg/ml and 3.125 mg/ml concentration could inhibit the growth of gram positive and gram negative bacteria. The presence of different chemical components was confirmed by thin layer chromatographic technique. The results of study revealed that the medicinal plants possess different phytochemicals, and have very promising antibacterial activity against gram positive and gram negative bacteria up to our knowledge. So, the effectiveness of these naturally occurring medicinal plants should be considered for the improvement of new methodologies to treat bacterial infections instead of synthetic antibiotics.

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Poster

Industrial Microbiology **P085**

Characterization of PHA Producing Bacteria Isolated from Produced Water (PW)

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Produced water (PW), a byproduct along with oil and gas is produced in oil industry and is comprised of various organic and inorganic compounds. These compounds are used to accumulate polyhydroxyalkonates that have acquired significance in terms of their structural versitality, biocompatibility, biodregradablity and strong association with conventional plastics. These polymers are produced by gram positive as well as gram negative bacteria under stress conditions.Three samples of produced water (PW) were used to isolate PHA producing bacteria because it contains hydrocarbons in the same zone. The biochemical analysis of produced water (PW) samples revealed less than 0.4 mg/ml of glucose concentration using O-Toludine method and more than 0.7 mg/ml of protein concentration using Bradford test. Bacteria were identified by means of certain staining techniques and biochemical tests. Gram staining test revealed seven negative strains in 1st sample of PW, one negative strain and five positive strains in 2nd sample of Pw and two negative strains in addition to three postive strains in 3rd sample of PW. On the basis of Nile blue screening and Sudan staining, seven strains were observed for producing fluorescence and black granules out of eighteen strains. Glucose was used as a sole source of carbon and optimized for enhanced growth of PHA on minimal media. Strain V was selected for PHA extraction and it produced 11.34% of PHA after 72 hours of incubation. Positive strains were further characterized on molecular grounds and DNA was extracted. A fragment 0.7 kb of of PhaC gene was amplified.

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Poster

Human Genetics **P086**

TMPRSS6 rs855791 Polymorphism and Associated Factors in Female Patients of Iron Deficiency Anemia

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Iron Deficiency Anemia is the most common worldwide anemia with many clinical complications.The aim of this study was to assess the TMPRSS6 polymorphism rs855791 that influences the susceptibility of iron deficiency anaemia in Pakistani females. Initially, two camps were arranged for both males and females. From the subjects at these two camps, it became clear that IDA is more prevalent among females, so next two camps were arranged only for females. Complete blood count and biochemical parameters were performed in CitiLab and Research Centre, Lahore. Patients were further divided into Iron deficiency anemia, latent iron deficiency, negative iron balance and anemia of chronic diseases based on hematological parameters. DNA extraction, PCR of the gene TMPRSS6 and restriction fragment length polymorphism analysis, by stu1 restriction enzyme, of the samples was performed at the Department of Microbiology and Molecular Genetics. Certain behavioral and environmental factors have been found to be strongly associated with IDA and were assessed with the help of a questionnaire. It is evident that Iron deficiency anemia is more common among females than males. These results suggested that homozygous CC for TMPRSS6 rs855791 may play a protective role against Iron deficiency anemia. Dietary habits, menstruation and pregnancies in females contribute a great deal in development of Iron deficiency anemia among females. If these factors are controlled, we may be able to reduce the prevalence of IDA among females of Pakistan. Further studies in larger number of patients are necessary to identify potential polymorphisms. This may help for management of this problem.

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Poster

Plant-Microbe Interaction **P087**

Multifarious Beneficial Attributes of Phosphate Solubilizing Bacteria Affecting *Triticum aestivum* under Different Treatments

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In soil, phosphate is present in different organic and inorganic forms. The inorganic forms of phosphate are not available to plants. Plants can only take up the organic phosphate from soil. Insoluble phosphate which can be converted to soluble form by the activity of Phosphate Solubilizing Bacteria (PSB). The purpose of this study was to isolate and evaluate the potential of PSB on wheat production and soil fertility under different treatments. Initially twenty eight bacterial strains were isolated and based on good solubilization abilities, four selected strains were used for this study including *Acinetobacter baumanii*-JA10, *Pseudomonas plecoglossicida*-R14, *Pseudomonas putida*-SL8, and *Pseudomonas aeruginosa*-SpA. Evaluation for plant growth promoting activities was done by isolates. The strain SpA showed maximum ammonia production, HCN production and siderophore production. Strain SL8 shows maximum auxin estimation (66.385 µg ml^{-1}). Phosphate solubilizing bacteria reduce their activity in presence of pesticides but they still have the ability of phosphate solubilization. Phosphate solubilizing bacteria in the presence of pesticides (Chlorpyrifos and Pyriproxyfen) showed no decline in plant growth which indicated that bacteria have the ability to breakdown the phosphate group present in the pesticides and solubilize phosphate, Pseudomonas putida showed increase in number of root, length of root and length of shoot in wheat plants in in-vitro conditions. Furthermore significant increase in plant growth parameters was observed by inoculated strains in the presence of phosphate and pesticide treatments in greenhouse conditions. Phosphate solubilizing bacteria are potential bio-inoculants for wheat plant in natural and stressed conditions.

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Poster

Diabetes **P088**

Molecular Analysis of Pyridoxine Level between Female Diabetic Popultion of Lahore and Sheikhupura

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Diabetes is one of the most widely occurring human disease. Worldwide prevalence has risen over the past two decades. According to the world health organization (WHO), it is reported that diabetes is sixth leading cause of death. In the present research work, the aim is to compare pyridoxine level between diabetic patients of Punjab University Lahore premises and D.H.Q hospital Sheikhupura. 100 samples of diabetic patients and 50 samples of normal healthy controls are collected from PU Lahore premises and D.H.Q Sheikhupura. To assess the pyridoxine level, serum level of samples is estimated and analyzed initially using different standard referred assays, protocols and then high-performance liquid chromatographic (HPLC) assay of all the samples is performed. Data obtained by performing all assays is analyzed. The variation in the results of samples of diabetic female patients of both cities and healthy group is mapped and pyridoxine level appeared to be low in both female diabetic populations and extremely low in Sheikhupura female diabetic population. The result may suggest effective method for early diagnosis of risk for this disease. It is suggested that pyridoxine level can be the early indicator of diabetes onset. Moreover, pyridoxine supplementation may reduce the rate of disease incidence

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Environmental Microbiology **P089**

Isolation, Characterization and Optimization of Hexavalent Chromium Resistant Bacterial Strains from Tannery Effluent

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Hexavalent Chromium is a known mutagenic and carcinogenic agent. Several indigenous chromium resistant bacteria present in the tannery water which can reduce the toxic effects of chromium compounds. Sample was collected from a tanning industry near Kasur, Pakistan. Six different chromium resistant bacterial strains (Cr1, Cr2, Cr3, Cr4, Cr5, Cr6) were isolated on the LB- media at an initial concentration of 1000µg/ml (K_2CrO_4) by serial dilution method. Morphological characterization revealed that all bacterial colonies were raised, off-white in color and with entire margins except Cr6 which was yellow in color with undulate margins. Gram staining analysis showed that all strains were gram positive cocci. Reduction potential studies revealed that all these strains could remove up-to 80% Cr (VI) removal at an initial concentration of 1000µg/ml. Bacterial strains were grown at three different temperatures i.e. (28°C, 37°C and 45°C) and variable pH (5, 7 and 9) for the optimization of best growth conditions. Optimum temperature, pH for all six strains were 37°C and 7, respectively. Glucose and trypton were screened out as best carbon and nitrogen source for these strains. Findings of the present study revealed that these bacterial strains can be utilized as potential agents for the remediation of chromium contaminated environment.

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Poster

Human Genetics **P090**

Molecular Genetics of BRCA2 Gene Associated with Breast Cancer

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Conventionally, breast cancer was considered as a single disease but recent researches have described it as a complex disease that is characterized by high degree of medical heterogenecity. It involves a strong association between environmental and genetic factors due to which it is a multi-factorial disorder with a contribution of hereditary factor of 5-10 percent. It contributes 23 percent of cancer cases and 14 percent of cancer related deaths. The aim of present study is the identification of single nucleotide polymorphisms of BRCA2 gene that is associated with breast cancer in Pakistani females. For this purpose the blood samples are collected from Jinnah hospital and genetic analysis will be done. The control healthy and malignant samples are taken and their DNA are extracted by modified chloroform isoamylalcohol method and their SNPs genotyping will be done by using tetra-arm PCR. Genetic contrast modeling and chi-square analysis will be done to find the mode of inheritance of genetic variants. It will be concluded from the present study that BRCA2 gene polymorphisms can be used as a prognostic markers for breast cancer research.

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Human Genetics **P091**

Interaction of APOE Variant Arg176Cys and NOS3 Variant Glu298Asp with Cardiovascular Disease in the Population of Pakistan

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Cardiovasular disease is the major general reason of death worldwide. According to the 2010 Global Burden of Disease study it was reported that cardiovascular disease caused 15.6 million mortalities globally that is 29.6% of all mortalities. Diabetes, age, use of antihypertensive as well as lipid-lowering drug, body mass index (BMI) (weight/height^2) (Kg/m^2), smoking, as well as Canadian Cardiovascular Society Functional Classification of Angina (CCS Class) are included in the cardiovascular risk factors. Many genes are associated with CVD, many of them are identified, and many are still being studied. The purpose of the study is to determine the association pattern of APOE variant Arg176Cys and NOS3 variant Glu298Asp with cardiovascular disease in the population of Pakistan. Blood samples from both control and CVD patients are collected from different locations of Pakistan and DNA isolation are done through manual procedure. Then target gene amplification will be performed by tetra-arm PCR and statistical analysis will be done to find out the association of APOE and NOS3 genes with cardiovascular disease.

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Microbial Genetics, Industrial Microbiology **P092**

Analysis of Pks-I, Nrps, Cyp P450 Hydroxylase And Glycopeptide Oxy B, Antibiotics Biosynthetic Gene Clusters in *Streptomycetes* Strains

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Gene based screening for the detection of antibiotics biosynthetic gene clusters in biologically active microorganisms is a modern strategies to search for new drugs. In this study the antimicrobials production potential of the *Streptomyces* strains was determined both by molecular and culture screening approach. The four biosynthetic genes clusters including; PKS-1, NRPS, polyene specific CYPs and glycopeptide Oxy B gene were investigated in the selected strains by PCR amplification, sequencing and by subsequent bioinformatics based approach. The screening results showed that among the 40 selected *Streptomyces* strains, 33 strains possessed the gene for NRPS; 17 strains for PKS-1; 4 strains for CYPs and none of them contain glycopeptide Oxy B gene. The bioinformatics based tools were applied to confirm the presence of CYPs, glycopeptide Oxy B, NRPS and PKS-1 proteins that plays an important role in the antibiotics biosynthesis pathways. The *Streptomyces* strains which exhibited the presence of polyene specific CYP gene in their genome were also screened for the production of antifungal metabolites. The *Streptomyces* strains including NR-1, NR-10, NR-14 and NR-15 were investigated by biological activity (antifungal activity) by agar plug and well diffusion assay and chemical profiling (TLC, HPLC) methods. In biological screening the antifungal activity was determined against *Fusarium oxysporum*, *Rhizoctonia solani* and *Aspergillus* isolate FN2. The selected strains exhibited pronounced antifungal activity against the fungal test strains. In chemical screening the methanolic extracts of these strains exhibited characteristic polyene like TLC and HPLC profile. Overall the study revealed that the selected *Streptomyces* strains harbour the antibiotics biosynthetic gene clusters for four major antibiotics structural classes, these strains may yield commercially useful antimicrobial agents if investigated for the purification and structure elucidation of the compounds.

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Poster

Biochemistry **P093**

TFII-I Isoforms act as a Co-Regulator Potentiating Nurr1-Induced Human TH Expression During Dopaminergic Neurogenesis

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Nurr1 plays a vital role in development and maintenance of midbrain dopaminergic neurons. Our previous study showed that Nurr1 actively represses human tyrosine hydroxylase (hTH) transcription in hNSCs, while it activates hTH expression via NBRE-A in DA cells. To identify the interacting protein partners of Nurr1 to regulate hTH expression during DA neurogenesis, we performed DNA pulldown assay and identified TFII-I, a multifunctional transcription factor having four isoforms.Expression of TFII-I isoforms was analyzed by PCR and western blot. Immunoprecipitation was performed for protein-protein interactions and ChIP for interaction of proteins to promoter. Mutants were constructed by PCR based strategy. Luciferase assay was performed to analyze promoter activity.TFII-I expression switch from TFII-IΔ to TFII-Iγ³ isoform was observed in midbrain of embryonic mice from E9.5-E13.5. TFII-IΔ preferentially interacts with SUMOylated Nurr1 and occupies hTH promoter in hNSCs resulted in repression of hTH activity while TFII-Iγ interacts with Nurr1 on hTH and enhanced hTH promoter activity in DA cells.TFII-I binding sites, an E-Box and an Inr element are found up- and down-stream of the NBRE-A respectively. In addition, ELM analysis and IP showed that only TFII-IΔmodified by SUMO1 at K221 and K240 in putative motifs. SUMO modified TFII-IΔ in a transcriptional complex with SUMOylated Nurr1 on the NBRE-A element, makes a complex in functional SCM and represses hTH promoter activity in hNSCs. Furthermore, SUMO deficient forms of TFII-IΔ showed enhanced binding and resulted in hTH activation in hNSCs only. Mutation of E-box and Inr showed that TFII-IΔ represses hTH activity via Inr. Our data specified expression of TFII-I isoforms in midbrain of mouse and established opposing role of TFII-IΔand γaccountable for Nurr1 mediated repression and activation of hTH respectively during dopaminergic neurogenesis.

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Antimicrobial Agent & Chemotherapy **P094**

Antimicrobial and Chlorhexidine Mouthwash Resistance of Dental Plaque Bacteria

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Chlorhexidine is used as a disinfectant and in oral care mouthwashes. In this study dental plaque bacteria were plated on media containing 2 µg/ml chlorhexidine gluconate. Resistant bacteria were characterized by 16S rDNA sequencing and antibiotic resistance profiles were determined using the disc diffusion method. The isolates were found to be variably resistant to multiple drugs including ampicillin, kanamycin, gentamycin and tetracycline. Biofilm formation and planktonic growth in the presence of chlorhexidine gluconate or a chlorhexidine-containing mouthwash showed a dose response. Two species, *Chryseobacterium* culicis and *Chryseobacterium* indologenes were able to grow in broth culture and form biofilm in the presence of 32 µg/ml chlorhexidine supplied as pure chlorhexidine gluconate or from a commercial mouthwash (Savacol). However they did not grow when media was supplemented with the antiseptic solution Dentalife as chlorhexidine source. Exposure of biofilms to the undiluted mouthwash for intervals from 5-60 s showed dose response growth inhibitions with maximum inhibition after 30 s exposure. In practice therefore, use of antiseptic solutions such as Dentalife in the dental clinic and use of chlorhexidine mouthrinses for at least the minimum 30 s recommended by the manufacturer can help combat bacteria resistant to a range of antimicrobials.

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Poster

Environmental Microbiology **P095**

Evaluation of Azad Kashmir's Soil Bacterial Flora for Biopolymer Production

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Soil samples for this study were collected from Tatta Pani, Azad Kashmir. The isolated strains were screened for the production of PHA and EPS. Nile Red and Sudan Black staining techniques were used to screen PHA producers. EPS production was checked by using ice chilled ethanol. Strain AJ2 and AJ3 were selected due to their ability to produce high amounts of both PHA and EPS. Time profiling and optimization of PHA and EPS production was done for these selected strains. Glucose, glycerol and molasses were used as carbon sources for PHA production. PHA production was checked at different conditions including high pH, high temperature and at different time intervals. Highest yield of PHA was given by strain AJ3 i.e 89.43% with molasses under normal incubation conditions. When grown at 55 °C for 24 hours, Strain AJ3 showed highest PHA accumulation. At alkaline pH, strain AJ3 gave 34% with molasses. Strain AJ3 (7 g/100ml) produced Maximum EPS with glucose. The phylogenetic analysis of 16S rRNA gene showed that strain AJ2 has 100% resemblance with *Micrococcus yunnanensis* (MF496374). While strain AJ3 resembled with *Psychrobacter sp*. (MF496375). The isolated strains significantly produced EPS and PHA.

