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Molecular Characterization of Methicillin Resistant *Staphylococcus aureus* Strains Isolated in Kerala, South India

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The aim of the present study is to report the prevalence, antimicrobial susceptibility pattern molecular characteristics of methicillin resistant *Staphylococcus aureus* strains and the emergence of vancomycin intermediate *Staphylococcus aureus* (VISA) strains in Kerala, India. The study was conducted during January 2006 to December 2007 on 70 strains obtained from pus cultures of patients from various hospitals in Kerala, India. Organisms were isolated, cultured and identified as per standard routine procedures. Susceptibilities to thirteen commonly used antibiotics were tested by agar diffusion method as recommended by CLSI. Minimum inhibitory concentrations of oxacillin, ciprofloxacin and vancomycin were determined using standard protocol. Plasmid profile analysis of the strains carried out and the central resistance determinant *mec*A and internal control gene *fem*A were isolated and sequenced. Cassette chromosome typing carried out as per standard procedures. Among the 70 strains isolated 13 of them showed reduced susceptibility to vancomycin and two isolates were resistant. All the strains were resistant to oxacillin and ampicillin and uniformly sensitive to gentamycin. *mec*A gene was isolated from 88% strains and sequence analyzed. The strains were found to be Hospital Associated-MRSA (HA-MRSA) with type III cassette chromosome. This study reveals the high prevalence of MRSA and a gradual emergence of VISA strains in Kerala. This is greatly due to the irrational and overuse of antibiotics like vancomycin and partly due to negligence on the part of health care workers in acknowledging the prevalence of MRSA and VISA strains and initiating appropriate strategies to control their spread. Careful use of existing antibiotics and regular monitoring of strains circulating in a particular hospital at regular intervals is necessary to control the spread of multidrug resistant strains and to prevent the emergence of even more serious strains. *(Current Research in Bacteriology 2 (1): 1-6, 2009;* ***doi****: 10.3923/crb.2009.1.6)*

Bacterial Symbionts of Reef's Invertebrates as a Sustainable Source of Marine Natural Products

Ocky Karna Radjasa and Agus Sabdono

Marine invertebrates are mainly accumulating within coral reef ecosystems such as soft corals, sponges, tunicates and bryozoans have long been recognized as the prolific sources of structurally unique and diverse natural products since they

provide a large proportion of bioactive compounds with different biological activities. Unfortunately, the supply of these bioactive natural products is usually insufficient to meet the ultimate development of most marine natural products. The concentrations of many highly active compounds in reef's invertebrates are often minute, accounting for less than 10^{-6}% of the wet weight. This problem has been viewed as the most significantthreat regarding the development of pharmaceutical from reef's invertebrates. The secondary metabolites from bacterial symbionts, on the other hand, are a rapidly growing field, due to the suspicion that bioactive metabolites obtained from invertebrates may be produced by their bacterial symbionts. In particular, from sustainability point of view, isolating bioactive-producing bacteria is obviously offers a much better approach than cultivating and harvest invertebrates, which are in most cases extremely difficult. Bacteria isolated from living surfaces, in particular from reef's invertebrates, are a promising source of natural products. It is expected that still quite a few parts of unexplored culturable bacterial symbionts exists in the reefs. Such information might be desirable, as these bacterial symbionts may serve beneficial purposes as the source of secondary metabolites including novel marine natural products. *(Current Research in Bacteriology 2 (1): 7-13, 2009;* ***doi****: 10.3923/crb.2009.7.13)*

Phylogenetic Diversity of the Causative Agents of Vibriosis Associated with Groupers Fish from Karimunjawa Islands, Indonesia

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A molecular-based study was conducted to estimate the richness of the causative agents of vibriosis associated with groupers from Karimunjawa islands, North Java Sea, Indonesia. Moribound grouper fish were collected from the cage cultures and a total of 32 isolates were isolated from external wound and kidney of groupers. Based on the repetitive sequence-based PCR (rep-PCR) and Koch postulate test, eight isolates were chosen for further sequencings. On the basis of the sequence analysis, the data showed that the causative agents are closely related with *Vibrio natriegen*, *V. oliviceaus*, *V. fortis*, *V. alginolitycus*, *V. harveyi*, *V. parahemolitycus*, *V. damsela* and *V. carchariae*, respectively. Present study highlighted the effectiveness of rep-PCR in rapid grouping and estimating the richness of the causative agents of vibriosis associated with the groupers. *(Current Research in Bacteriology 2 (1): 14-21, 2009;* ***doi****: 10.3923/crb.2009.14.21)*

Two Pathotypes of *Xanthomonas oryzae* pv. *oryzae* Virulence Identified in West Africa