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Aquatic Microbiology **P096**

Characterization of Extracellular Enzyme Producing Bacteria Isolated from Arabian Sea and Ravi River

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Bacteria are famous to produce many bioactive molecules and extracellular enzymes of commercial importance. Marine and freshwater bacteria occupy prominent position in producing commercially important enzyemes. To investigate bacterial extracellular enzyme prodcing activity samples were collected from Arabian Sea, Karachi and Ravi River, Lahore-Pakistan. Samples were treated to isolate pure bacterial cultures by serial dilution method. Twenty three different bacterial strains were selected and identified by their morphological, physiological and biochemical characteristics. Gram staining, spore staining, acid-fast staining, catalase test, mannitol fermentation , glucose fermentation, pigment test, voges proskauer test, methyl red test, DNAse, novobiocin sensitivity tests were performed to identify the isolated bacterial strains. Phylogenetic analysis was done by 16S rRNA sequencing. These isolated strains were further screened for the production of various extracellular enzymes (Pectinase, Amylase, Tannase, Protease, L-Glutaminase, Gelatinase, Cellulose, DNAse, Lecithinase). It was found that out of these selected 23 bacterial isolates, 13 strains showed pectinase activity, 9 had amylolytic activity, 17 had l-glutaminase activity, 20 had gelatinase activity, 5 had DNAse activity and 8 strains had lecithinase activity. While tannase, protease and cellulase activity was not exhibited by any of the 23 isolated strains. The 6 strains which showed maximum number of extracellular enzyme production were selected for furthur testing for their maximum tolerable concentration for different antibiotics and heavy metals.

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Poster

Nanobiotechnology **P097**

Comparative Cytotoxicity Study of Silver Nanoparticles Synthesized by Utilizing Bacterial Cells and Plant Extracts

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Metal nanoparticles are chemically and physically different in properties than the bulk material (e.g. mechanical qualities, higher surface areas, lower melting points, specific optical densities and specific polarizations) properties that may demonstrate appealing in different modern applications. Nanoparticles can be synthesized chemically, physically or biologically. Nanoparticles synthesis by synthetic (chemically) methodologies are costly as well as environment unfriendly. Consequently, there is an urge to build up economical and eco-friendly biological procedures (microbial and plant synthesis) that do not utilize poisonous or toxic substances for nanoparticles synthesis. Silver metal has been utilized widely in various applications due to its antimicrobial effect. The research work involves two major portions, first: the biological synthesis of safe and eco-friendly silver nanoparticles at lab scale, second: the demonstration of positive and negative effects of these silver nanoparticles on explant of *Solanum tuberosum*. For microbial synthesis, *Exiguobacterium aestuarii* and *Achromobacter xylosoxidans* were used. For plant mediated synthesis extracts of plants and spices were used. The invitro studies of *Solanum tuberosum* explant showed that AgNPs synthesized by bacteria have positive effects on plant development as compared to AgNPs synthesized by plants.

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Poster

Bioinformatics **P098**

In Silico Screening of Phytochemicals as Agonists of ADRB2 to Treat Asthma

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Asthma is a worldwide disease and is also common in Pakistan. The most commonly used drugs are beta-2 agonists which bind to beta-2 adrenergic receptor (ADRB2) and corticosteroids as well. In humans, ADRB2 is a member of the G protein-coupled receptor (GPCR) superfamily. GPCR is very important now-a-days as 40% modern drugs target this receptor. Overuse of corticosteroids and beta 2-agonists has many side effects and human become resistant to these drugs. So, people are moving again towards herbal medication. In this study, different phytochemicals were screened. In silico, many ligands, including commercially available drugs and phytochemicals were docked with ADRB2 by using PyMol. And then druglikeness properties of selective phytochemicals were estimated by SwissADME. Total 194 ligands were docked with beta-2 adrenergic receptor (ADRB2). Out of these, 6 were commercially available drugs and 188 were phytochemicals. Percentage efficiency of phytochemicals was calculated as compared to drugs. Only 18 compounds having high affinity with ADRB2 receptor were selected. And ADME properties of 17 compounds were evaluated by SwissADME. These were studied by different aspects including physiochemical properties, lipophilicity, pharmacokinetics, drug likeness and medicinal chemistry. 18 phytochemicals with affinity ≤- 9 kcal/mol were derived from 12 unique plants. By correlation of all the parameters of the results of SwissADME with binding energy of phytochemicals with ADRB2 concluded that fraction coefficient, rotatable bonds and log Po/w, log kp are directly related with binding energy. Whereas the number of hydrogen bonds acceptor is inversely related to binding energy. It was concluded that in plants oat, barberry, sorrel, lemon, holy basil, opium poppy, waterland weed, baikal skullcap, thyme and cacao have potential to become an effective medicine. Whole plant can be used in herbal medicines with no side effects.

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Poster

Environmental Microbiology **P099**

Air Quality in Operation Theaters Of Urbanized Hospitals of Lahore, Pakistan

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The current study explores the air quality in orthopaedic operation theaters of six urbanized hospitals of Lahore, Pakistan. The objective of study was to assess the air quality in terms airborne microbial concentration and particle dust in operation theatres with different ventilation systems.Air samples (duplicates) were collected during surgeries by active sampling (Filtration method) on mixed cellulose ester filter (MCE; 0.45µm pore size, 47 mm diameter) using high volume air sampler. Considering ACGIH guidelines, 36 L/min and 20 minutes were chosen as a sampling strategy after performing some optimization test for time and bacterial cfu/m3 recovery. The bacterial plates were incubated at 37°C and observed after 48h; bacterial aerosols were observed and identified using standard microbial procedures. Particle concentration was simultaneously monitored to bacterial sampling for 20 min using DRX Aerosol Monitor (TSI Model 8533) in the size range from 1 to 15µm. Highest microbial counts were recovered from a naturally ventilated operation theater (7.3×107 CFU/m^3) of a government hospital; while the lowest count was found in a mechanically ventilated operation theater of a private hospital (1.0×10^3 CFU/m^3). *Staphylococcus aureus*, *Bacillus*, *Micrococcus*, *Enterobacter* and other coagulase negative *Staphylococci* were recovered from different operation theatres. Particle mass concentration did not correlate with the bacterial counts ($P>0.05$) in this study.This might be because of limitation of active sampling and cultural count method; culture method represents only a fraction of bioaerosol load while a direct reading particle monitor can account all the particles in a set size range. Moreover, complexity, duration of surgery, cleaning activities and proximity of operation theater to road were found to affect microbial concentration and particle dust levels.

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Poster

Nanobiotechnology **P100**

Green Synthesis of Silver Nanoparticles and their Antimicrobial Activity

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In this work silver nanoparticles were synthesized by indigenous bacteria usin g $AgNO_3$ (silver nitrate) as substrate. This paper aims to provide the biological approach for the synthesis of silver nanoparticles. This might be beneficial to control the nosocomial infections triggered by MRSA. The current study is the extracellular synthesis of silver nanoparticles by using the cell free filtrate of bacterial strains isolated from the soil. The optimization study was also carried out to obtain the maximum production of silver nanoparticles. These nanoparticles were confirmed and characterized by UV-Vis spectroscopy and Transmission Electron Microscopy (TEM). Out of 167 strains 3 appear to be best for the synthesis of silver nanoparticles and showed very good antimicrobial activity against MRSA. This is the green approach for the production of AgNPs, as there was no previous work done on the synthesis of silver nanoparticles by bacteria in this region of Southern Punjab, Pakistan and these nanoparticles can be used to treat nosocomial infection.

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Poster

Public Health **P101**

Awareness of Sickle Cell Anemia

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The objective of conducting the survey is appraising the awareness level about the sickle cell anemia among the general population of Punjab, Pakistan. A structured questionnaire having separate segments revealing sickle cell anemia's causes, symptoms and treatments etc. is accordingly filled by the people with their consent. All the data obtained was questioned and analyzed by Chi-square test and percentage analysis centered on the basis of age and education. On the basis of these aspects people are more aware about the sickle cell anemia

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Poster

Human Genetics **P102**

Analysis of Single Nucleotide Polymorphisms of DNA Repair gene-BRCA1 in Malignant Breast Tumors

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Breast cancer is the most common cancer among the females, worldwide. It can be inherited and sporadic. Many environmental factors are involved in the development of the disease. BRCA1 is the breast cancer susceptibility gene, located on chromosome 17, normally involved in DNA-damage repair pathways. Malignancy of breast tissues can occur due to the variations in BRCA1 gene. A case-control study was done by using malignant breast cancer samples to study the variants of BRCA1 gene. Six tag SNP's were studied. All the SNP's showed the deviation from Hardy Weinberg Equilibrium. Furthermore, an increased risk of malignant breast tumors is associated with the major allele. Seven haplotypes were significantly associated with the malignant tumors and TGCAGG haplotype appeared to increase 18 times more risk as compared to others with p-value <0.0001. Hence, it is concluded that variants of BRCA1 are strongly associated with the predisposition of the malignant breast tumors.

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Poster

Pharmaceutical Microbiology **P103**

Antibacterial Activity and Phytochemical Analysis of Extracts of *Caesalpinia bonduc*, *Tinospora cordifolia* and *Quercus infectoria*

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Infectious diseases are one of the real issues in developing as well as developed countries. Conventional medicinal plants are generally used to treat microbial disease because of their rich antimicrobial properties and less cost. Several kinds of medicinal plants are used for the treatment of infectious diseases. In this study, 3 medicinal plants *Caesalpinia bonduc* (karanjuwa), *Tinospora cordifolia* (Giloy) and *Quercus infectoria* (Manjakani or Majuphal) were used to analyze their antibacterial potential and phytochemical compounds. These plants are commonly used for the treatment of the epidemic diseases such as dengue and malaria. The different parts of the plants such as seed cover, bark, stem was extracted using different solvents like ethanol, methanol and ethyl acetate. Antibacterial activity of these extracts was tested against *Bacillus* (KC881030) and *Pseudomonas* (KC1031) by agar well diffusion method. Future study will be focused on the characterization of these antibacterial compounds. Phytochemical analysis of *Caesalpinia bonduc*, *Tinospora cordifolia* and *Quercus infectoria* have revealed the presence of flavonoids, alkaloids, tannins, phenolic compounds and Phyto sterols. Hence, present findings support the presence of antibacterial properties in these plants and this will help in the development of effective antimicrobial drugs.

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Industrial Microbiology **P104**

Isolation and Characterization of Biopalstic Producing Bacteria from Various Soil Samples

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Bioplastics, literally mean as "life plastic", are mainly derived or synthesized by certain microbiota. Unlike traditional petroleum based plastics, bioplastics are not involving petrochemicals, carbon origin is purely biotic and they are biodegradable. Taking this as motivation, bacteria were sampled from oil contaminated soil (Oil storage diggings at railway workshop, Pakistan), highly salty ground soil (Soil near khewra salt mines, Pakistan) and general agricultural soil (without fertilizers and pesticides). The purpose of this research was to organize and study the collection of such bacteria that have the ability to produce or biodegrade bio-plastics. Further checking out their genetics, origin, habitat and characteristics, that allow us to manipulate them and utilize them for the beneficial purposes related to production, degradation and environmental cleanliness. Using Nutrient agar with the stress of oil and salt, bacterial colonies were obtained from the samples. Three different colonies from each sample were streaked to purify and their morphology was studied. Gram staining of each purified colony was done. Out of 9 colonies, 6 were rod like (bacillus), found in clusters orientation. They were gram positive and spore forming bacteria while the rest 3 were spherical in shape (cocci). Two of these 3 were gram negative and 1 was gram positive. All these 3 were in clusters orientation. Further sequencing and purpose related studies of these isolates are in progress yet.

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Human Genetics **P105**

Effect of Olive Oil in Cholesterol Level of Diabetic Patient

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Diabetes mellitus is a metabolic disorder in all over the world. Diabetes can leads to long term life threating complication. High frequency of diabetes in Pakistan needed the discovery of new therapeutic methods to control the disease complication. In the present research work, the level of Cobalamin in diabetic patients have been assessed and identified by the recent advance technologies. And also check the valuable effect of olive oil in the cholesterol level, olive oil can help in diabetes in a number of ways as long term inflammation is through play role in diabetes and complication of disease. Serum of 50 sample with same age and sex that are normal healthy control and that are diabetic was collected from Diabetic Clinic Health Center University of the Punjab Lahore. Biochemical parameters and the total serum Cobalamin level were estimated by high liquid chromatography. Extra virgin Olive oil will be used, and will be given to the diabetic patients as 2 table spoon per day after meal for the period of three month. Samples of 50 diabetic patients group and control groups were then examined by HPLC and evaluated the levels of Cobalamin. The result were significant and level of Cobalamin and were reported down in these patients as compare to normal healthy control. Cholesterol level from the lipid samples of Diabetes patients shall be considered by techniques. Assessments of these levels will be helpful in not only early diagnosis but also in prediction of diabetes mellitus.

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Microbiology **P106**

Characterization of Some Biosurfactant Producing Bacteria from Indigenous Environment

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Biosurfactant producing bacterial strains isolated from variety of oil contaminated soils and carried out screening tests to check biosurfactantproducing capacity. Screening tests include Emulsification Capacity Assay and Drop Collapse Assay. The bacterial strains which showed positive results were selected. Selected bacterial strains were further carried out for their morphological, biochemical and physiological characterization. Mostly the strains were gram positive rods, while others were gram negative. Only 9-M, 5-T, 1-F and 8-F were motile and all other strains were non-motile. Biochemically all the strains catalase positive, 2-M, 4-S, 5-S, 10-S, 3-F and 8-F were oxidase negative. Various results were observed for bacterial resistance against ampicillin, erythromycin, tetracycline and chloramphenicol. All the strains were tetracycline sensitive except 4-S. Bacterial strains were sensitive towards mercuric sulphate, cobalt chloride and silver nitrate at all concentration. Four bacterial strains were able to resist mercury and cobalt while only one strain out of sixteen resisted the stress of silver. All the bacterial strains showed resistance towards metals at their different concentrations. Biosurfactant producing bacterial strains showed oil degradation potential at various concentration of oil (1%, 1.5 % and 2%) which was determined by decolonization of redox indicator 2, 6 dichlorophenol indophenol. Three strains out of sixteen showed high oil degradation potential 1-M, 4-T, and 2-F; hence they were selected as effective oil degrading bacterial strains.

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Antimicrobial Agents & Chemotherapy **P107**

Screening of Indigenous *Streptomycetes* for Identification of Antimicrobial Compounds Against Extreme Drug Resistant (XDR) *Acinetobacter* Involved in Nosocomical Infections

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To determine the current status of antibiotics resistance in Acinetobacter in Lahore, Pakistan and to screen indigenous streptomycetes isolated from various regions in Pakistan for activity against XDR Acinetobacter strains. Acinetobacter strains, isolated from different patient samples, collected from the Chugtai labs Lahore, Pakistan. These isolates were identified by using microbiological, biochemical and molecular genetics (16S rRNA gene sequencing) techniques. Antibiotics sensitivity pattern of Acinetobacter strains were determined by Kirby Bauer disk diffusion assay against a panel of 22 antibiotics specified by CLSI guidelines 2013 against *Acinetobacter*. Methanolic crude extracts of 18 Streptomyces strains were obtained by solvent extraction method and screened for antimicrobial activity by well diffusion and disk diffusion assays against XDR Acinetobacter strains. For partial identification of bioactive secondary compounds, chemical screening of active extracts was done by TLC and HPLC analysis. A total of 100 *Acinetobacter* strains were recovered from different patient samples. 100% of tested isolates exhibited resistance to Amoxicillin/Clavulanic acid, Ampicillin/Sulbactam, Cefepime, Cefoperazone, Ceftazidime, Cefotaxime, Imipenem, Meropenem, Ciprofloxacin, Levofloxacin, Moxifloxacin and Piperacillin/Tazobactam. While 96.8% were resistant to Trime/Sulphamethoxazole, 92.9% to Amikacin, 87.1% to Tobramycin, 64.5% to Cefoperazone/Sulbactam and 63.2% to tetracycline. However, resistance to Tygacil was only 6.5%. A very promising antimicrobial response of crude extracts of streptomycetes was observed against XDR Acinetobacter strains. The maximum zone of inhibition was given by the extract of Streptomyces strain 15 that was up to 20 mm in disc diffusion assay against the XDR Acinetobacter strain 955-29. Different types of bioactive metabolites were also documented by chemical screening of extracts of *streptomycetes*.

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Nanobiotechnology **P108**

Antibacterial Properties of Composite Resins Incorporating Organic Silica and Zirconia Nanoparticles Against *Staphylococcus* and *Bacillus* Bacteria

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At micro scale, properties of material are same as bulk but at nano scale they show different physical properties. Nanoparticle based dental resin composites increases wear and fatigue resistance in comparison with micro-filled composites and favors the accomplishment of restorative with improved and long term depiction. In this study nanoparticle based dental fillers were prepared by using different ratios of zirconia and silica nanoparticles with commercially used dental filler and their micro hardness, biocompatibility and other mechanical properties were evaluated. Zirconia and silica possess many advantages like high mechanical properties, high flexural strength and ability to bond to the enamel surface as compared to commercially used dental ceramic fillers. In comparison specimens having cavity filled with 100% self-cure were regarded as controlled group. Lowest and highest hardness number were found for self-cure (100%) and silica (50%) with zirconia (50%) respectively. Highest hardness recorded was ~1614 HV with durability of 9.77 years. Constituents of these nano fillers inhibit the second carries formation by controlling the biofilm growth by enhancing the antibacterial activity of commercially used dental fillers. Well plate method was used to calculate antibacterial activity. Zone of inhibition for these nanoparticles was found to be 28 mm against bacteria.

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Nanobiotechnology **P109**

Antimicrobial Study of Zirconia Coated Teeth

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Nanomaterials possess valuable biomedical applications. Zirconia has exceptional mechanical strength, hardness and fracture toughness. The use of zirconia in dentistry and medicine has promptly extended over past few decades. The present study aims is how to protect the human teeth. For this purpose, freshly extracted human teeth were collected from dental hospital and kept in saline solution for two weeks for preservation. Freshly extracted human teeth were firstly coated with five different commercially available toothpastes i.e Close up, Kodomo, Forhan's, Colgate, Pigeon for just maximum one minute and then dipped in different drinks or beverages (coke, lemon, green tea, black tea, milk coffee, milk, egg yolk) for time interval 15 min, 30 min, 45 min & 60 min. Zirconia has potential applications for protective teeth coatings. As-synthesized Zirconia NPs have been added in five different commercially available toothpastes i.e. Close up, Kodomo, Forhan's, Colgate, Pigeon. It has been observed that Zirconia NPs added toothpastes showed the effective results as it reduced the weight loss difference and increased the surface hardness of the teeth enamel. Hardness was determined by Micro Vickers hardness indenter and weight of the teeth was measured by analytical balance. When ZrO_2 NPs are added to Colgate then it showed high value of hardness ~1337 HV which is compatible for teeth coating. Well diffusion method was used to investigate antibacterial activity. As-synthesized Zirconia NPs has showed the strong inhibition zone of 29 mm against pathogenic bacteria. Among all the toothpastes, Colgate toothpaste showed maximum inhibition zone of 20 mm.

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Plant-Microbe Interaction **P110**

Analysis of Potency of Plant Growth Promoting Rhizobacteria on *Zea mays* Growth

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Rhizosphericbacteria, or rhizobacteria, are often generally designated as those having symbiotic relationships with plants but not contributing to the soil profile. Rhizobacteria which enhances the plant growth, are known as plant growth promoting rhizobacteria (PGPR) and this ability might be due to specific features of ammonia production, sidrophore production, phosphate solubilization and other traits of these bacteria. In this study, total 30 rhizospheric bacteria were isolated from several plants but twenty five bacterial strains were further tested for plant growth promotion traits. For the identification of growth promoting traits of isolated bacterial strains, different tests including HCN production, ammonification and auxin production tests were done. In order to test the beneficial effects of these bacteria on plants, plant-microbial interaction assay was done. For this purpose, *Zea mays* plant was chosen. Effect of bacteria on plant growth was observed. All of the bacterial strains showed specific effects on plant growth. Results revealed that these PGPR caused more enhanced growth as compared to control plants. Three strains were strong ammonia producers and also three strains were strong HCN producers but only two strains were auxin producers. In conclusion, these PGPR may further be used in agriculture and horticulture research where the plant growth is required to be enhanced because the chemical fertilizers which are used for enhanced crop production may be dangerous in terms of nutritional value and may also be dangerous for biological pests which act as natural pesticides.

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Nanobiotechnology **P111**

Antimicrobial Study of Titania Nanoparticles used as Teeth Coatings

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Titania (TiO_2) is well known for its excellent mechanical strength, being corrosion resilient and has numerous biomedical applications due to its bio-compatibility. The aim of study was how to protect the human teeth from demineralization. Five dentifrices were taken in which one medicated, two baby dentifrices, and two were used by general community and mixed TiO_2 Nano particles in them and investigated the effect of different foods (selected on the basis of pH) whole pH spectrum was covered, Eatables used coke, lemon, green tea, black tea, coffee, milk, egg yolk for experiment. Micro Vickers Hardness test and Optical microscopy was done to investigate mechanical properties and surface morphology and antibacterial effect also investigated. TiO_2 nano particles exhibited excellent results and enhanced mechanical strength. In milk, sample hardness increased to 1056HV for Forhan's toothpaste, which showed TiO_2 is compatible for teeth coating. Well diffusion method was used to investigate antibacterial activity.TiO_2 nanoparticles exhibited maximum zone of inhibition 14 mm and 17 mm at 8mg/ml and 10mg/ml respectively. Close up and Colgate when treated with TiO_2 nanoparticles exhibited maximum zone 30 mm, 27 mm respectively at 10 mg/ml. Forhan's and Pigeon exhibited maximum zone of 19 mm and 20 mm at 8 mg/ml respectively.

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Metagenomics **P112**

Isolation and Characterization of Salt Resistant Bacteria and Metagenomic Analysis of Saline Soil

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Salinity is considered as one of the major factors for limiting the growth of life as it affects the cellular processes as well as the plant yield, thus results in the decreased biological diversity. The present was performed to analyze the saline soil by culture dependent as well as culture independent techniques. Hypersaline area, located near Faisalabad was selected for the isolation of salt resistant species. Among all the isolated strains, *Pseudomonas putida* J8 was observed to show highest resistance against NaCl i.e. 5M. The species were also checked for cross metal resistance. *Bacillus subtilis* J1 and *Pseudomonas* sp. J5 showed the maximum resistance against all the tested metals (Se, As, Ni, Co, Cu and Zn). while checking UV resistance, *Pseudomonas putida* J8 showed maximum resistance against 20 minutes UV exposure. The strains were also checked for desiccation tolerance. The results showed that the highest viability was showed by *Bacillus subtilis* J1 i.e. 20%. . The bacteria with the multiple resistances can be used as the potential candidates for the reclamation of salt affected area. Metagenomic study includes the analysis of hypersaline soil samples in comparison with normal soil samples. Results of Heatmap and phylogenetic tree showed that the microbial community of saline samples (FSA and FSB) was originated from same ancestor, while that of the normal soil samples (NSA and NSB) was sharing the same origin. The samples were studied at different classification units for checking the relative abundance of different groups in different samples. Both of the saline samples showed the highest relative abundance for the family Haloacteriaceae, while the family was totally absent for normal soil samples. For normal soil samples, Acidobacteria Group-6 was the highest abundant family, but showed very low relative abundance in saline samples. The technique can be used as an effective tool for exploring and studying the uncultivable life.

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Diabetes and Molecular Biology **P113**

Olea Europea as a Lipid Lowering Therapy in Diabetic Patients of Lahore Region

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Studies reported that Extra virgin Olive Oil (EVOO) improves the glycemic index and lipid profile in diabetic patients but never been studied in the diabetic population of Pakistan. So the aim of this study was to check the health benefits of EVOO in the diabetic population of Pakistan. 100 Diabetic patients were given 10ml of EVVO on daily basis and blood samples were taken before the start of the therapy to check the level of Glucose, lipid profile, total cholesterol, HDL-Cholesterol Triglycerides, LFT's, HbA1c and Serum Creatinine. After 3 months of therapy blood samples were taken to check the effect of EVOO on these parameters. The results are promising as EVOO containing meal lowers the level of triglycerides and overall lipid profile as compared to control group without EVOO. this is the first study of EVOO on the diabetic patients of Pakistani population which shows the improvement in the glucose level and lipid profile.

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Antimicrobials **P114**

Screening for In Vitro Antitumor and Antimicrobial Activity of Rare *Micromonospora* Genera Isolated from Humus Rich Soil Samples

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The purpose of our study was to isolate the rarely encountered genera of actinomycetes i.e. *Micromonospora* for the screening of their antimicrobial and antitumor activity. A total of five humus rich soil samples were collected and after the pretreatment of the soil samples, almost 12 strains presumed to be the members of the genera *Micromonospora* were selected. The isolates were characterized morphologically, biochemically through following the proper guidelines of International Streptomyces project (ISP) mentioned in the Bergey's manual. The isolates were cultivated in glucose yeast-malt extract media and extraction of the metabolites produced by them was done through solvent extraction by using ethyl acetate. The final extracts were dried and dissolved in methanol. These methanolic crude extracts were checked for their antimicrobial activity by using well diffusion method and antitumor activity was determined by MTT assay. The antimicrobial activity was checked against seven different test strains and antitumor activity was determined against HCT116 colorectal carcinoma cell line. The methanolic crude extracts were further evaluated through chemical screening which included; Thin layer chromatography (TLC) and High performance liquid chromatography (HPLC), which showed the production of diverse range of compounds by these strains. The bioactive strains will be phylogenetically identified through 16S rRNA sequencing. The isolated strains from humus rich soil belonging to the genera Micromonospora were found to be potential producers of antimicrobial and antitumor compounds. In future, the bioactive metabolites produced by the isolates in our study could be used as a source of pharmaceutical agents after their purification and identification through spectroscopic techniques (LC/MS and NMR).

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Industrial Microbiology **P115**

Fatty Acid Utilizing Bacteria Produce Sustainable Bio-Polymers

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Bio-based materials are gaining a lot of attention these days and there are ever increasing efforts to make their production sustainable. Co-production of exo and endo-polymers seems to be a good idea as one compound is produced inside of cells while other is excreted outside. For this purpose, an experimental effort was done that focuses on the isolation of bacterial strains with the ability to degrade complex organic molecules and produce exopolysaccharides and poly-3 (hydroxy-butyrate). The water and soil samples for bacterial isolation were collected from a tire recycling factory. The production of PHB and EPS was optimized using glucose, cholesterol, and palmitic acid as carbon sources. The selected organisms were classified up to specie level by 16s ribosomal RNA identification and were found to be *Rhodococcus pyridinivorans*-NK19 (KY703220) and *Acinetobacter junni*- NK10 (KY703219). Bacterial strain *Rhodococcus pyridinivorans* NK19 seemed to produce more PHB as compared to *Acinetobacter junni* NK10, 54 and 40.5% respectively when supplemented with glucose. Among other selected carbon sources palmitic acid and cholesterol was supplied to bacterial isolates and considerable amounts of PHB were observed 43 and 37% respectively. The maximum EPS production was 43.5 and 197 g/l after 72 hours of incubation by NK19 and NK10 respectively. The FTIR analysis of extracted PHB was showed good resemblance with pure PHB and peaks for ester linkage and ester carbonyl group (1711 cm/1).

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Medical Microbiology **P116**

Chlamydia trachomatis and Infertility in Women

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Chlamydia trachomatis is recognized as human pathogens. These are the most prevalent sexually transmitted infections (STI) recognized throughout the world. Worldwide, the magnitude of morbidity associated with sexually transmitted chlamydial infections is enormous. The highest prevalence rates are found among young adults who have frequent partner change rates. Symptomatic, asymptomatic, or latent infections or their sequelae can also cause chronic inflammation of the cervix and endometrium urethritis and cervicitis, and pelvic inflammatory disease (PID), ectopic pregnancy and tubal factor infertility. Chlamydial PID is the most important preventable cause of infertility and adverse pregnancy outcome. Chlamydial infections, like STI in general, are primarily a woman's health care issue since the manifestations and consequences are more damaging to the reproductive health in women than in men. To prevent the severe sequelae and spread of disease, diagnosis and treatment of infected individuals is very necessary. Historically, cell culture technique was used for diagnosis of *C. trachomatis* infections. This was followed by the DFA (direct fluorescent antibody) techniques, and today NAATs (nucleic acid amplification tests) are most widely used techniques. However, with the availability of newer diagnostic the diagnosis has become fast and easy.

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Industrial Microbiology **P117**

Purification and Partial Identification of Bioactive Secondary Metabolites of *Streptomycetes* Strain 12M

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The increasing drug resistance in pathogens against the available antibiotics imposes a burden on the pharmaceutical industry and thus the demand for novel bioactive secondary metabolites from natural sources is increasing. In this study, biochemically characterized streptomycetes strain, 12 M was cultivated through large scale fermentation of almost 10 liters in glucose yeast-malt extract media. After fermentation, the solid phase and liquid phase of the fermented broth were separated by using bed extraction and treatment was done by using solvents i.e. methanol or ethyl acetate respectively. Then both phases of these extracts (solid and liquid phase) were purified by manual column chromatography using silica gel, preparative TLC and sephadex column. The antimicrobial activity of each fraction as well as purified compounds was determined by agar well assay and microwell plate against gram positive (MRSA) and gram negative pathogenic (*E .coli* and *Pseudomonas*) strains. Cytotoxicity of the crude extract of strain 12 M has been determined by microwell cytotoxicity assay against *Artemia salina* but cytotoxicity of the purified fraction is yet to be determined. Similarly, anticancer activity of the crude extract was determined by MTT and sulforhodamine B (SRB) against HCT116 colorectal carcinoma cell line but anticancer activity of pure fractions is yet to be determined. The crude extract of streptomycetes strain 12M showed antimicrobial activity against MRSA with 15 mm zone of inhibition, 60% cytotoxicity against Artemia salina, 71% anticancer activity against against HCT116 colorectal carcinoma cell line. In future, the fractions showing significant antimicrobial and antitumor activity will be identified through LC/MS and Nuclear magnetic spectroscopy (NMR) and maybe we will be able to report new compounds from the strain 12 M.

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Bioinformatics **P118**

Screening of Phytochemicals and Synthetic Compounds to Evaluate their Therapeutic Response Against Bipolar Disorder

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Bipolar Disorder is a neuro-psychiatric disorder, formerly called manic depression,characterized by mood swings between energy (manic) and clinical depression. It is also known as manic-depressive illness, triggering unusual changes in mood, energy, activity levels, and the capability to execute routine tasks. There are numerous genes that cause bipolar disorder, but the CACNA1C gene, encoding a subunit of the L-type voltage-gated calcium channel is one of the best-supported susceptibility genes for bipolar disorder. This gene encodes an alpha-1 subunit of a voltage-dependent calcium channel. Dysregulation of calcium ions (greater influx) is a cause of bipolar disorder. By inhibiting the extra influx of calcium ions by protein Voltage-dependent L-type calcium channel subunit alpha-1C"bipolar disorder can be treated.Through Computer Aided Drug Development (In-Silico Drug Development) the protein "Voltage-dependent L-type calcium channel subunit alpha-1C" is targeted to inhibit the abnormal influx of calcium ions to access the therapeutic response. Bioactivity of chemically synthesized compounds is compared with the phytochemicals.Sequence of protein was interpreted; 3D structure of protein is constructed based on homology modelling and active site is predicted. Phytochemicals and synthetic compounds were assessed and analyzed. Docking analysis is performed to determine the number of interactions and binding energies. Lead compound is retrieved on the basis of Lipinski rule of 5and by passing through the ADME filter. Pharmacodynamics and pharmacokinetic properties of lead compound are analyzed. Lead optimization is performed.Lead compound is procured on the basis of low IC50 value and low binding energy. Bioactivity of chemically synthesized compounds is compared with derived phytochemicals to assess the therapeutic response towards bipolar disorder.