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Pathotyping analysis of 50 Xanthomonas oryzae pv. oryzae (Xoo) isolates from seven West African countries against 18 rice cultivars was carried out to identify and characterize Xoo virulence. The study revealed two major pathotypes (Pta and Ptb) of Xoo virulence. Pta has 29 virulence (Vr) Xoo isolates while Ptb has 21 mildly virulence (MVr) Xoo isolates. Pta has three subgroup pathotypes (Pta1, Pta2 and Pta3) and Ptb has two subgroup pathotypes (Ptb1and Ptb2). At country level the study revealed the presence of Pta1, Ptb1 and Ptb2 in Niger, Pta3, Ptb1 and Ptb2 in Benin and Nigeria, Pta1, Pta3 and Ptb1 in Burkina Faso, Pta1, Pta3, Ptb1 and Ptb2 in Mali, Pta1, Pta2, Pta3, Ptb1 and Ptb2 in Guinea and Pta1, Pta2, Ptb1 and Ptb2 in the Gambia. The existence of five subgroups was likely due to mutations and interactions among isolates that originally constituted Pta and Ptb pathotypes. The study revealed information on Xoo virulent population structure in West Africa as well as possible Xoo pathogen migration between these countries and this provide useful information for selection and deployment of cultivars with durable resistance to BLB disease in West Africa. *(Current Research in Bacteriology 2 (2): 22-35, 2009;* ***doi****: 10.3923/crb.2009.22.35)*

Antibiotic Susceptibility and Genetic Analysis of *Vibrio* Species Isolated from Reverine Environment

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The resistance profile and its correlation with mobile genetic elements were investigated in 11 *Vibrio cholerae,* 10 *V. parahaemolyticus*, 12 *V. vulnificus,* 11 *V. fischeri,* 10 *V. proteolyticus* and 5 *V. mimicus* isolated from River Narmada. All the 59 isolates of *Vibrio* species were examined for their susceptibility/resistance against 14 commonly used antibiotics against *Vibrio* species. More than 50% isolates showed resistance against five commonly used antibiotics viz., ampicillin, ceftadizime, erythromycin, chloramphenicol, cefuroxime. Plasmid of 6 kb was detected in 11 resistant isolates and class 1 integron was detected in 16 resistant isolates. SXT element was not found among resistant isolates. The present study indicated that plasmid and Class 1 integron mainly contributed to the circulation of multidrug resistance determinants in *Vibrio* species isolated from river Narmada. *(Current Research in Bacteriology 2 (2): 36-49, 2009;* ***doi****: 10.3923/crb.2009.36.49)*

Ceftriaxone-Sulbactam Combination: Microbial Analysis by Variation of Ratios and Comparative Disc Diffusion

S.M. Shrivastava, S. Kumar and M. Chaudhary

Development of β-lactamase provides resistance to bacteria against cephalosporins. Ceftriaxone, a third generation cephalosporin has also lost it's effectiveness in clinical practices. However, it is the current trend to use combinations of β-lactam antibiotics and β-lactamase inhibitors as they have come up as the ideal solution. The potential combination with ceftriaxone is of sulbactam, a β-lactamase inhibitor. This combination is used in clinical practice for achieving better therapeutic value. In present study, comparative microbial analysis of various ratios of ceftriaxone, sulbactam and sulbactomax, a Fixed Dose Combination (FDC) of ceftriaxone and sulbactam has been performed by Minimum Inhibitory Concentration (MIC) analysis. Comparative evaluation of susceptibility discs of FDC of ceftriaxone and sulbactam with ceftriaxone is done under time stress to find out possibility of development of resistance in *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Proteus vulgaris*, *Escherichia coli* and Methicillin Resistant *Staphylococcus aureus* (MRSA). In the results of MIC, 2:1 ratio of ceftriaxone: sulbactam has shown better bactericidal activity than the ratio of 1:6.66 and 1:3.33. Antibiotic Susceptibility Test (AST) demonstrated that ceftriaxone-sulbactam, apart from being more bactericidal, has less chances of resistance development, when compared with ceftriaxone alone. It may be concluded that ceftriaxone-sulbactam in the ratio of 2 :1 has better bactericidal properties and reduces the probability of resistance development. *(Current Research in Bacteriology 2 (2): 50-55, 2009;* ***doi****: 10.3923/crb.2009.50.55)*

Application of Random Amplification of Polymorphic DNA, Antibiogram and Serotyping for Differentiating *Streptococcus agalactiae* Clinical and Environmental Isolates from Kuwait

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The aim of this study was to assess the phenotypic and genotypic diversity among human and environmental isolates of *S. agalactiae* from Kuwait. A total of 87 isolates were collected from clinical and environmental samples. Conventional typing methods were performed by stereotyping test (slid agglutination) and antimicrobial susceptibility test (disk diffusion) method. Molecular typing method

was performed by RAPD analysis to study genetic variability at the molecular level. Fifty six of the isolates were positive for *S. agalactiae* by PCR and culture method. The bacterial isolates showed 100% sensitivity to the ampicillin and ciprofloxacin antibiotics, but 75% sensitivity to chloramphenicol and 66% sensitivity to the erythromycin antibiotics. Serotype III was predominant 26.7%, followed by serotype V, Ia and VI. Serotypes found among isolates from environment samples included V 60%, III 40%. Twelve genotypic patterns were generated using a single arbitrary RAPD primer, conventional phenotypic typing methods presented less significant discriminatory power comparing to molecular. Serologic analysis data showed to certain extent correlation with molecular data using genetic clustering and similarity indices generated by RAPD-PCR. The detection of DNA polymorphism between isolates within a serotype confirmed earlier reports of the heterogeneous nature of individual GBS serotypes. *(Research Journal of Microbiology 4 (1): 1-12, 2009;* ***doi:*** *10.3923/jm.2009.1.12)*