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Environmental Microbiology **P119**

Isolation, Identification and Antibiotic Susceptibility Testing of Bacteria from Computer Keyboard of Hospital and University Settings

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Computer emerge as continuous vital device for our day to day life. But the computer keyboard is always neglected as a place of harboring of microorganism and their transmission. To isolate, identify and antibiotic susceptibility testing of bacteria from house, university and hospital settings. Seven different samples were collected from Women University Multan, Nishtar Hospital Multan and House. All of the sample collected were directly swabbed on blood agar and 30 colonies were picked on the basis of colony morphology and further purified on nutrient agar. Purified strains were identified on the basis of cell morphology and many biochemical testing include catalase test, starch hydrolysis, glucose fermentation, mannitol salt agar test, voges proskauer, citrate utilization, 6.5% NaCl and blood agar test were performed for identification.Three bacterial species: *Staphylococcus* spp, *Bacillus* spp and *Corynebacterium* spp were identified. Among these *Bacillus* species were dominated. Antibiotic susceptibility test was also performed for detection of antimicrobial resistance in strains. Majority of strains were sensitive few were resistant and intermediate, against broad spectrum antibiotics that was use with different concentration i.e. Ciprofloxacin 5 µg, Ampicillin 10 µg, Gentamycin 10µg , Amikacin 30µg, Amoxicillin 25µg and Erythromycin 15µg. Among these antibiotics majority of strains show higher sensitivity to ciprofloxacin and amikacin. Highest diversity and bacterial count were obtained from hospital settings. 100% contamination were observed and many pathogenic species were determined. As in Hospital settings more harmful bacteria isolated as compared to the University settings. So it is recommended that good personal hygiene is necessary and cleaning of computer on daily basis to lessen the number of bacteria on computer keyboard.

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Antimicrobial Agents & Chemotherapy **P120**

Antimibacterial Activity of Nanoparticles Synthesized from Citrus

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Nanoparticles have a wide range of applications in areas such as biomedical sciences, chemical industries, electronics and energy science. The biological means of nanoparticles synthesis are more advantageous than the physical and chemical methods, as it involves simple methodology that is non-toxic and obtained at a minimum cost. Silver nanoparticles were synthesized using citrus peel as a reducing agent by green synthesis. The nanoparticles synthesis was monitored by using the UV-visible spectroscopy. *Staphylococcus aureus*, *Proteus* sp., *Bacillus cereus*, *Clostridium* sp. and *Aeromonas* sp. Results were recorded by measuring the diameter of zone of inhibition. Nanoparticles showed antibacterial activity against Gram- negative as well as Gram-positive bacteria. Higher zone of inhibition (7.1±0.173 mm and 9.5±0.005 mm) was observed in case of *Clostridium* sp., from dried citrus peels. The study is helpful in describing the antibacterial potential of citrus peel synthesized nanoparticles.

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Virology **P121**

Isolation and Characterization of Lytic Bacteriophages against *Enterobacter cloacae*

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Alarming antibiotic resistance has been seen over the past few decades. One of the greatest challenges of the present time is to find out the effective strategies against antimicrobial resistance. Phage therapy could be the alternative option to combat bacterial resistance. In the present study, lytic bacteriophage EBP isolated from sewage sample and characterize at molecular level for therapeutic use. Bacteriophage shows activity against several clinical isolates of *Enterobacter cloacae*. EBP remain viable at pH 5- 8 and show stability against a temperature range from 25°C-80°C. Long term storage at low temperature cause no effect on its antimicrobial activity. 20 minutes were the latent period of EBP. Single step growth curve showed the burst size of 252 particles per cell. EBP showed narrow host range as it do not show any activity against other enterbacter species. Next generation sequencing confirmed the genome size of EBP to about 174 kb which is quite big then the normal range. This molecular characterization of EBP opens the door for further studying this phage activity and to find out its therapeutic use.

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Poster

Human Genetics **P122**

Case Control Analysis of RAD50 Gene Polymorphisms in Cancer Patients from Local Breast Population

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Breast carcinoma is characterized as multifactorial disease that can be caused by a high number of risk factors that are genetic or environment factors and has significant association with each other. Genetic alterations are contributed by mutations and genetic variants that are found in susceptibility genes of tumor or gene of DNA repair pathway. In this study we took 60 control and 60 breast cancer patients from Pakistan. Their DNA is extracted by manual method then PCR was done. The evaluation of their significant association with risk of breast cancer was accomplished by various analytical investigations. A widely accepted finding of reported research was that studied genetic variant was found in strong significant association with breast cancer incidence including the clinical factors.

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Poster

Cancer Biology **P123**

Lipid-Load in Peripheral Blood Mononuclear Cells: Impact of Food-Consumption and Dietary Macronutrients, Extracellular Lipid Availability and Demographic Factors

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Lipid content in the peripheral blood mononuclear cells (PBMCs) has recently gained attention of the researchers working on nutritional regulation of metabolic health. Previous works have indicated that the metabolic circuitries in the circulating PBMCs are influenced by dietary-intake and macronutrient composition of diet. In the present work, we analyzed in detail the impact of diet and dietary macronutrients-including carbohydrates, proteins and fats- on PBMCs' lipid-load. The overall analyses revealed that dietary carbohydrates and fats synergistically induce triglyceride accumulation in PBMCs. On the other hand, dietary fats were shown to induce significant decrease in PBMCs'cholesterol content. The effect of various demographic factors "including age, gender and body-weight- on PBMCs' lipid-load was also studied. Body-weight and age were both shown to affect PBMCs' lipid-load. Our study fails to provide any direct association between extracellular lipid availability and cellular cholesterol content in both, freshly isolated and cultured PBMCs. Cultured PBMCs and human monocytic cell line THP-1 showed increase in cellular triglyceride levels when cultivated under lipoprotein deficient medium. The presented work significantly contributes to the current understanding of the impact of food-consumption, dietary macronutrients, extracellular lipid availability and demographic factors on lipid-load in PBMCs.

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Poster

Molecular Biology **P124**

Mutataional Screening of Exon 2,19,21 and 26 of the SCN1A in Dravet Syndrome Patients

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Dravet syndrome is a common neurological disorder effects 0.5 to 1% of the world population, it appears at the age of first year. Dravet syndrome is a genetically determined disorder, about 75% cases are due to genetic variation. The de novo mutation in SCN1A gene is the major cause of dravet syndrome, about 600 de novo mutations are reported in the SCN1A gene in dravet syndrome patients. The other genes in which mutations are reported in dravet syndrome patients are PCHD19, SCN1B and GABRG2, among them 16% cases are due to mutations in PCDH19 gene.The fact is that this disorder is prevalent in Pakistani population, but no genetic study related to SCN1A gene mutation has been reported. We tried to sequence the exon 2, 19, 21 and 26 of the SCN1A gene in 5 dravet syndrome patients.In the current study no mutation was found in the coding region of the SCN1A gene and in the intronic regions nearby upstream and downstream regions. Rather, known single nucleotide polymorphisms (SNPs) were found both in coding and non coding regions of the SCN1A gene. The two SNPs (rs767750534) and (rs752789580) were identified in untranslated regions (5'UTR188A>T) and (5'UTR195A>C) in the single patient (EP-16) at the positions (166915233) and (166915243) respectively in exon 2. The exon 19 has SNP (rs763755211) in the non coding region (c.22 A>C) in one affected individual at (166868615) chromosome 2. In the case of exon 21 single SNP (rs745918943) was found in the intronic variat region (c. 28 G>C) at position (166002465). The exon 26A has a single SNP (rs565537621) in one patient (EP-15) in the untranslated region (3'UTR 320T>G) at position(166847735). In the case of exon 26B the variation (c. 495C>G) is known SNP (rs199988999) present only on the reverse strand of (EP-12). This variation is missense leads to change in amino acid (Gln>Arg). While in the case of exon 26 C no SNPs are reported.

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Poster

Food Microbiology **P125**

Assessment of Microbiological Quality of Selected Spices (Fennel and Corriander)

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Spices are used for the culinary purpose and aromatic properties to get better taste. The objective of this research was to determine the microbial contamination in several spices available in different markets. Spices are easily contaminated with pathogenic organisms. 30 packed and 40 unpacked samples of fennel and coriander were collected from different areas of the Punjab. Established and standard microbiological methods were used for the detection and enumeration of different microorganisms present in the samples using standard media. The total number of aerobic bacteria, *Staphylococcus aureus*, spore formers as well as presence or absence of *Salmonella* and *Shigella* were observed by using standard microbiological methods. High count (10^{-5}-10^{-7}) of aerobic bacteria were found in most of the samples. About more than 50% of the samples contain high amount (>10^{-3} CFU/g) of *Staphylococcus aureus*. Spore formers are also detected in large amount (>10^{-5} CFU/g) in 45% of samples. *E.coli* was detected in 2 samples only. The total count of bacteria also depends upon the type of packaging. More than 75% of packed samples contained less than (<10^{-2} CFU/g) and exhibited lower level of such organisms. *Bacillus cereus* were present in 30 samples from which mostly were polythene packaged. The overall samples collected for the study were contaminated with the microorganisms which constitute the public health risk and it alarms to improve the hygiene practice properly. Irradiation is also a tool for elimination microorganisms from dry spices.

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Poster

Antimicrobial Agents **P126**

Identification of *Bacillus* Strains Producing Antimicrobial Agents and Optimization of Agents

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Soil is considered as a promising environment for discovery and isolating the bacterial strains that are capable of producing natural products like bioactive substance against their competitors present in soil. These substance inhibit the growth of other microorganism thereby providing an advantage to these microorganism. This study is based on identification and isolation of these bioactive compounds from *Bacillus* strains, as *Bacillus* is one of the important genera that produce these substances. Firstly *Bacillus* spp. isolated from different soil samples and are selected by identification through different Morphological test, like Gram staining, Spore Staining, Capsule staining, and confirmed by different Biochemical tests, like Catalase, Oxidase, Urease test, Of glucose, Methyl red, Voges proskauer, Nitrate reduction, Denitrification, Indole, starch hydrolysis, Gelatin hydrolysis, and Hydrogen Sulfide tests. Then their antimicrobial activity is determined against Gram +ve and Gram -ve test bacteria by stabbing and agar well diffusion methods using different specialized media, like Landy's, Optimized and Brain heart infusion broth, that enhances the development of these bioactive compunds. A total of 9 *Bacillus* strains are selected on the basis of production of significant zone of inhibition of test bacteria. Next the optimized physical parameters like pH, temperature and aeration for these active strains are determined and cell free supernatants having antictobial activities were characterized on the basis of their stability to temperature, pH, and enzymes effect. Finally mimimum inhibitory concentration of these substances is evaluated. Our findings highlighted the importance of soil *Bacillus* isolates for the production of compounds with interesting bioactivities that may contribute to the drug research field.

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Poster

Environmental Microbiology **P127**

Study of Bacterial Growth Pattern and their Oil Degradation Capacity in Produced Water

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Petroleum is a very important energy source of the industrialized world. The environmental impacts in petroleum industry comes from exploration and production, transportation, refinement and product utilization. Removal of hydrocarbons from Produced Water (PW) by using biological treatment is the main focus of this study. PW is the waste water which is produced in huge quantities along with oil from subsurface during petroleum extraction. It causes contamination of aquifers and nearby areas, also hydrocarbons present in PW are carcinogenic in nature. The substances of main environmental concern are aromatic hydrocarbons including monocyclic and polycyclic aromatic hydrocarbons (PAH) along with dispersed oil, alkyl phenols (AP), heavy metals and naturally occurring radioactive material (NORM). Present study is focused in three selected sites of Eastern Potwar. Produced water samples were collected from all three sites. 47 bacterial strains were isolated out of which 36 were gram positive and 11 were gram negative. The Colony forming unit (CFU) value of location A was 3.5×10^2 CFU/ml, location B was 1.3×10^2 CFU/ml and location C was 2.3×10^3 CFU/ml. A lab scale experiment was conducted to check bacterial growth from which five strains were selected. These strains were analyzed on selected media and their growth was observed by checking their Optical Density (OD). Maximum oil degradation potential of 72% and 70% with CFU value 4.4×10^6 CFU/ml and 4.5×10^6 CFU/ml were observed in 2 strains respectively. The PW characteristics were also studied. Total Dissolved Solids (TDS) in PW of location A, B, and C were 34745 mg/L, 4124 mg/L and 64932 mg/L, respectively. Chemical Oxygen Demand (COD) of PW from location A was 4208 mg/L, location B was 695 mg/L, and location C was 11896 mg/L. The Biological Oxygen Demand (BOD) of PW samples of location A and B, and C were 703 mg/L, 174 mg/L and 506 mg/L, respectively. Further research work is in progress.

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Poster

Molecular Biology **P128**

Study of Type 2 Diabetes in Association with Gly1057Asp Polymorphism of IRS-2 Gene in the Residents of Punjab, Pakistan

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Diabetes is a complex syndrome with multiple etiologies. It occurs either due to impaired action and secretion of insulin or due to defective β-cells that ultimately leads to hyperglycemic conditions. IRS proteins have a significant part in insulin signaling pathways. Many studies have indicated increased prevalence of diabetes in the inborn population of Pakistan hence we aim to investigate the relationship between type 2 diabetes and Gly1057Asp polymorphism of IRS-2 gene in the population of Punjab, Pakistan. A total 484 samples (268 cases and 216 controls) were analyzed using PCR-RFLP assay. Allelic and genotyping frequencies were calculated and compared with anthropometric traits. The current study revealed that G1057D polymorphism is highly associated with diabetes (OR=1.62; CI=1.22-2.16; P=0.001) and age has a significant impact on this polymorphism (p=0.042).

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Industrial Microbiology **P129**

Comparative Screening of Natural Compounds from *Streptomycetes* for In Vitro Antitumor Activity by MTT and Sulforhodamine B (SRB) Bioassays Against HTC116 Colorectal Carcinoma Cell Line

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Cancer is a broad term used for malignant diseases defined as highly uncontrolled proliferation of malignant cells which collectively forms solid tumors and metastasize throughout the body, which may ultimately leads to death. The colorectal carcinoma is one of the most frequently observed cancers in developing countries, although recently a marked increase in its prevalence has also been observed in developed countries. At first stage the 50 Streptomyces extracts from the cholistan desert were screened against the brine shrimp (*Artemia salina*) for cytotoxicity or larval mortality in a microwell plate assay. The 17 Streptomyces extracts exhibited high cytotoxicity up to 60-70% larval mortality against *Artimia salina*. Further screening by MTT (3-(4, 5-dimethyl thiazol-2-yl)-2, 5-diphenyl tetrazolium bromide) and Sulforhodamine B (SRB) antitumor bioassays against HCT116 cell lines indicated a promising in vitro antitumor response of the selected *Streptomyces* strains. In case of SRB assay 5 *Streptomyces* extracts including; AK4, AK7, AK12, AK13 and AK29, were found highlyeffective which exhibited more than 80% cell mortality at 200 µg/µl, 100µg/µl and 50µg/µl dilutions, with IC 50 values 86.7, 87.7, 85.9, 84.5, 86.9µg/µl respectively. The results of % mortality by MTT and SRB was compared witheach other, the SRB antitumor bioassay provides more reliable and efficient estimation of the crude extracts of Streptomyces strains against HCT116 cell lines. The results illustrated the presence of commercially useful *Streptomyces* strains exhibiting antitumor properties against colorectal carcinoma.These *Streptomyces* strain should be investigated further by cultivating on large scale, for the purification and structure elucidation of the active compounds responsible for the antitumor activity against colorectal carcinoma.

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Industrial Microbiology **P130**

Biological Screening and Chemical Profiling of *Actinomycete* Strains Isolated from Extreme Environment

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The aim of this study was to isolate the antibiotic producing *actinomycetes* strains from the desert soil of the District Bahawalpur, Punjab, Pakistan. A total of twenty one *actinomycetes* strains were isolated from desert soil and were characterized morphologically, physiologically and biochemically. The 16S rRNA sequencing of three isolates (Q2-A, Q2-S and Q2-12) recognized them as different species of the genus Streptomyces. Conventional methods of biological screening including; cross streaked, agar plug and well diffusion assay were used to screen the bioactive actinomycetes strains against MDR bacterial pathogens. The remarkable antimicrobial activity against (multi-drug resistant) MDR pathogens was observed through the well diffusion method by using methanolic crude extracts of isolated actinomycetes. The techniques of TLC and HPLC were used for chemical screening. Overall the desert actinomycetes isolated in this study has the potential of inhibiting the multi-drug resistant pathogenic strains like MRSA, *Acinetobacter* and *Pseudomonas* etc. So the extreme desert environment holds the valuable actinomycetes in its habitat which could be valuable in getting new antimicrobial compounds.

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Immunology **P131**

Effect of Different Food Habits on CCL11 Expression; An Aging Biomarker and its Effects on Lung and Male Reproductive Tissues

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Aging can be defined as gathering of various deleterious changes in the cells and tissues which increase the risk of death and disease with increasing age, leading to some intense outcomes in various tissues of the body. Along with genetic factors, many epigenetic factors and the lifestyle of an individual also effect the process of aging. The basic aim of this study was to check the effect of different food types contributing in the process of aging. For this purpose, an aging biomarker named CCL11 was selected. Carbohydrate rich and fat rich food was selected to determine their contributory effect in elevating the expression of CCL11. To check the elevated concentration of CCL11, Enzyme linked immunosorbent assay was performed. There was an increase in the expression of CCL11 due to the carbohydrate and fat rich food while no effect was observed in the calorie restricted diet. Histology was done on the testis and lungs of mice to check the effect of elevated CCL11 on the morphology of thse tissues. Physiological defects in the morphology of testis and lungs indicated that the given type of foods contributed in inducing early aging. This study opens the doors for further studying different biomarkers in future to study the process of aging in detail.

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Pharmaceutical Microbiology **P132**

Study of Antimicrobial Activity of *Suaeda Fruticosa* against Some Pathogenic Bacteria

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To assess the leaves of *Suaeda fruticosa* for its antibacterial activity and to recognize the nature of phytochemicals in the selected plant. The leaves of *S. fruticosa* had been drawn out in different solvents and tested against the clinical bacterial isolates.The DMSO filtrate was found to be effective against the bacterial strains as compared to the methanol filtrate by giving greater zone of inhibition 2.5 cm against *K. pneumoniae*. It was also found that phytochemical extract as phenol and flavonoid giving 2.5 cm and 4cm inhibition zone against *P. aeruginosa* and *S. enterica*, respectively. Ampicillin was used as standard. The MIC of all extracts and phytochemical components was found to be effective with the minimum range of 1.25 µl out of 50ul with all the pathogenic strains and MBC was not showing visible growth. *S. fruticose* is an exemplified weed plant which has been scientifically validated as an excellent source of anti-microbials. The leaves have been proven to contain the maximum phytochemical constituents and as an excellent source of various lead molecules that can be developed into an anti-microbial drug.

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Biofilms **P133**

Multiple Metal Resistant Biofilm Producing Bacterial Strains Isolated from Paper

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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 purpose of this study was to isolate the toxic metal resistant biofilm producing bacterial strains in paper. For the present study the strains was first isolated and then further screened for multiple heavy metal resistant such as Cu, Ni, Zn, Se separately and in a combination i.e., Cu, Ni, Zn and Cu, Ni, Zn, Se at the concentration of 50, 100, 200 and 500 µg/ml. Resistant strains were picked out and further analyzed for their biofilm formation under the normal conditions as well as in case of metallic stress added 100 µg/ml. Total 02 out of 08 strains showed multiple heavy metal resistant at the concentration of 500 µg/ml. These 2 strains showed different showed different results like some metallic stress show decrease in bacterial growth over the control treatment while in some cases metallic stress was the reason of increased bacterial growth of bacterial cells. Paper as it is formed in industries so a list of steps towards its formation or manufacturing can be the cause of metal accumulation in the metal which enables the metal resistant bacteria to grow on them.

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Nanobiotechnology **P134**

Control of *Escherichia coli*, a Pollution Indicator Via its Own Biosynthesized Nanoparticles

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Good public health depends on good monitoring of water quality. Fecal contamination in fresh and marine water bodies is causing water pollution which is a serious problem resulting in various health issues. Matter of fact is the emerging infectious diseases and the development of multidrug resistance in the pathogenic microorganisms. Fecal contamination in water is measured using primary pollution indicator organisms, notably *Escherichia coli*. This study considers current gold standard analysis for *E. coli* and its remediation by using latest nano-biotechnology approach. This technique emerges as an eco-friendly against many multi-drug resistant bacteria. The test bacterium was isolated from three different fresh water samples grown on EMB Agar medium and identified as *E. coli* bacteria. Morphological and biochemical analysis was performed. The biosynthesis of selenium nanoparticles employing *E. coli* was analyzed by color change and UV-Vis spectroscopy. To confirm the presence of selenium nanoparticles XRD analysis will be performed. Further characterization of these selenium nanoparticles will be done by TEM and SEM and their antimicrobial activities will be studied against isolated bacteria *E. coli*. In conclusion, this antimicrobial activity will suggest that how these can be used for further water purification by causing death of *E. coli*.

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Environmental Microbiology **P135**

Characterization of *Bacillus amyloliquefaciens* 6A: A Novel Kerosene Oil Degrading Bacterium

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The present investigation is dealing with the use of bacterial strain (*Bacillus amyloliquefaciens*) isolated from kerosene contaminated industrial wastewater and identified biochemically and by ribotyping, to degrade kerosene hydrocarbons from aqueous and soil environments. The bacterial strain possesses kerosene degradation potential and could remove 13%, 39%, 40% and 64% kerosene oil as compared to the microorganisms indigenously present in industrial effluent which could degrade only 14%, 16%, 27% and 28% kerosene oil after 2, 4, 6 and 8 days, respectively. Kerosene biodegradation behaviour was evaluated by studying biodegradation kinetics, half-life and thermodynamics to determine the suitability of the process. Kerosene biodegradation process followed pseudo first order kinetics and found greatly influenced with temperature. Degradation rate constants and overall degradation rate was calculated from line waver burke plots obtained from Michaelis-Menten equation. Moreover, hydrocarbon fractions of kerosene oil before and after bacterial treatment were estimated by GC-MS and thin layer chromatography (TLC). Moreover, enzyme activity of crude extracellular lipase was calculated and relationship between concentration of kerosene oil and enzyme activity (oil degradation) was established by Michaelis-Menten plot. The enzyme followed the single substrate Michaelis-Menten kinetics with Vmax (maximum rate) 9.251 (µg.ml-1) min-1 and Km (Michaelis constant, substrate affinity) 8.325 $\mu g.ml^{-1}$ for kerosene oil. Furthermore, hydrocarbon fractions of kerosene oil before and after bacterial treatment were estimated by GC-MS and thin layer chromatography (TLC) which revealed the degradation of many aliphatic and aromatic hydrocarbon constituents of kerosene.

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Environmental Microbiology **P136**

Azo Dyes Decontamination Potential of Bacteria Isolated from Industrial Effluents

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Pakistan has a broad network of industries which is greatly required for the economic development of the country. Synthetic dyes, especially azo dyes are of great use in many industries such as textile, cosmetics, paper, leather and pharmaceutical etc. The discharge of wastewater from these industries proved to be fatal for the ecosystem as well as living organisms. As dyes are non- degradable so they remain stable in the wastewater. Physiochemical methods use to degrade pollutants are not economic friendly and they generate by products which are also toxic in nature. A recent approach is to treat the wastewater with microorganisms which reduces the cost of wastewater treatment. The present study was designed to isolate dye degrading bacteria and to characterize them morphologically and biochemically. Wastewater samples were collected from Kot Lakhpat industrial estate, Lahore and azo dye resistant aerobic and anaerobic bacteria were isolated. The dye degradation conditions were optimized and degradation was also checked on large scale. Microbial and phytotoxicity of this treated wastewater was also checked. TLC, HPLC and FTIR were also performed to analyze the degraded products.

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Poster

Bioinformatics & Gene Expression **P137**

Evaluation of MicroRNA and their Target Genes in Obese Population of Pakistan

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Obesity is a complicated, multifactorial disease linking environmental, genetic and metabolic factors. It has been escalating at an alarming rate globally since the past two decades to the level that it is now an epidemic affecting millions of individuals worldwide, including Pakistan. Understanding the molecular aspects of the development of fat cell and adipogenesis in obesity, it is important to identify therapeutic targets and new biomarkers for development of anti-obesity treatment. The purpose of our study was to conduct a meta-analysis to analyze obesity specific miRNAs and their differential expressed genes. Identification of a list of miRNAs that were involved in the inflammatory processes and development of obesity was conducted. We carried out functional enrichment analysis of their target genes and gene expression analysis to explore the association of the selected miRNAs in regulating pathways that might be a contributory factor of obesity. Moreover, network illustration was carried out to elucidate the inter-relationship between 7 microRNAs and their target genes. Out of 7 miRNAs, the gene expression was analyzed using qRT-PCR on 3 miRNAs hsa-miR-21-5p, hsa-miR-143-3p and hsa-miR-145-5p including their target genes. Expression on 50 obese patients and 50 controls samples was carried out. All the three miRNAs are upregulated in our study and their target genes STAT3, IL6-R, MYO5A, LOX, and TNFRSF11A except the expression of MAP3K7 showed downregulation. The dysregulation of the candidate genes shows their contribution in the development of obesity in Pakistani population. Though miRNA study needs constant investment mostly in the areas of metabolic health and diseases in which their therapeutic potential will be fully realized.

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Biochemistry **P138**

Determination of Amylase Activity by Starch Hydrolyzing Bacteria

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Starch hydrolyzing bacteria (SHB) are considered as the important source of producing amylases and have many important commercial applications in different industries such as textile, brewing, baking, paper and detergent. In the current study, we isolated and screened SHB from soil as the cheap source of these bacteria and optimum growth conditions of isolates were determined thereafter. Phenotypic and biochemical characterization indicated that isolates belong to genus *Bacillus* and confirmed by 16S rRNA genetic analysis. Both qualitative and quantitative assays were performed for determination of amylase activity. Effect of pH and temperature on enzyme activity was also determined. The effect of media composition and concentration of substrate on enzyme production and enzyme activity was revealed by inoculating bacteria in different media and by performing assays at different starch concentrations. The study revealed the optimum growth at 30˚C-45˚C and pH 7-9 for the isolates. Starch hydrolysis ratio was calculated on qualitative plate assay that was 1.8-2.8 and further confirmed by analyzing enzyme activity quantitatively in percentage; 44%-83%. In addition the identified enzyme from different isolates showed maximum activity at pH range of 5-9, with 0.1% starch concentration and at 45˚C-75˚C indicating it's thermophilic and alkaliphilic nature. Thin layer chromatography on silica gel plate was also performed to confirm the starch hydrolysis by amylolytic action. Molecular weight of amylase was 60-70 KD, determined by SDS PAGE analysis of crude enzyme by using acetone precipitation method. In conclusion, the production of amylases from indigenous amylolytic bacteria could positively affect the local industry. Such bacteria could be optimized and genetically manipulated for large scale amylase production.

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Poster

Soil & Agriculture Microbiology **P139**

Applications of *Actinomycetes* in Agriculture for Yield Enhancement and Disease Control

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Actinomycetes are gram positive filamentous bacteria, and have high GC content in their genome. They are frequently found in soil (often associated with plant roots), in water bodies, air and in remains of plants. They form filaments and are responsible for the earthy smell of fresh and healthy soil due to the production of a volatile compound called ”geosmin”. In the field of agriculture they play a lot of important roles like they are involved in the recycling of organic matter by oxidizing the organic leftovers of plants and leaving behind the nitrogenous and mineral compounds needed for the growth of plants thus improving the health and fertility of both soil and improve plant growth. Similarly they also inhibit the growth of harmful pathogens by antagonizing there activity thus they can be used as bio control agents, they produce extracellular enzymes, plant growth promoting compounds and bioactive secondary metabolites like antibiotics and stimulates plant growth even in stress conditions. *Actinomycetes* have characteristic biodegradation ability thus they can degrade pesticides and related compounds that can affect the health of soil and plant. *Actinomycetes* don’t contaminate the environment they maintain the stability of soil by the formation of compost piles, stable humus and breakdown of complex plant residues like cellulose and animal residues.

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Poster

Environmental Microbiology **P140**

Biosynthesis and Genetics of Polyhydroxyalkanoates by Newly Isolated *Pseudomonas aeruginosa* IFS and 30N using Inexpensive Carbon Sources

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Poly (3-hydroxyalkanoates) (PHAs) are the bioplastics that are stored in many genera of bacteria as carbon and energy storage polyester granules. PHAs have established themselves as strong competitors to conventional plastics. This study reports the isolation of PHA-accumulating bacteria from local environment and their PHA characterization. Two potential strains identified as *Pseudomonas aeruginosa* strain IFS (Accession no. JQ041638) and *P. aeruginosa* strain 30N (Accession no. JQ041639) based on 16S rRNA gene sequence identity were cultivated under nitrogen limited conditions to study their PHA biosynthesis capabilities. The strain IFS and strain 30N produced 1.36 and 1.40 gl^{-1} dry biomass with percentage PHA contents of 44.85 and 45.74%, respectively, when grown on glucose as carbon source. The PHA was identified as poly (3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV) by fourier transform infrared spectroscopy (FTIR) and gas chromatography coupled with mass spectrometry (GC-MS). The PHA synthase genes of these strains were isolated, sequenced and analyzed using bioinformatic tools that showed they belonging to type 2 PHA synthases and presented their evolutionary relationships with PHA synthases of other *Pseudomonas* species. The experimental results of this study highlight the importance of these strains for future use of bacterial biopolymer production growing on simple and inexpensive carbon sugars.

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Poster

Plant Microbe Interaction **P141**

Isolation and Screening of PGPR from Rhizosphere of *Cannabis sativa*

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The main objective of this study was to isolate rhizospheric bacteria from the rhizosphere of *Cannabis sativa*. Strains were identified by Gram's staining and by morphological characteristics. Final taxonomic status of the bacterial strains was confirmed by 16S rRNA gene analysis. Nine bacterial strains showed similarity with genus *Bacillus* while B16 and B17 showed similarity with *Serratia marcescens* and *Stenotrophomonas maltophilia*, respectively. The auxin production ability of the isolated strains was tested by colorimetric method in the presence and absence of L-tryptophan. Maximum auxin production in L-tryptophan amended medium was shown by *B. subtilis* (B15), *St. maltophilia* (B17) and *B. subtilis* (B11). Rooting assay and pot trials was done to check the plant gowth promoting ability of the isolated strains. Seeds of *Vigna mungo* were treated with bacterial strains single cultures and grown in culture tubes. Pot trials were performed under laboratory conditions. In rooting assay, *B. oceanisediminis* (40%) and *B. subtilis* (84%) showed significant response. In pot trials, *B. zhangzhouensis* and *B. subtilis* recorded 75% and 65% increases for shoot length, respectively, over control. For fresh weight, *B. subtilis* (34%) and *B. aryabhattai* (33%) recorded significant improvements, over control while in case of dry biomass *B. subtilis* and *Bacillus* sp. showed significant increases over control. Bacterial strains isolated from *C. sativa* showed good potential for in vitro auxin production.

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Poster

Virology **P142**

Prevalence of Acute Hepatitis B in Non-Hospitalized Patients of KPK Region

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The objective of study was to estimate the prevalence of acute hepatitis B virus and to assess the correlation of HBV serological and molecular markers for the confirmation of acute hepatitis B infection in non-hospitalized patients of KPK region. HBV is responsible for a self-limited infection known as acute HBV infection in healthy and immuno-competent individuals who recover within six months. Total of 946 samples taken from the patients were screened to detect the hepatitis B infection including 500 (52.85%) samples from males and 446 (47.15%) samples from females. It was shown that out of the total samples 212 (22.41%) samples were found positive for HBsAg. A gender based difference in the hepatitis B infection rate can be clearly seen in this study where 114 (53.77%) hepatitis B patients were males and 98 (46.23%) were females. The highest prevalence of hepatitis B was observed in the age group of 20-29 years where 62 (30.66%) patients were hepatitis B positive. In this study 53 (25%) HBsAg positive samples were anti-HBc IgM positive, categorized as acute hepatitis B patients while 135 (63.67%) HBsAg positive patients were anti-HBc IgG positive. Hence the total anti-HBc positive patients were 188 (88.67%) out of the total of 212 HBsAg positive patients. The HBsAg positive patients found negative for the antibodies such as anti-HBc IgM and anti-HBc IgG were 11.32%. In this study out of the total HBsAg positive patients 208 (98.41%) had elevated ALT levels and 197 (93%) had elevated total bilirubin levels. The HBV infection was detected by PCR amplification in 136 (64.15%) HBsAg positive patients. In this study out of the total of 136 (64.15%) PCR positive samples the HBV infection was observed in 71.69% of anti-HBc IgM and 53.84% of anti-HBc IgG positive samples.