Helminth Contamination of Lettuce and Associated Risk Factors at Production Sites, Markets and Street Food Vendor Points in Urban and Peri-Urban Kumasi, Ghana

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The study assessed contamination levels of lettuce with helminth parasites and associated practices that may influence contamination levels at farm, market and street food vendor points in urban and peri-urban Kumasi. Three farms, three market sites that sold lettuce purchased from the selected farms and 20 street food vendors, who purchase their lettuce from these markets, were studied. Samples of lettuce, irrigation water and refreshing water (water used for keeping lettuce fresh throughout the day) were collected from these sites and analyzed for helminths eggs/larvae using standard methodology. Helminths on the lettuce leaves, irrigation water and refreshing water in the farms and markets were mostly *Ascaris lumbricoides*, with some *Shistosoma*, *Hookworm*, *Trichuris trichura*, *Taenia*, *Clonorchis* and *Strongyloides* larvae. Helminths eggs on lettuce leaves ranged between 4 and 14 100^{-1} g wet weight and 3 and 25 eggs L^{-1} in irrigation water on the farms and between 2 and 7 100^{-1} g wet weight and 4 and 15 eggs L^{-1} in refreshing water in the markets. Helminths egg counts on lettuce leaves on two farms were 40-52.9% more when compared with the farms' irrigation water but one farm had 40.5% more in irrigation water when compared with the lettuce leaves and these differences were significant. Helminths eggs on lettuce from the

two farms were 50 and 60% higher when compared with its corresponding market samples and 23.5% higher in one market when compared with its farm source. Helminths eggs in street food lettuce samples analysed from the selected areas were only *Ascaris* and *Shistosoma* eggs ranging between 0 to 2 eggs 100^{-1} g wet weight. Helminths eggs for both farm and market samples exceeded the recommended level of <1egg L^{-1}. Education on farm practices, post harvest handling and washing methods at both market and street food vendor sites and improved hygienic practices at consumer level may help reduce their numbers and minimize the risk. *(Research Journal of Microbiology 4 (1): 13-22, 2009;* ***doi:*** *10.3923/jm.2009.13.22)*

Nosocomial Legionnaires' Disease Outbreak in Tehran

R.H. Doust, A.M. Mobares, S. Mirkalantar, J. Aslani and A.I. Fuladi

The study took placed during the summer time of 2007. A 20 years old university hospital with 600 beds equipped with central air conditioning. No special disinfection program was achieved for the hospital water supplies at the time of investigation. The hospital is supplied by city water and sewages organization and is treated with standard chlorination. To analysis the first nosocomial outbreak of Legionnaires' disease in a major university hospital of Iran. Seventy Broncho Alveolar Lavage specimens were obtained from patients with pneumonia. In addition 20 water samples of various hospital points were screened for the presence of *Legionella* species and free-living amoebae. Six nosocomial cases occurred over an 8 weeks period, between the first and last case detection. *Legionella* isolates from the patients matched the water sample isolates. *L. pneumophila* were grown up from only 3 out of 70 samples, while the bacteria *mip* gene were detected from additional three cases. *L. pneumophila* (serogrup 1) were isolated from two hospital sites. Since, *Legionella* positive patients had been admitted to the hospital at least 2 weeks prior to sampling, the cases could be assumed as hospital acquired Legionnaire's disease, originated from hospital water supplies which should be treated for effective disinfection. *(Research Journal of Microbiology 4 (1): 23-30, 2009;* ***doi:*** *10.3923/jm.2009.23.30)*

Production and Freeze-Drying of Leben Lactic Starter

Z. Manel, M'hir Sana, M. Abdeslam, T. Philippe and H. Mokhtar

The production of two strains of lactic acid bacteria isolated from Tunisian fermented milk (Leben): *Lactococcus lactis* var. *lactis* (SLT6 and SLT10) was investigated in fed-batch process. The final biomass production after 8 h was

upper than 10^{10} cells mL^{-1} for both strains. The strains present an important growth rate (0.95±0.03 h^{-1}) and short generation time. The conversion yield ($Y_{x/s}$) is 0.12 and 0.14 g g^{-1} for SLT6 and SLT10, respectively. The survival after freeze-drying is 22 and 37% for SLT6 and SLT10, respectively. *(Research Journal of Microbiology 4 (1): 31-37, 2009; **doi**: 10.3923/jm.2009.31.37)*

Bio-Control of *Vibrio fluvialis* in Aquaculture by Mangrove (*Avicennia marina*) Seeds Extracts