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Medical Microbiology **P143**

Antibacterial Activity of *Lactobacilli* against Extended Spectrum B-Lactamase Producing Isolates from Post-Operative Urinary Tract Infections in Cardiac Patients

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Lactobacilli (LAB) can serve as better supportive supplements in boosting the immune response and act as remedy against life threatening infections. Isolation and characterization of LAB against Extended Spectrum β-Lactamase (ESBL) producing bacteria isolated from postoperative urinary tract infections of cardiac patients. ESBL producing isolates were characterized by using phenotypic tests including: combination disc (CD) and double disc synergy test (DDS) and PCR based detection tests. *Lactobacilli* were isolated from commercial yogurt and milk samples. Characterization was done by using biochemical battery. Well Diffusion assay was used to check antibacterial activity. Test organisms were obtained from study conducted in Punjab Institute of Cardiology (PIC), Lahore, Pakistan from Oct-2016 to Feb-2017. ESBLs producing strains were isolated from post-operative urinary tract infections (UTIs) in cardiac patients. Fourteen strains were selected as potential probiotics as they possessed strong antibacterial activity against ESBLs. These strains showed variable zone of inhibition, Heat, proteinase k and Sodium dodecyl sulphate (SDS) treatments showed that the antibacterial compound is protein in nature. *Lactobacilli* isolated from dairy products may be used against ESBLs. Isolated strains were resistant to low pH and were able to inhibit the growth of ESBLs.

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Genomics **P144**

Identification of Putative Cis-Regulatory Elements in Promoter of ADP3 Protease Gene Expressed Upon Thermo-Stress in *Arabidopsis thaliana* and its Orthologues

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In ATP-dependent protease-3 (ADP3) multifunctional enzyme involved in the proteolysis of complex, abnormal and unstable protein molecules to their units present in mitochondrial matrix. Its expression is increased upon thermal stress, suggesting that it may play a role in the heat shock response. Although, it is up-regulated upon heat stress, the underlying molecular mechanism controlling its expression is still largely unknown. Current research work intends to discover the gene-regulatory-network and molecular base controlling the expression of ADP3 by unraveling the evolutionarily Conserved Non-coding Sequences (CNSs) in the -1000 bp promoter region by analyzing upstream promoter sequences (counted from the translation initiation codon; ATG). Comparative genome-wide bioinformatic analysis performed for identification of evolutionarily preserved regulatory sequences, which revealed three highly conserved upstream non-coding sequences (CNSs). Consensus sequences from conserved sequence logo showed the position of CNS1 at -37 bp to -65 bp, CNS2 at -162 bp to -182 bp and CNS3 at -375 bp to -391 bp of *Arabidopsis thaliana* promoter counted from the ATG. Thus identified putative cis-regulatory elements in the promoter region of ADP3 gene, are expected to allow physical binding of upstream regulatory proteins which are also yet to be known. These novel putative cis-regulatory might have role in controlling the thermo-tolerance or thermo-memory response upon heat stress in *Arabidopsis thaliana* and in the plant species harboring the orthologues of ADP3 protease.

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Microbiology **P145**

Different Bacterial Strains Isolation from Spoiled Food Samples Causing Potential Gastrointestinal Infections and their Control by Home Remedy

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Bacterial gastroenteritis happens when bacteria causes an infection in gut or intestine. This causes inflammation in stomach and intestines. One may experience symptoms like vomiting, severe abdominal cramps, and diarrhea. Although viruses cause many gastrointestinal infections, bacterial infections are also common. Some people call this infection "food poisoning." Bacterial gastroenteritis can result from poor hygiene. Infection can also occur after close contact with animals or consuming food or water contaminated with bacteria (or the toxic substances produced by bacteria). The bacterial strains isolated from the raw spoiled food have different genetic and metabolic characteristics. Randomly selected samples were used for the isolation of beneficial strains, a total of twenty bacterial strains were isolated and purified. Selected bacterial strains were subjected to biochemical tests and identified as *Bacillus* sp. and *Corynebacterium* sp. predominantly. These bacteria were further subjected to antibiotic susceptibility and heavy metal resistance tests. Antibiotic susceptibility was checked against Metronidazole, Ciprpfloxacin and Ceftriaxone and heavy metal resistance against chromium and mercury. All strains were showing resistance against Metronidazole and most of them showed resistance against chromium heavy metal. Bacterial strains resistant to antibiotics were analyzed for the antibacterial activity of natural extracts of various medicinal plants. Studies are in progress to observe the genetic behavior of bacteria resistant to different antibiotics and heavy metals. In conclusion, screening of different medicinal herbs as antibacterial home remedy will be useful to replace antibiotics.

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Medical Microbiology **P146**

Isolation, Identification and Drug Resistance of Skin Aerobic Flora from Acne Individuals of Haripur City

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Acne vulgaris is chronic inflammatory disease of skin cause by pilosebaceous glands present in the skin tissues of face, chest, neck and upper back. Acne vulgar is cause by anaerobic *propionibacterium*. In case of acne, skin normal aerobic flora also involved and cause chronic acne infection. The current study deals with the isolation, biochemical identification and antibiotic susceptibility patterns of aerobic bacterial species responsible for acne vulgaris in the individuals of Haripur city. In this research study, 100 patients visiting district hospital Haripur with acne problem were enrolled during January to June 2017. After informed consent, the pus/blood samples were collected from the effect face skin area and transported via transport media to microbiology research lab. Biochemical test performed on respective isolated species. Antibiotics susceptibility was determined using Kirby Bauer disc diffusion method for a set of antibiotics like ampikacin, erythromycin, ampicillin, ceftazidin, ciprofloxcine, amoxiclline, gentamycin, ceftriaxone, cefotaxime, vancomycin, pencilline and pipercilline. Results of current study showed that 50% affected individuals were male and 50% were female. Regarding associated risk factors associated with Acne were also observed. In female higher no of cases have mild to moderate acne condition compared to male individuals. The prevalence of *Staphylococcus aureus* was found to be higher in acne individual compare to *Streptococcus pyogen* and *Staphylococcus epidermidis*. Resistance pattern analysis of the identified pathogens isolated from acne individuals was also determined. *Staphylococcus aureus* showed resistance to different antibiotics like ampicillin, ceftazidin, ceftriaxone, vancomycin. In case of *Streptococcus pyogen* all the isolated samples showed 99% resistance to pencilline, amoxicillin, vancomycin, while in the case of *Staphylococcus epidermidis* 95% resistance to amoxcilline, vancomycine and ceftrazidine was found.

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Environmental Microbiology **P147**

Arsenic Resistant Purple Non Sulphur Bacteria Isolated From Fish Pond, Rice Paddies and Industrial Effluents

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Purple non-sulphur bacteria have certain habitats, they can be found in aquatic environment such as marine water, waste water, fish pond, activated sludge etc. These can grow as photoautotrophs, photoheterotrophs, and chemotrophs. Depending on the availability of carbon source, the presence and absence of oxygen and light these switch from one metabolism to the other. These are also known to resist many heavy metals such as arsenic, nickel, copper, selenium etc. PNSB can grow aerobically as well as anaerobically. Many of these bacteria are also known to respire arsenic in the presence of different carbon sources. In this study we isolated arsenic resistant PNSB from rice paddies, fish pond and industrial effluents. Samples were taken from water and soil interface. Then enrichment was done to increase the number of PNSB. Different colonies of PNSB were obtained after spreading and these were purified by quadrant streaking. Colony morphology of PNSB were noted and different identification tests were performed. All the isolates were oxidase and catalase positive. These bacteria will be identified by 16S rRNA gene sequencing. PNSB are also known to produce hydrogen which can be used as biofuel. Such bacteria can be used for waste water treatment where they can oxidize different organic compounds along with detoxification of arsenic.

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Environmental Microbiology **P148**

Characterization of Purple Non-Sulfur Bacteria on the Basis of Carbon Source Utilization and Heavy Metal Resistance

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Purple non-sulfur bacteria are a major group of phototrophic anoxygenic microorganisms which are distributed extensively in environment especially in aquatic areas, where they appears to have a major role in respiration in the presence of oxygen and anoxic fermentation. These bacteria have different modes of nutrition and growth. In this study, we isolated these anoxygenic bacteria from rice paddies, fish pond and industrial waste. The enrichment was performed in minimal salt succinate (MSS) media followed by isolation through plating and incubation anaerobically. The isolates were gram negative rods and were pigmented in anaerobic conditions in presence of light. The ability of PNSB to utilize different carbon sources such as propionate, sodium lactate, sodium acetate, citrate, ammonium acetate and ammonium oxalate was evaluated. Many PNSB play important role in detoxification of heavy metals by converting them in less toxic form. Minimum inhibitory concentration against arsenic as well as resistance against different metals such as Pb, Hg, Co, Cr, Mo and Se were determined. Purple non-sulfur bacteria being metabolically diverse can serve a lot of different biotechnological purposes. It can play a major role in single cell protein as well as in nitrogen fixation.

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Food Microbiology **P149**

Compositional, Physicochemical and Functional Properties of Phosphocaseinate

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Globally, milk is extensively used for the production of value added dairy products owing to its rich nutritional profile. Similarly, innovative dairy products can be formulated by changing the functional characteristics of milk caseins. Current research was planned to assess the compositional, physicochemical and functional characteristics of phosphocaseinate powder prepared from buffalo milk. Chemical profiling of the tested powder revealed that it contains total protein, non casein nitrogen, non protein nitrogen, moisture, ash and total solids as 87.82±0.28, 0.79±0.02, 0.48±0.01, 4.22±0.10, 8.57±0.11 and 95.76±5.10%, respectively. Furthermore, phosphocaseinate powder contains sodium (6.79±0.39 mg/100g), potassium (2.98±0.75 mg/100g), calcium (9.68±0.08 mg/100g), magnesium (10.44±0.03 mg/100g) and phosphorus (12.25±0.05 mg/100g). Different suspensions of phosphocaseinate powder i.e. T1 (1%), T2 (2%), T3 (3%), T4 (4%) and T5 (5%) were used to optimize the functional properties. The results revealed that T5 treatment has the highest foaming stability (89.17±3.79%), foaming capacity (49.15±2.82%), gelling strength (13.54±0.09%), micellar hydration (2.30±0.11 mL/g), water absorption capacity (4.56±0.09 mL/g), oil absorption capacity (1.71 mL/g) and protein solubility (37.09±1.48%) as compared to the other treatments.

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Food Microbiology **P150**

Risk Assessment of Fumonisins in Dry Fruits Samples from Three Districts of KPK

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Mycotoxins are secondary metabolites of fungi present in different dry fruits and cause real toxic side effects on plant, animal and human. Illness caused by mycotoxins is called mycotoxicosis. Cancer may become a major problem in many developed countries. It is an epidemic disease in Europe and worldwide. The main aim of the present study was to check fuminosins presence in dry fruits. Strains of fungi mainly *Aspergillus*, *Fusarium*and *Penicillium* are responsible for fuminosins. Forty five samples were collected from different shops of Haripur, Havelian and Abbottbad, Pakistan. These forty five samples were incubated for the toxigenic fungal growth on potato dextrose agar. The isolated colonies were identified by using compound microscope. Fuminosin were extracted through methanol from ground dry fruits. After inoculation, different strains of fungi were formed. Fuminosins were quantified by ELISA reader using commercial ENZYME LINK IMMUNOSORBANT ASSAY (ELISA) kit. *Aspergillus niger*, *Fusarium verticilloides* and *Fusarium moniliforme* were the common isolated fungi. In Haripur sample maximum fuminosins concentration was 391.5ppb in A9 while minimum concentration was 61ppb.

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Soil Microbiology **P151**

Contribution of Zinc Solubilizing Bacteria in Growth Promotion and Zinc Content of Wheat

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Zinc is an imperative micronutrient required for optimum plant growth. Zinc solubilizing bacteria are potential alternatives for zinc supplementation and convert applied inorganic zinc to available forms. This study was conducted to screen zinc solubilizing rhizobacteria isolated from wheat and sugarcane, and to analyze their effect on wheat growth and development. Fourteen exopolysaccharides producing bacterial isolates of wheat were identified and characterized biochemically as well as on the basis of 16S rRNA gene sequences. Along these, 10 identified sugarcane isolates were also screened for zinc solubilizing ability on five different insoluble zinc sources. Out of 24, five strains,were selected for pot scale plant experiments. $ZnCO_3$ was used as zinc source and wheat seedlings were inoculated with these five strains, individually, to assess their effect on plant growth and development. The effect on plants was analyzed based on growth parameters and quantifying zinc content of shoot, root and grains using atomic absorption spectroscopy. Plant experiment was performed in two sets. For first set of plant experiments (harvested after 1 month), maximum shoot and root dry weights and shoot lengths were noted for the plants inoculated with *Rhizobium* sp. (LHRW1) while *E. cloacae* (PBS 2) increased both shoot and root lengths. Highest zinc content was found in shoots of *E. cloacae* (PBS 2) and in roots of *P. agglomerans* (EPS 13). For second set of plant experiment (harvested after 3 months), *Pantoea dispersa* (EPS 6), *P. agglomerans* (EPS 13) and *E. cloacae* (PBS 2) significantly increased shoot dry weights. However, significant increase in root dry weights and maximum zinc content was recorded for *Pseudomonas fragi* (EPS 1) inoculated plants. While maximum zinc content for roots was quantified in the control plants indicating the plant's inability to transport zinc to grains, supporting accelerated bioavailability of zinc to plant grains with zinc solubilizing rhizobacteria.

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Microbiology **P152**

Optimization of Amylase Production from an Endophytic Fungi *Aspergillus niger* Isolated from *Tagetes erecta*

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The present study reports the isolation of endophytic fungi from *Tagetes erecta* and its optimization for amylase production. Stem and leaves of *Tagetes erecta* were collected from campus garden of University of Haripur, KPK Pakistan. Sections were cut from the surface sterilized plant samples and incubated on Potato Dextrose Agar for fungal growth. Endophytic fungi isolated from *Tagetes erecta* plant were proceeded for amylolytic activity on starch modified agar. Among the twenty five isolates of fungi, NFL-32 showed highest activity and identified as *Aspergillus niger* was selected for further optimization studies. Effect of variouscultural parameters like incubation period, temperature, pH, nitrogen and carbon sources on amylase production was studied in liquid fermentation media. Themaximal productivity of amylase was achieved at 40ºC and at pH 7.0. Among the various carbon sources, sucrose at 1% gave the highest amylase production. Among different nitrogen sources 0.3% yeast extract was found to be optimum at the agitation rate of 200 rpm. Results revealed the diversity and the novel finding of amylase production from *Tagetes erecta* endophytes.

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Environmental Microbiology **P153**

Anoxygenic Growth of Purple Non Sulphur Bacteria in the Stress of Arsenic

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Purple non sulphur bacteria are the most metabolic diverse group of the phototrophic purple bacteria, exhibit a remarkable adaptability in their anaerobic metabolism. They are ecological important due to their participation in many biogeochemical cycles. Due to the presence of Calvin cycle in purple non sulphur bacteria, they have ability to grow as a photoheterotroph anoxically, photoautotroph and chemoheterotroph in aerobic conditions. They produce carotenoids and bacteriochlorophyll in the presence of light. Reduction in oxygen tension induced the carbon dioxide fixation, photosynthesis and anaerobic respiration. In this study, PNSB were isolated from the habitats having low concentration of oxygen, including paddy fields, fish pond, activated sludge, hospital waste water ditches. They were isolated on minimal salt succinate (MSS) enriched media.10 pigmented isolates were obtained after plating and anaerobic incubation. All isolates were gram negative rods. They acquire energy for growth by arsenic redox transformations. Fermentation of different carbon sources like oxalate, benzoate, acetate, lactate, propionate and succinate was determined for energy purposes. Minimum inhibitory concentration against arsenic and ability to resist many metals like Pb, Cr, Hg, Ni, Se, Co, Mo, was evaluated. PNSB showed the arsenite oxidase and arsenate reductase activity under anoxic conditions. Elucidation of phylogenetic structure of PNSB can be done by the analysis of 16S rRNA gene sequences. PNSB has most significant role in the purification of industrial waste water, biohydrogen and biopolyesters production.

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Environmental Microbiology **P154**

Indigenous Sulphate Reducing Bacteria of Extreme Environments of Hot Springs of Pakistan

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Anaerobic microorganisms usually thrive in extreme habitats where bacterial species adapt. Many such anaerobes are thermophiles that thrive in extreme environments such as hot-springs. Thermophilic extremophiles not only have well-adapted systems to resist local heat but also have developed the mechanisms to use varied electron donors rather than traditional oxygen which is scarce in such conditions. The sulphate reducing ability of a group anaerobes is well documented by the production of large amounts of H_2S in those areas. In this study, we attempt to isolate sulphate reducing bacteria (SRB) from hot springs of Kashmir, Pakistan, followed by their identification by 16SrRNA gene analysis in regard to special focus on sulphate reduction gene. The sulphate reducing ability will be identified via turbidimetric method that estimates the decrease in sulphate concentration in reducing cultures. Furthermore, sulphate reducing genes will also be amplified and sequenced. SRB are the most significant group among bacteria causing microbially induced corrosion (MIC) due to production of H_2S in their course of metabolism which precipitates with iron forming iron sulphides causing corrosion of iron.

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Cancer Biology **P155**

Measurement of DNA and/or Protein Concentrations as a Normalization Strategy for Lipid Data from Adherent Cell Lines

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Our research group is actively involved in studying the impact of metabolic stress on cellular lipid levels in cancer cell lines cultivated in 2D or 3D cell culture system. This data on cellular lipid levels is observed to display several unwanted variations. These variations are mainly introduced in the data because both metabolic stress and 3D cell culture systems make cell counting procedure almost impossible. Cells growing in 3D models are very tightly clumped and are not converted back to a single-cell suspension even after trypsinization. Moreover, cells under metabolic stress particularly cells cultivated under lipid-reduced medium are shrunken and distorted that makes cell counting a very difficult task. Moreover, for metabolic studies MTT assay is also not the best assay to measure cell viability. Although the results from MTT assay normally correlate with the number of viable cells growing in standard culture conditions, the rate of tetrazolium reduction reflects the general metabolic activity or the rate of glycolytic NADH production. The rate of MTT reduction can change with culture conditions (e.g., pH and glucose content of medium) and the physiological state of the cells. In addition, cells growing in 3D may have a different rate of metabolism than the cells growing in 2D system. In order to interpret our data on cellular lipid levels we needed to have an appropriate normalization strategy because ineffective or poorly chosen normalization methods can lead to erroneous conclusions. In this study, we have compared several methods for normalization of lipid data. Common choices for normalization include total protein concentration, cell count, and DNA concentration. Here, we compared and observed that all the three strategies –protein concentration, cell count, and DNA concentration– exhibit strong linear correlations with seeded cell number, but DNA concentration was found to be the most useful method.

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Cancer Biology **P156**

Measurement of DNA and/or Protein Concentrations as A Normalization Strategy for Lipid Data from Non-Adherent Cell Lines

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Our research group is actively involved in studying the impact of metabolic stress on cellular lipid levels in cancer cell lines cultivated in 2D cell culture system. This data on cellular lipid levels is observed to display several unwanted variations. These variations are mainly introduced in the data because metabolic stress makes cell counting procedure almost impossible. Cells under metabolic stress particularly cells cultivated under lipid-reduced medium are shrunken and distorted that makes cell counting a very difficult task. Moreover, for metabolic studies MTT assay is also not the best assay to measure cell viability. Although the results from MTT assay normally correlate with the number of viable cells growing in standard culture conditions, the rate of tetrazolium reduction reflects the general metabolic activity or the rate of glycolytic NADH production. The rate of MTT reduction can change with culture conditions (e.g., pH and glucose content of medium) and the physiological state of the cells. In order to interpret our data on cellular lipid levels we needed to have an appropriate normalization strategy because ineffective or poorly chosen normalization methods can lead to erroneous conclusions. In this study, we have compared several methods for normalization of lipid data. Common choices for normalization include total protein concentration, cell count, and DNA concentration. Here, we compared and observed that all the three strategies –protein concentration, cell count, and DNA concentration– exhibit strong linear correlations with seeded cell number, but DNA concentration was found to be the most useful method.

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Environmental Biology **P157**

Isolation, Identification and Characterization of Halophilic Bacteria from Saline Plains

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Halophiles belong to extremophilic group of microorganisms that can flourish in saline and hyper saline environments. Halophiles are mostly known to produce vast variety of industrially important enzymes. The objective of the study was isolation, identification, characterization, and phylogenetic analysis of halophilic bacteria. 18 Saline soil samples were collected from 6 different sites and 3 samples taken from each site of saline plains of khewra Punjab, Pakistan. Bacteria isolated from saline soil through dilution method at high concentration of NaCl. Bacterial isolates that exhibited 1.0 M salt resistance were characterized based on colony morphology. Further, characterization was doing through salt minimal inhibitory concentration, by measuring the ability of the bacteria to resist UV radiation, and bacterial tolerance against desiccation. Phylogeny of isolated halophiles will be done through 16S rRNA gene analysis. Exploring such extreme environments offer potential solutions to many industrial problems and can also give more insights into mechanisms that allow life to persist in such extreme habitats.

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Biotechnology **P158**

Bifidobacterium: An Introduction and Applications as "Probiotics"

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Bifidobacteria constitute the dominant colonic microbiota, represent up to 25% of the cultivable faecal bacteria in adults and 80% in infants. As probiotic agents, bifidobacteria have been the target from past few decades to study their efficacy in preventing and treating animal and human gastrointestinal disorders. *Bifidobacteria*, gram positive group of microorganism previously known as *Bacillus bifidus* due to its bifid morphology, are nonmotile, non-spore former and anaerobic bacteria. They have the ability to synthesize amino acids, riboflavin, thiamine and especially the vitamin B2 and B6 all of which are the requirement of healthy GIT. These all properties along with their ability to inhibit the growth of other bacteria especially in GIT promote their potential as probiotics which are defined as the live microbial feed supplements that improve the properties of indigenous microflora and improve the health of the host. In the gut flora, *B. Bifidus* produce bacteriocin named bifidin which can inhibit the growth of pathogens; such as *Enterococcus*, *Listeria*, *S. aureus* and *Pseudomonas*. Probiotics are not new, they have been consumed for years in fermented food where they maintained the efficacy of the fermented food and now a days, their addition in dairy products, increase their shelf life. Many bacterial genera have been used in making probiotics but regarding the safety concerns, the lactic acid bacterial group including *Lactobacilli*, *Bifidobacteria*, and *Streptococci* are the best one but due to the dominancy of *Bifidobacteria* in gut flora always highlight this genera for its application in probiotics. So, by using probiotics containing *Bifidobacteria* the therapeutic options can increase to prevent urogenital diseases, alleviation of constipation, prevention of infantile diarrhoea, control of inflammatory bowel diseases and irritable bowel syndrome.

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OMICS **P159**

Automated Knee Replacement Detection using Radiological Symptoms

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Knee replacement surgeries have experienced an exponential growth in the past few decades. Knee issues are very frequent among people of all ages. Osteoarthritis is one of the most common reason behind knee arthroplasty. Key features of osteoarthritis include subchondral sclerosis, joint space narrowing, osteophytes and cyst formation. These parameters are continuous and therefore result in varied decisions by the doctors. The objective of this research is to develop a computer-aided design (CAD) which can assist doctors to decide whether knee should be replaced or not. In contrast to existing CADs that use clinical symptoms, the proposed system uses radiological symptoms is this analysis. The knee radiographs are pre-processed to remove the noise and to enhance the contrast. The region of interest (ROI) is marked and histogram of gradient (HOG) features are computed. In the next step, edges of femur and tibia are detected to measure the joint space width using the Canny edge detection algorithm which gives a binary image containing edges of bones. The joint space width of knee is estimated from this binary feature using a ratio-formula. The performance of the proposed algorithm is evaluated on 50 knee radiographs, collected from a local hospital. The dataset comprises radiographs of patients suffering with different level of osteoarthritis. The key features in x-ray images that show the radiological symptoms of knee to be replaced are marked by the radiologist and orthopaedic surgeons, which show the severity of arthritis and serve as the ground truth in our study. The quantitative evaluation showed that the proposed algorithm achieved more than 80% accuracy with respect to the ground truth in measuring the joint space width. The results show that our algorithm is able to effectively measure the joint space width which can be assistive for surgeons to take decisions on knee surgery.

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Cancer Biology **P160**

Matrix Metalloproteinase 9 Expression in Squamous Cell Carcinoma of Head and Neck; A Prognostic Marker

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In tumor development, there is a mutual interaction between tumor, stromal and inflammatory cells. These all release different matrix metalloproteinases (MMPs) for extracellular matrix (ECM) degradation. MMPs affect the tumor microenvironment by degrading all ECM & Basement Membrane to rupture physical barriers, regulate & increase tumor induced angiogenesis, vascular remodeling & disrupt local tissue to promote invasion & metastasis leading to poor prognosis. To determine the MMP 9 expression in SCC and find if any relation between SCC grade and MMP 9 expression. 49 patients presenting with various histological grades of SCC were selected during study period of April 2013-Dec 2013. Clinical & gross examination findings were noted followed by immunohistochemical staining of SCC samples with anti MMP 9 antibody. Sections were microscopically scored for intensity (0-3) & proportion (0-3). Overall score was obtained by adding intensity & proportion scores. Tumors were categorized into low & high expression groups. Among 49 cases of SCC, n=18(36.7%) showed strong staining intensity (score 3) of MMP 9 antibody staining in tumor cells, n=14 (28.6%) showed moderate staining intensity (score 2), n=16(32.7%) showed weak intensity &only n=1 showed negative staining. For n=3 (6.1%) cases overall expression was weak while n=46(93.9%) cases showed high expression. Statistical relation between histological grade & overall expression was found to be significant by applying Mann-Whitney U test. Immunohistochemical findings of our study predict that MMPs produced by both tumor cells as well as adjacent stroma play significant role in tumor progression. There is marked expression of MMP 9 in SCCs with a high histological grade of malignancy. In this regard, antibodies against specific MMPs or MMP blocking agents may represent helpful approaches as adjuvant in treatment for patients at the initial stages of SCC.

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Poster

Cancer Biology **P161**

Morphological Study and Determination of Lymphocytic Infiltrate in Follicular Adenoma and Papillary Carcinoma Thyroid in Local Population of Lahore, Pakistan

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To determine the peritumoral and intratumoral lymphocytic infiltration in biopsies of patients with follicular adenoma and papillary carcinoma thyroid. 50 diagnosed cases, 25 each of follicular adenoma and papillary carcinoma thyroid were recruited according to inclusion and exclusion criteria. Relevant clinical data and morphological findings were recorded along with the density of peritumoral and intratumoral lymphocytic infiltrate was ascertained microscopically. Among 25 cases of follicular adenoma 19 (76.0%) were females and 6 (24.0%) were males and a female to male ratio was 3.1: 1. While among 25 cases of papillary carcinoma 18 (72.0%) were females and 7 (28.0%) were males and a female to male ratio was 2.1:1. This shows female predominance in both groups. The ages of patients were divided into three age groups; Group 1(G I, 10-34 years), Group 2 (G II, 35-59 years) & Group 3 (G III, 60-84 years). When the frequency of follicular adenoma and papillary carcinoma was related with age groups, on applying Fisher's Exact Test to observe the statistical relation of follicular adenoma and papillary carcinoma with age; it was found to be significant (p= 0.046). This shows young age preponderance in follicular adenoma while in papillary carcinoma greater number of cases was present in 60-84 age groups. All of the biopsy sections were examined thoroughly under the microscope to observe the lymphocytic infiltrate (both intratumoural and peritumoural) within the histological section. When this lymphocytic infiltrate was graded according to the density, it depicted a higher frequency of papillary carcinomas showing a marked degree of lymphocytic infiltrate while majority of follicular adenomas showed moderate degree of lymphocytic infiltrate.

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Poster

OMICS **P162**

Molecular Characterization of a Cytosolic Malate Dehydrogenase Gene (GhcMDH1) from Cotton

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Malate dehydrogenase (MDH) is a key enzyme that catalyzes the reversible oxidation of oxaloacetate to malate and plays an important role in the physiological processes of plant growth and development. However, cytosolic malate dehydrogenase (cMDH), which is crucial for malate synthesis in the cytosol, still has not been extensively characterized in plants. Here, we isolated a cytosolic malate dehydrogenase gene, designated as GhcMDH1, from Gossypium hirsutum and characterized its possible molecular function in cotton fiber for the first time. The cloned cDNA of GhcMDH1 is 1520 base pairs in length, and has an open reading frame of 999 base pairs, encoding for 332 amino acid residues with an estimated molecular weight of 35.58 kDa and pI of 6.35. Sequence alignment showed that the deduced amino acid sequence of GhcMDH1 protein shared a high similarity to other plant cMDHs. Confocal and immunological analysis confirmed that GhcMDH1 protein was subcellularly localized to the cytosol. Quantitative real-time PCR revealed that GhcMDH1 was constitutively expressed in all vegetative cotton tissues, with slightly lower levels in roots than stems and leaves. Interestingly, the transcripts of GhcMDH1 were detected in 5~25 days post anthesis (DPA) fibers and highly abundant at 15 DPA fibers. The total MDH activities and malate contents of cotton fibers were positively correlated with the fiber elongation rates, suggesting that GhcMDH1 may function in malate synthesis in fast fiber elongation. In agreement with this suspicion, the recombinant His-GhcMDH1 protein mainly drives the reaction towards malate generation in vitro. In conclusion, our molecular characterization of the GhcMDH1 gene provides valuable insights to further investigate the malate equilibrium in cotton fiber development.

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Poster

Human Genetics **P163**

Anthropometric and Metabolic Indices in Assessment of Type and Severity of Dyslipidemia

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It has been shown that obesity is associated with increased rates of dyslipidemia. The present work revisits the association between plasma lipid levels and classical indicators of obesity including body mass index (BMI). The significance of various anthropometric/metabolic variables in clinical assessment of type and severity of dyslipidemia was also determined. Recently described body indices, a body shape index (ABSI) and body roundness index (BRI), were also assessed in this context. For the present cross-sectional analytical study, the participants (n = 275) were recruited from the patients visiting different health camps. Participants were anthropometrically measured and interviewed, and their fasting intravenous blood was collected. Plasma lipid levels were accordingly determined. The values for different anthropometric parameters are significantly different between dyslipidemic and non-dyslipidemic participants. Receiver operating characteristics curve analyses revealed that all the tested variables gave the highest area under the curve (AUC) values for predicting hypertriglyceridemia in comparison to other plasma lipid abnormalities. BRI gave slightly higher AUC values in predicting different forms of dyslipidemia in comparison to BMI, whereas ABSI gave very low values. Several anthropometric/metabolic indices display increased predictive capabilities for detecting hypertriglyceridemia in comparison to any other form of plasma lipid disorders. The capacity of BRI to predict dyslipidemia was comparable but not superior to the classical indicators of obesity, whereas ABSI could not detect dyslipidemia.

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Poster

Microbiology **P164**

Effect of Ascorbic and Citric Acid on the Growth of Wild Type and Div Mutants of *B. subtilis*

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The basic purpose of this work is to evaluate the effect of acids on growth of spoilage bacteria *B. subtilis*. We isolate the strains of *B. subtilis* wild type and div mutants in N-agar. To check the effect of acids on the growth of wild and div mutants of *Bacillus subtilis* we use ascorbic and citric acid of 10% stock solution. Then use different concentration of acids e.g. 75ug/ml, 100ug/ml, 200ug/ml, 300ug/ml, 409ug/ ml respectively inculcated in them and noticed the growth by differing acid concentration. After this we inoculated our strains in N-broth and after 24 hour intubation we set O.D of inoculum at 0.2 for 600nm wavelength. Then gave inoculum of these strains in the N-broth with concentration of Ascorbic and citric acid 0ug/ml, 200ug/ml, 300ug/ml, 400ug/ml, also reference tubes without inoculum and incubate for 24 hour at shaker. After 24 hour take O.D of each tube and make dilutions. We Took third dilution and plated at N-agar after it incubate and count viable colonies and count CFU. This will shows the effect of Ascorbic and citric acid on the growth of *B. subtilis* cells.

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Environmental Microbiology **P165**

Effect of Temperature and Calcium Chloride on Wild Type and Div Mutants of *Bacillus subtilis*

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In the present work, effect of high temperature and calcium chloride on the wild type strain (PY79) and *div* mutants (1A314, 1A315, 1A316, 1A317, 1A318) was studied. *Div* genes are actually cell division genes that are involved in minicell production and formation of long filaments in mutants. PY79 differs from mutant strains in colony morphology especially in colony margins. At high temperature of 50^{o}C, morphology and staining behavior of mutants is greatly affected as compared to wild type. Long filaments and minicell production was observed more in *div* mutants than wild type. On growing with different concentration of calcium chloride from 10% stock (0ug/ml, 500ug/ml, 1000ug/ml, 2000ug/ml, 5000ug/ml), not only morphology and staining properties are effected but aggregation of cells was also observed. As $CaCl_2$ concentration increases, the aggregation of cells increases. Hence temperature and calcium chloride greatly affect the cell morphology, cell division properties and staining behavior of mutants having mutation in *div* genes.