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The microbial community associated with mangrove plant (*Avicennia marina*) in Safaga (Red Sea) was studied, the heterotrophs (TVC), *Vibrio*, *Aeromonas* and *Staphylococcus* counts in sea water were 56000, 200, 300 and 160 cfu mL^{-1}, respectively. The mangrove stems harboured lower values and the roots harboured higher values. The dominant heterotrophs isolated from the roots and stems were: *Bacillus*, *Vibrio*, *Aeromonas* and *Pseudomonas*. Different extracts of the different parts of the plant (seeds, leaves, stems and roots) were applied on different bacterial pathogens such as: *P. aeruginosa*, *V. fluvialis*, *V. vulnificus*, *S. faecalis*, *E. coli*, *S. aureus* and *B. subtilis*. The chloroform extracts showed considerable activities against the different pathogens, while the activity of the ethanol extracts showed lower values. The chloroform seeds extracts inhibited the growth of all pathogens efficiently and recorded the highest activity unit (AU = 25.0) against the fish pathogen *V. fluvialis*. Chemical composition of the extract contained carbohydrates, proteins and lipids (2.58, 0.74 and 0.074 mg), respectively, in addition to flavonoids, triterpenoids, lignin and tannin (8.6, 3, 11 and 8%), respectively. The study extended to apply these extracts on *Nile tilapia* sp. (*Oreochromis niloticus*) aquaculture, 2.5 and 5 ml L^{-1} of the chloroform seeds extracts were applied, 5 ml L^{-1} showed satisfied results while the efficiency ranged from 64.1% in the second day to 79.4% in the six day. *(Research Journal of Microbiology 4 (1): 38-48, 2009; **doi**: 10.3923/jm.2009.38.48)*

Isolation and Characterization of 3-*N*-Trimethylamino-1-Propanol Degrading *Arthrobacter* sp. Strain E5

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The aim of this study was to screen for microorganism that able to utilize 3-*N*-trimethylamino-1-propanol (homocholine) as sole source of carbon and nitrogen and to see which mechanism is followed in the degradation of this compound by

soil microorganisms. A gram-positive bacterium, designated, as strain E5 was isolated from soil. The strain was identified as *Arthrobacter* sp. strain E5 based on the phenotypic features, physiologic and biochemical characteristics and phylogenetic analysis. The cells of strain E5 displayed primary branching at the exponential phase and fragmented into irregular rod and coccoid elements at the stationary phase. The colonies were yellow in color, convex, round and entire with smooth and regular margins on both homocholine and nutrient agar medium. Comparative 16S rDNA sequencing studies indicated that strain E5 fall into *Arthrobacter nicotinovorans* subclade where it forms a monophyletic group with the type strains of *Arthrobacter nicotinovorans* and *Arthrobacter histidinolovorans*. Metabolites analysis by capillary electrophoresis and gas chromatography-mass spectrometry showed trimethylamine as a major metabolite beside β-alanine betaine and trimethylaminopropionaldehyde. Therefore, the possible degradation pathway of homocholine in *Arthrobacter* sp. strain E5 is through consequence oxidation of alcohol group (-OH) to aldehyde (-CHO) and acid (-COOH), respectively and thereafter cleavages of C-N bond providing trimethylamine and alkyl chain. *(Research Journal of Microbiology 4 (2): 49-58, 2009;* ***doi:*** *10.3923/jm.2009.49.58)*

Occurrence of Antibiotic-Resistant and Plasmid DNA Harbouring Bacterial Pathogens in Stressed Polluted Water Environment of Lake Manzala, Egypt

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This study aims at characterization of microbial pollution of Lake Manzala, bacteriological investigation of water and fish and isolation of antibiotic resistant and plasmid harbouring bacterial strains. The study revealed high levels of pollution in the water and fish samples taken from the most important sites (Kapoty, Bashtier and Mataryia areas), representative of human activity and the different ecosystems in the Lake water environment. The testing for total suspended solids (TSS), ammonia and nitrates, demonstrated that figures exceeded national and international standards. The fish-tissue samples gathered from two different sites yielded high concentration of bacterial count by plate count method. Total viable bacteria (TVB) reached more than 10^4 cfu mL^{-1} in water samples and 10^5 cfu g^{-1} in fish samples, particularly in Kapoty and Mataryia areas. Faecal coliform counts reached 10^2 cfu mL^{-1} in water samples and 10^3 cfu g^{-1} in fish samples. The API-20E test kit was used for the identification of eighty isolates of different bacteria strains. The bacterial strains *Stenthorpohomonas maltophilia, Proteus mirabilis, Escherichia coli* and *Erwinia* sp. were common species found in the samples of the study and demonstrated multi-drug resistance. These strains

harbored β-Lactamases and plasmid DNA; characteristics that can be attributed to the stressed water environment of the polluted Lake Manzala. *(Research Journal of Microbiology 4 (2): 59-66, 2009; **doi:** 10.3923/jm.2009.59.66)*

Cellulase Production by *Trichoderma longi*, *Aspergillus niger* and *Saccharomyces cerevisae* Cultured on Plantain Peel

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In this study, three fungi: *Trichoderma longibrachiatum*, *Aspergillus niger* and *Saccharomyces cerevisiae* were cultured on plantain peel, a cellulosic waste. The waste was dried, pre-treated with alkali and steam, re-dried and then blended. The powdered waste was then used as substrate in shake-flasks which contained Mineral Salts Medium (MSM) and inoculi of the three test fungi. Fermentations were initially carried out in flasks containing the MSM, waste substrate and the inoculum at pH 5.0, 1% substrate concentration, 10% inoculum size and cultured on a rotary shaker at 29±1°C for 5 days to verify cellulase production by the organisms from the waste substrates, then for 7 or 9 days while varying different fermentation parameters. Cellulase activity and amount of glucose produced by the three test organisms from the waste substrate was determined and compared. Glucose production was optimized by varying the fermentation parameters: Time, pH, Substrate concentration, Inoculum size and Temperature. The results obtained from the fermentations showed that *Trichoderma longibrachiatum* produced the highest amount of glucose among the cultures tested (1.64 mg mL^{-1}). This was produced from plantain peel at pH 5.0 and temperature of 45°C on day 7 of fermentation. The highest amount of glucose produced by *Aspergillus niger* from plantain peel was 1.18 mg mL^{-1} at pH 4.5 and temperature of 45°C on day 7 of fermentation. The highest amount of glucose produced by *Saccharomyces cerevisiae* was 1.00 mg mL^{-1} at pH 3.5 and temperature of 45°C on day 5 of fermentation. *(Research Journal of Microbiology 4 (2): 67-74, 2009; **doi:** 10.3923/jm.2009.67.74)*

Construction of pcDNA/*fim*H Cassette as a DNA Vaccine Candidate Against Urinary Tract Infection and Evaluation of *fim*H Transcripts in COS7 Cell Line

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Uropathogenic *Escherichia coli* is one of the major agents of urinary tract infection. Since it has intracellular propagation, cellular immune response is so

important in this case. Accordingly, a genetic construct for inducing of cellular immune system was designed. At first, chromosomal DNA extracted from *E. coli* 35218 and *fimH* gene amplified with this template by PCR. PCR product inserted to pcDNA.1 eukaryotic expression vector and confirmed the recombinant vector by sequencing. The COS7 cell line transfected with a complex of pcDNA/*fim*H and ExGen 500 poly cationic polymer. Expression of *fimH* gene in COS7 was confirmed by RT-PCR. Consequently, pcDNA/*fim*H cassette could express inserted *fim*H gene in eukaryotic cells and is a valuable DNA candidate cassette for urinary tract infection vaccination. This is the first prompt to designing a DNA vaccine against urinary tract infection that caused by Uropathogenic *Escherichia coli*. *(Research Journal of Microbiology 4 (2): 75-81, 2009;* ***doi:*** *10.3923/jm.2009.75.81)*

Biocatalytic Production of a Commercial Textile Dye (Indigo) from a Xenobiont

S. Mutnuri, C. Bandi and A. Ganguly

A Gram negative rod SCV1 was isolated from oil contaminated garage soil. This bacterial strain was used for the production of indigo-a commercial textile dye after induction on xenobiotics like diesel, naphthalene and salicylate. The specific rates of indigo formation are 0.30, 0.38 and 0.35 mg $mL^{-1}\times h$ for diesel, salicylate and naphthalene induced bacterial strain SCV1. The bacterial strain SCV1 was hydrophobic in nature as evident from hydrophobicity measurements. Hydrophobic nature gives the advantage to the bacterial strain in adhering to the hydrocarbons. The results of the indigo production by different substrates induced bacterial strain SCV1 suggest that the diesel induced the maximum at 1.75 and 2 mM concentrations. It is also suspected that the uninduced culture i.e., SCV1 enriched on nutrient broth produced other indigoid compounds other than indigo. *(Research Journal of Microbiology 4 (3): 82-88, 2009;* ***doi:*** *10.3923/jm.2009.82.88)*

PHA Production Using Low-Cost Agro-Industrial Wastes by *Bacillus* sp. Strain COL1/A6

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Recycling of wastes generated from agro based industries for polyhydroxyalkanoate production is not only crucial for waste management but also in economizing and commercializing the polymer. In this study, the

heterotrophic bacterium *Bacillus* sp. strain COL1/A6 isolated from humus was biologically characterized and explored for its potential to synthesize PHA using agroindustrial wastes. Qualitative analysis using Nile blue A staining revealed that starch, wafer residue, citrus pulp and cane molasses proved to be excellent carbon substrates for PHA accumulation. Growth and PHA producing ability of the isolate on cane bagasse and rice chaff improved after dilute acid hydrolysis. Highest cellular PHA content was obtained using wastes such as hydrolyzed wafer residue (62.41±1.04% of dry cell wt.) followed by cane molasses (54.68±1.36% of dry cell wt.) and hydrolyzed citrus pulp (47.5±1.01% of dry cell wt.). This is the first report wherein a *Bacillus* sp. has been reported to grow and utilize wastes such as wafer residue and citrus pulp as carbon feedstock for PHA production. *(Research Journal of Microbiology 4 (3): 89-96, 2009;* ***doi:*** *10.3923/jm.2009.89.96)*

Antimicrobial Activity of Titanium Dioxide Nanoparticles Synthesized by Sol-Gel Technique

Vilas S. Desai and Meenal Kowshik

The process of Heterogeneous Photocatalysis (HP) using titanium dioxide photocatalysts is a field of immense research potential for researchers worldwide. TiO_2 as a photocatalyst has been widely applied for air and water remediation. This study reports the synthesis of a visible light responsive nanosized TiO_2 photocatalyst by a modified sol-gel process. The synthesized TiO_2 photocatalyst exhibits photocatalytic activity against some common pathogenic microorganisms such as *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Staphylococcus aureus* under visible light illumination. TiO_2 is known to exhibit photocatalytic activity under UV light irradiation, the results obtained in this study using solar irradiation are very promising and enables the use of cheaply available solar energy for the process of photocatalysis. *(Research Journal of Microbiology 4 (3): 97-103, 2009;* ***doi:*** *10.3923/jm.2009.97.103)*