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Poster

Microbiology **P166**

Isolation and Chemical Characterization of Microorganisms from Soil of Multan, Pakistan

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Soil humus is rich in nutrient, so it has vast diversity of microorganism. Due to higher temperature, the soil of Multan is excellent in harboring thermophilic bacteria as cellular volume of microorganism is too small to maintain cellular temperature that is different form ambient temperature, so cellular temperature of microorganism is the same as that of their surroundings. Bacteria that are isolated at 42℃ are characterized through biochemical testing. Mostly isolates were found gram positive microorganisms. Biological silver nanoparticles are made with the help of $AgNO_3$ by using isolated *Bacillus* species and *Corynebacterium* species and the antibacterial activity of silver nanoparticles was checked. In future research, silver nanoparticles will be used against intestinal pathogens, agricultural pathogens and multi drug resistant pathogens. Enzymes isolated from thermophilic bacteria have immense industrial applications because they are stable at high temperature. They are most suitable for biogenic nanoparticles manipulation by using metals.

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Poster

Human Genetics **P167**

GWAS Implicated Risk Variants in Different Genes Contribute Additively to Increase the Risk of Coronary Artery Disease (CAD) in the Pakistani Subjects

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Coronary artery disease (CAD) remains the single most important cause of mortality worldwide. Many candidate and GWAS genetic variants have been identified in the recent years. In the current study, we selected six SNPs from various genes that have originally been identified in GWAS studies and examined the association of SNPs individually and as a genetic risk score (GRS) with CAD and blood lipid levels. 624 (404 cases and 219 controls) subjects were genotyped for variants rs10757274 in CDKN2A gene, rs17465637 in MIA3 gene, rs7025486 in DAB2IP gene, rs17228212 in SMAD3 gene, rs981887 in MRAS gene and rs1746048 in CXCL12 gene, by TaqMan and KASPar allele discrimination techniques. Serum lipid parameters were measured using commercially available kits. Statistical analyses were done using SPSS version 22. Individually, the single SNPs were not associated with CAD ($p<0.05$). However, the combined GRS of 6 SNPs was significantly higher in cases than controls (4.89±0.11 vs 4.58±0.08, $p=0.024$). Among blood lipids, GRS showed significant positive association with serum triglycerides levels ($p=0.022$). The GRS was quantitatively associated with CAD risk and showed association with serum triglycerides levels, suggesting that the mechanism of these variants is likely to be in part at least through creating an atherogenic lipid profile in subjects carrying high numbers of risk alleles.

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Poster

Agricultural Microbiology **P168**

Evaluation of Herbicidal Activity of *Aspergillus phoenicis* Metabolites for the Management of Parthenium Weed

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Noxious alien weed parthenium (*Parthenium hysterophorus* L.) has invaded most parts of Punjab and Khyber PakhtunKhwa, Pakistan during the last two decades. Although some chemical herbicides are very effective against this weed, however, owing to ill effects of these agrochemicals, scientists are in search of nature friendlyalternatives. In the present study, metabolites of a fungus *Aspergillus phoenicis* were assessed for their herbicidal activity against parthenium weed. Metabolites were prepared by growing the fungus in malt extract broth and potato dextrose broth for three weeks. Original (X) as well as diluted (½X) metabolites were tested for their herbicidal activity in laboratory bioassays against germination and growth of parthenium. Metabolites prepared in malt extract proved more inhibitory to germination as well as shoot and root growth of parthenium than those prepared in potato dextrose broth. Original metabolites prepared in malt extract and potato dextrose broths reduced germination by 99% and 34%, shoot dry biomass by 99% and 95%, and root dry biomass by 99% and 77%, respectively. In pot trial, original (X) and concentrated (2X) metabolites prepared in malt extract broth were sprayed on 1-week, 2-week and 3-week old parthenium plants. 1-Week old plants were found the most susceptible to spray where 62% and 70% reduction in shoot dry biomass was recorded due to original and concentrated metabolites, respectively.

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Human Genetics **P169**

PCOS: "The Perfect Hormonal Storm"

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PCOS is a condition with a range of reproductive and metabolic features that affects 4-18% of women of all ages. Millions of women around the world with PCOS go undiagnosed and number of women who do not know what PCOS is, also very high in Pakistan. The present survey project was designed to check PCOS awareness and its possible risk factors in local population of Lahore. It was descriptive cross sectional study. Different educational institutes of Lahore were visited for almost 1 year (Aug 2016 – Oct 2017) to collect required data of teenage girls for present study project. A well-defined questionnaire with proper inclusion and exclusion criteria was applied for sampling. Total 350 samples from different educational institutes were collected. The criteria of age group for present study was (13 – 27). A total of 250 teenage girls were selected. The significant outcomes showed that, 48% teenage girls were interested in enriched carbohydrates diet from which 32% were suffering from obesity. Almost 43% were having family history of high blood pressure and 38% were having family history of diabetes. 20% having family history of PCOS. Almost 36% teenage girls were having acne problems, 32% were having Hirsutism and 27% were having menstrual irregularities. From present study it was concluded that teenage girls having obesity and with positive family history of PCOS and diabetes can be considered for underlying major risk factors of PCOS. Early diagnosis and treatment can avoid the possible complications. So, they should improve their dietary habits to avoid weight complications by adopting active life style along with proper physical exercise. They should have early discussion of irregular menstrual cycle and pay more attention on menstrual complications. So, avoid later on more complications like infertility in future.

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Poster

Medical Microbiology **P170**

Prevalence of Acute Hepatitis B in Non-Hospitalized Patients of KPK Region

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The objective of study was to estimate the prevalence of acute hepatitis B virus and to assess the correlation of HBV serological and molecular markers for the confirmation of acute hepatitis B infection in non-hospitalized patients of KPK region. HBV is responsible for a self-limited infection known as acute HBV infection in healthy and immuno-competent individuals who recover within six months. Total of 946 samples taken from the patients were screened to detect the hepatitis B infection including 500 (52.85%) samples from males and 446 (47.15%) samples from females. It was shown that out of the total samples 212 (22.41%) samples were found positive for HBsAg. A gender based difference in the hepatitis B infection rate can be clearly seen in this study where 114 (53.77%) hepatitis B patients were males and 98 (46.23%) were females. The highest prevalence of hepatitis B was observed in the age group of 20-29 years where 62 (30.66%) patients were hepatitis B positive. In this study 53 (25%) HBsAg positive samples were anti-HBc IgM positive, categorized as acute hepatitis B patients while 135 (63.67%) HBsAg positive patients were anti-HBc IgG positive. Hence the total anti-HBc positive patients were 188 (88.67%) out of the total of 212 HBsAg positive patients. The HBsAg positive patients found negative for the antibodies such as anti-HBc IgM and anti-HBc IgG were 11.32%. In this study out of the total HBsAg positive patients 208 (98.41%) had elevated ALT levels and 197 (93%) had elevated total bilirubin levels. The HBV infection was detected by PCR amplification in 136 (64.15%) HBsAg positive patients. In this study out of the total of 136 (64.15%) PCR positive samples the HBV infection was observed in 71.69% of anti-HBc IgM and 53.84% of anti-HBc IgG positive samples.

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Environmental Microbiology **P171**

Isolation, Identification and Characterization of Bacteria from Rhizospheric Saline Soil

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Every environment has its own unique attributes which dictate the number and type of life present in that environment. Hypersaline soil is one such environment where excess of salt restricts the growth of organisms. However halophilic and halotolerant bacteria have special mechanisms that allow them to grow in this extreme environment. The objective of this study was to explore the microbial life of rhizospheric hypersaline soil of the salt plains present near Khewra mines, Punjab, Pakistan. Rhizospheric halotolerant bacteria were isolated on a media having 1M salt concentration. These halophiles were morphologically characterized. Further characterization was done by measuring the minimum inhibitory concentration (MIC) of salt for that bacteria, ability of these bacteria to survive at UV exposure and how much desiccation they can stand. These bacteria will be identified through ribotyping method. Unculturable bacteria of this environment will be identified based on 16S ribosomal RNA analysis of metagenome. Exploring such extreme environment can provide us with unique bacteria which might be used in research conducted to understand the mechanisms that allow the survival of life in extreme conditions. These bacteria can also be the source of a number of substances of industrial and agricultural importance.

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Food Microbiology **P172**

To Determine Antimicrobial Activity of Soy Bean Extract in Milk

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Antimicrobial activity of soy bean extract was checked on four strains (E2, C1, F1 and B1) that were isolated from milk sample. Strains were purified and then identified by applying different biochemical tests. Then strains were inoculated in test tubes containing milk, soy bean extract and combination of both in different proportions. The results showed that the strains show maximum growth in milk but in the presence of extract there is retardation of their growth. The same results were obtained in plates too when agar plates containing milk, extract and combination of both were inoculated with strains. The strains showed growth with lysis zones in milk but no growth was obtained on rest of the plates in which soy bean extract was added. Later on antimicrobial activity was also confirmed by agar disc diffusion method in which discs were inoculated with suspension of strains and they were placed on agar plates containing milk, extract and combination of both. Clear zones of inhibition were obtained in milk agar plates but no zones were obtained in plates containing soy bean extract due to its antimicrobial activity. Hence it is clear that when soy bean extract is added to milk it inhibits the growth of microorganisms and increase the shelf life of milk. Further i am also aiming to check the antimicrobial activity of soy bean extract on dry milk powder and then finally to determine the sequence of protein from extract which i most likely to show antimicrobial activity.

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Poster

Medical Microbiology **P173**

Evaluation of bla SHV, bla TEM and bla OXA Encoding Clinical Isolates from Chronic Tonsillitis using Phenotypic and Molecular Technique: First Report from Pakistan

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To evaluate bla SHV, bla TEM and bla SHV encoding clinical isolates in chronic tonsillitis using phenotypic and molecular techniques. The study was conducted in Department of Microbiology and Molecular Genetics, University of the Punjab, Lahore from August, 2016 to August, 2017. Sample processing, identification and characterization of isolates was done by using (CLSI, 2016) criteria. Antibiotic susceptibility testing by using disc diffusion assay and biofilm forming ability was analyzed by ring test and slime production test. Combination disc test was used for phenotypic detection of antibiotic resistance genes. Multiplex-PCR assay was used to check presence of bla SHV, bla TEM and bla OXA genes. 16S rRNA sequencing and phylogenetic analysis was performed. Here, variable resistance pattern was observed against applied antibiotics. 100 % resistance towards azotreonam and penicillin was observed. While 60-85 % resistances were observed against cephalosporins. Biofilm formation increased with the passage of time. 77 % strains indicated positive combination disc test. Multiplex-PCR indicated 60 % strains harbored tested genes. 40 % bla SHV genes, 30 % bla TEM genes and 60% bla OXA genes were observed among selected isolates. GenBank Accession number obtained for *Klebsiella pneumoniae* was KY810693 and for *S. aureus* was KY810692. In conclusion, *K. pneumoniae* and *S. aureus* came out to be common causative agents of tonsillitis in current study. Resistance towards multiple classes of antibiotics and strong biofilms of these micro-organisms explain the chronicity and recurrent nature of the infection. bla OXA genes were frequent among genes tested.

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Microbiology **P174**

Genetic Diversity of the Wild Grey Francolin (*Francolinus Pondicerianus*) from the Region Of Mianwali Kalabagh, Punjab, Pakistan

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In Pakistan, the Grey francolin (*Francolinus pondicerianus*) is present mostly drier areas of the Indus plains and now they spread in the Thar Desert in Sindh, Thal and Cholistan deserts in Punjab. Grey Francolin also present in the lower hills of the Makran and Lasbela districts in Balochistan, the Cherat and Kohat districts of Khyber Pakhtunkhwa Province, the salt range and agro-forestry tracks of the Pothwar Plateau in the Punjab and in the Margalla hills of Islamabad. Grey Francolin mostly present at open cultivated tracks and the grassland that having bushes and hardly found at 1200m altitude in Pakistan. The genetic diversity of francolin is needed to be conserved. The study on genetic diversity of grey francolin helped us to investigate the diversity of allelic frequencies of different microsatellite DNA markers or mitochondrial DNA (mtDNA) markers. Birds were sampled from Punjab. Collection site included Mianwali Kalabagh near Indus River and University of veterinary and animal sciences Lahore. For this purpose DNA was extracted from blood by organic method. We used four primers to illustrate the polymorphism information content of grey francolin from the region of Mianwali Kalabagh Pakistan. And random pattern of gene selected from NCBI. We obtained different values of Polymorphic information content (PIC) for each primer. We observed the high values of heterozygosity that showed the high opportunity of mating to the individuals of wild francolins then the captive. So the number of variety offspring produced they have more ability to live in wild environment. After the study of genetic diversity we found that it is important to conserve the allelic frequencies in a population rather than the population number.

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Microbiology **P175**

Epidemiological Studies of Dengue Fever in District SWABI KPK

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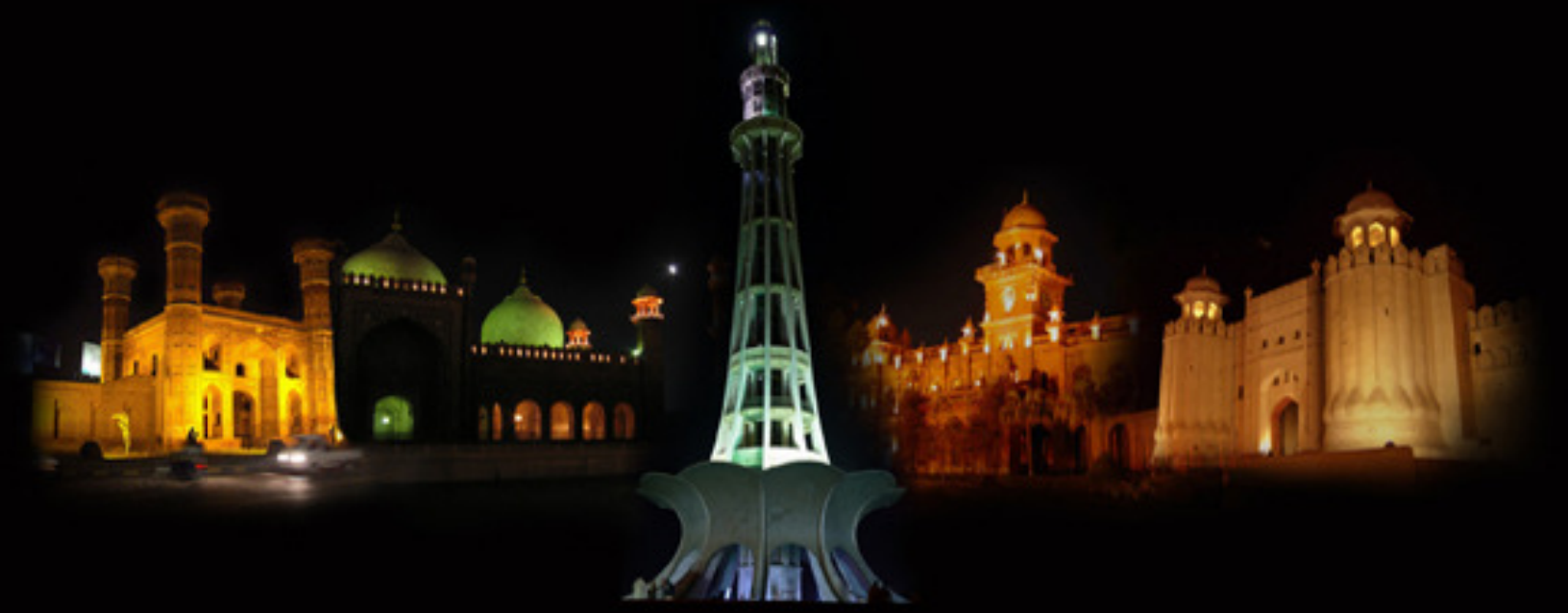
This study was set out to find out occurrence of dengue fever in district Swabi. A total of 196 blood sample were collected from patients having dengue infection on the basis of physical symptoms from Bacha Khan Medical Complex Swabi during August to October 2017. Serological test were performed for detection of IgM, IgG and NS1 against dengue. Out of 196 dengue cases, 123(62%) were male and 73(38%) were female. It was also observed that in district Swabi 21-30 age people were the main victim of dengue fever. The most affected areas in district Swabi were Topi (41%), Main Swabi (27%), Maneri (8%), Marghuz (6%), Shewa Ada (5%), Shah Mansoor (5%), Ghohati (4.5), and Chota Lahore (3.5%). The dengue fever affected patient ratio was 0.012% in Swabi. The prevalence of dengue fever was found more in male than female in Swabi areas.



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