Construction and Testing of EGFP Based Bacterial Biosensor for the Detection of Residual Tetracyclines in Milk and Water

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A plasmid containing a transcriptional fusion between *tetR* regulated *tet* promoter from plasmid pOT182 and Enhanced Green Fluorescent Protein (EGFP) gene was created and was transformed into *E. coli* JM109 and this strain was used as

whole cell bacterial biosensor for detection of tetracyclines in milk and water samples. The sensor strain *E. coli* JM109 (pJSKV41) was able to detect tetracycline in the range of 10-60 ng mL^{-1} sample and oxytetracycline in the range of 25-125 ng mL^{-1} of sample. When employed for detecting residual tetracyclines in pond water samples, the biosensor strain showed high sensitivity. Also the biosensor strain was able to detect residual tetracycline in goat milk even after 4 days of tetracycline treatment. *(Research Journal of Microbiology 4 (3): 104-111, 2009; **doi**: 10.3923/jm.2009.104.111)*

Genome Wide Single Nucleotide Polymorphism Analysis of *Mycobacterium* Species and Subspecies

S.K. Srivastava, M. Agrawal and M. Grover

In this study we report the reannotation of the genome of seven *Mycobacterial* species and subspecies. We have used bioinformatics tools for annotation and reevaluated each of the Protein-Coding Sequences (CDS) previously annotated and presented the combined results of recent database searches. We have also used comparative genomic tools to focus on comparative analysis as an effective strategy. Pair wise comparison between the various *Mycobacterium* strains was performed so as to predict the relationships between them. Among the wide variety of mycobacterium strains present, we selected seven and showed how their genome is interrelated by studying synteny with the genomes of various strains studied. The genome wide SNP analysis in the seven genomes of *Mycobacterium* sp. was also done in this study and the base by base changes in the genome of these seven subspecies were identified. The gene based SNPs were further classified into the marker SNPs (SNPs which are unique amongst all the seven studied species). Out of a total of 2073 SNPs, 966 were identified as marker SNPS. This study may be used for further analysis of host pathogen interactions at the pathway and product level. The present investigation will also be useful for study of evolutionary relationship. *(Research Journal of Microbiology 4 (3): 112-121, 2009; **doi**: 10.3923/jm.2009.112.121)*

Antibacterial Activity of *Leuconostoc lactis* Isolated from Raw Cattle Milk and its Preliminary Optimization for the Bacteriocin Production

Ram Lal Thakur and Utpal Roy

Leuconocin, a bacteriocin like inhibitory substance produced by *Leuconostoc lactis* an isolate from fresh raw cattle milk was inhibitory against *Bacillus cereus*,

Staphylococcus aureus, *Enterococcus faecalis* and interestingly to the gram-negative species like *Pseudomonas putida*, *E. coli* DH5α and *E.c oli* DH5α with pUC 18 vector. The inhibitory potential was confirmed both by spot assay and cut well agar assay as well with the cell-free supernatant of the test culture in Elliker's broth. MRS broth adjusted to pH to 7.0 and 6.8, respectively produced an inhibitory zone of 15-16 mm against *B. cereus*. This promising wild-type isolate was identified up to a species level by 16S rDNA-based PCR which showed a band at about 692 bp. The set of primer used appeared to be specific as it did not amplify the closely related species. The cell-free supernatant upon concentration by 5 fold (approximately) showed a much stronger biological activity and showed heat stability. This isolate thus appears to be novel as no bacteriocin so far has been reported from *Leuconostoc lactis*. Moreover, the bacteriocin was active against both gram-positive and gram-negative organisms. *(Research Journal of Microbiology 4 (3): 122-131, 2009;* ***doi:*** *10.3923/jm.2009.122.131)*

Graph Theoretic Approach on Metabolomic Networks of Mycobacterial Strains for Potential Drug Targets

V. Baths, V.V. Rohit Kumar, G.V.R. Praneeth and U. Roy

A special strain of *Mycobacterium tuberculosis*, H37Rv's Gluconeogenesis pathway is analyzed for clusters in the pathway using the principles of spectral graph theory to find out a drug target for tuberculosis. The software named Visant was used and the data set was obtained from KEGG. The large-scale properties of chemical reaction systems, such as metabolism, can be studied with graph-based methods. To do this, one needs to reduce the information, lists of chemical reactions, available in databases. There are several ways by which this reduction can be done even for the simplest type of graph representation. Present study is aimed to apply the knowledge of graphs and graph theoretic concepts to compare the metabolic network in *Mycobacterium tuberculosis*. The study is done on the gluconeogenesis pathway, a pathway that is important for the growth of *M. Tuberculosis* H37Rv strain. Each metabolite of the pathway is taken as node of a network with the edge between the nodes representing the reaction. Spectrum and spectral radius of this network were obtained using spectral graph theory, manually. The spectral radius of this network is found out to be 0.9254. *(Research Journal of Microbiology 4 (3): 132-137, 2009;* ***doi:*** *10.3923/jm.2009.132.137)*

Inhibition of *Candida albicans* and Two Selected Gram-Negative Pathogens by Polar *Enterococcus faecalis* and *Carnobacterium* sp.

R. Shekh, K. Upadhyay, S.M. Singh and U. Roy

The current study has the objectives to identify the polar microorganisms with the ability to produce antimicrobial substances with wide-spectrum potential to antagonize the multi-drug resistance *Candida albicans*, *Pseudomonas aeruginosa* and *putida*. As many as 218 bacterial strains were screened and isolated from 6 Antarctic Penguin rookery faecal samples at Larsemann Hills, East Antarctica and from arctic sea-water-glacier stream convergence samples for checking the production of antimycotic and antibacterial substances using the cut well agar assay. Seven selected bacterial isolates were grown at 15°C for 48 h and the cell free supernatant showed activity against either *Pseudomonas aeruginosa* and *putida* or four strains of *Candida albicans*. Three selected isolates produced antimicrobial substances (AMS) which inhibited 4 strains of multi-drug resistant *Candida* sp. and two other species of *Bacillus* inhibited one Candida strain. The isolates PR 210 and 211 were found to demonstrate a very strong fungicidal agent when concentrated. The present investigation led to the findings of the three AMS producers which were identified *Enterobacter hormaechii*, *Carnobaterium maltaromaticum*, *Enterococcus faecalis*, based on *16S rRNA* gene sequences and fatty acid compositions, respectively. The other two isolates were *Bacillus megaterium* and *B. mycoides* identified by 16 S rDNA phylogenetic analysis. *(Research Journal of Microbiology 4 (3): 138-142, 2009; **doi:** 10.3923/jm.2009.138.142)*

Application of PCR-Based Fingerprinting for Detection of Nontuberculous Mycobacteria among Patients Referred to Tuberculosis Reference Center of Khuzestan Province, Iran

A.D. Khosravi, S. Seghatoleslami and M. Hashemzadeh

The present study was conducted to determine the frequency of NTM by application of PCR-based Restriction Fragment Length Polymorphism (PCR-RFLP) among suspected tuberculosis patients. In total 150 clinical isolates from patients referred to TB reference laboratory were screened. Culture and biochemical tests were performed. The PCR-RFLP method based on

amplification of a 439 bp fragment of *hsp* gene involving genus specific primers was performed and the PCR products were digested with *HaeIII* and *Bst EII* restriction enzymes. Of total isolates tested, 100 isolates were culture positive (66.6%). Eighty out of 88 isolates that were subjected to RFLP, showed the identical restriction patterns similar to *Mycobacterium tuberculosis* (90.9%). Eight clinical isolates (9.1%) showed different restriction patterns, six isolates identified as *Mycobacterium intracellularae* and two isolates were *Mycobacterium gordonae* I. In conclusion, RFLP as a fast, cheap and accurate technique is a valid alternative for phenotypic identification of pathogenic and potentially pathogenic mycobacteria in the routine laboratory. *(Research Journal of Microbiology 4 (4): 143-149, 2009; **doi:** 10.3923/jm.2009.143.149)*

Spectra of Antibacterial Activity of Propolis (Promax-C) Samples from Two Localities of Adamaoua Province (Cameroon)

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Fifteen samples of Promax-C, ethanolic extracts of propolis collected from different hives situated in two localities of the Adamaoua Province of Cameroon were tested each against seven strains of bacteria namely *Samonella enterica*, *Staphylococcus aureus*, *Escherichia coli*, *Enterococcus faecalis*, *Listeria monocytogenes*, *Pseudomonas fluorescens* and *Bacillus subtilis*. The aim of this study was to evaluate the antibacterial activity of those Promax-C samples. Antibacterial activity essays were investigated by the determination of the zones of growth inhibition using the well diffusion method on agar medium and the evaluation of the Minimal Inhibitory Concentration (MIC) using the macrodilution method. All the Promax-C samples were active against the Gram positive bacterial strains except *E. faecalis*. On the other hand, there was no activity of those samples on the Gram negative bacterial strains studied. Considering the diameter of the inhibitory zones and the MIC values, the susceptibility of bacterial strains to the Promax-C samples decreased as follows: *L. monocytogenes*> *S. aureus*>*B. subtilis*. The most active sample was Promax-C8 from the Martap locality and the most susceptible bacteria was *L. monocytogenes*. The areas of the minor and major peaks of the phenolic compounds obtained by HPLC analysis were more important for the Promax-C8 sample, showing that the greatest activity of these antimicrobial components was probably linked to their higher contents in the samples. *(Research Journal of Microbiology 4 (4): 150-157, 2009; **doi:** 10.3923/jm.2009.150.157)*

Study of Bacteria Isolated from Orthopedic Implant Infections and their Antimicrobial Susceptibility Pattern

A.D. Khosravi, F. Ahmadi, S. Salmanzadeh, A. Dashtbozorg and E. Abasi Montazeri

The aim of the present study was, to determine the bacteriology of orthopedic implant infections and susceptibilities of isolated bacteria to the commonly used antimicrobial agents. One hundred and sixty five patients were investigated for early or late postoperative infections of orthopedic bone implants using conventional microbiological procedures. Antimicrobial susceptibility testing were then performed for the isolated bacteria according to the standard guideline. A total of 155 isolates were recovered (152 aerobes and 3 anaerobes). *Staphylococcus aureus*, *Klebsiella ozaenae* and *Pseudomonas aeruginosa* were the most common causative agents. In relation to onset of infection, about 72.9% of patients were with early; 22.6% with delayed and 4.5% with late infections. The correlation between infection onset and total number of isolated bacteria was found to be statistically significant. The majority of isolated bacteria were sensitive to vancomycin, ciprofloxacine and imipenem. In conclusion, present study showed that *S. aureus* was the most common recovered bacterium with high sensitivity to vancomycin as expected. knowledge of the commonly isolated organisms and their antimicrobial susceptibility patterns within a given hospital assists in the selection of appropriate antimicrobial treatment. *(Research Journal of Microbiology 4 (4): 158-163, 2009;* ***doi:*** *10.3923/jm.2009.158-163)*

Antibacterial Potential of Herbal Formulation

Archana A. Bele, Varsha M. Jadhav, S.R. Nikam and Vilasrao J. Kadam

Natural drugs are boon to mankind. They have few side effects as compared to allopathic medicine. This invention relates to herbal composition, having potent anti-bacterial and wound healing property. The formulation prepared is a gel, which is used for effective treatment of wounds and exhibits broad spectrum antibacterial action. Crude extracts of *Punica granatum* pericarp and *Curcuma longa* showed antibacterial activity against different strains of gram positive such as *Staphylococcus aureus*, *Bacillus subtilis* and gram negative microorganisms such as *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Proteus vulgaris* and *Enterobacter aerogenes*. The MIC is recorded as the lowest concentration of drug which showed clear fluid without turbidity. Minimum

inhibitory concentration of *Punica granatum* peel ranged from 0.05 to 3.2 mg mL^{-1} and for *Curcuma longa* MIC ranged from 5 to 320 mcg mL^{-1}. Formulation containing these extracts, showed significant zone of inhibition for 0.5, 1, 2.5, 5% of which 5% showed maximum zone of inhibition (ranging from 20.2 to 26 mm) as compared to marketed preparation. The present investigation revealed that gel formulation has potential antibacterial activity. *(Research Journal of Microbiology 4 (4): 164-167, 2009;* ***doi:*** *10.3923/jm.2009.164.167)*

L-Glutaminase Production and the Growth of Marine Bacteria

P. Jeya Prakash, E. Poorani, P. Anantharaman and T. Balasubramaniam

The search of salt-tolerant and thermo-stable bacterial L-glutaminase in the marine environment was done from Coleroon estuary, Muthupet mangrove and Mullipallam lagoon which possess different marine biotopes. The isolated and identified high potent strains were subjected in to comparative study between their growth and production to select the industrially potent strains. Within that the Mullipallam lagoon strain *Vibrio* sp. SFL-2 (Sethusamudharam Field Laboratory) had produced 352.4±0.23 IU (International Unit) of L-glutaminase in the 96 h of culture but their growth rate was more or less same as other strains. *(Research Journal of Microbiology 4 (4): 168-172, 2009;* ***doi:*** *10.3923/jm.2009.168.172)*

A Real-Time Polymerase Chain Reaction Based Assay for the Detection of *Escherichia coli* in Patients with Urinary Tract Infection in the Sudan

Humodi A. Saeed, Zahra K. Yousif, Mugahid M. El Hassan, Misk El Yamen A. Atti and Mansour M. Mansour

This study was undertaken in Khartoum State, Sudan, during the period May 2007 to March 2008. A detection system based on real-time PCR has been developed for detection of *Escherichia coli* strains in patients' urine. The optimized assay format included two PCR primers. Urine specimens (46) were collected from patients attending different hospitals in Khartoum State. Bacterial DNA was extracted from each urine specimen using the Phenol-Chloroform method. Real time PCR technique was adopted to detect *E. coli*. The study revealed that 45.7% of the specimens were positive for *E. coli*. The bacterium was more prevalent in female patients than in male

patients. Adult age group was more exposed to the pathogen than the children age group. Real-time PCR technique facilitated detection of *E. coli* directly in patients' urine without a need for bacterial culture. The technology could be easily adopted in hospital settings in the Sudan. *(Research Journal of Microbiology 4 (4): 173-177, 2009;* ***doi:*** *10.3923/jm.2009.173.177)*

Thiamine (Vitamin B_1) Plays a Critical Role on Sugar Utilization by the Phytopathogenic Fungus, *Ustilago esculenta*

Kuang-Ren Chung and Dean D. Tzeng

Ustilago esculenta, inducing edible galls in its host *Zizania latifolia*, exhibits an obligate requirement for thiamine (vitamin B_1) in axenic culture. The function of thiamine for growth in *U. esculenta* was investigated and compared with two closely related species, *U. maydis* (corn smut) and *U. scitaminea* (sugarcane smut). Sucrose was readily broken into glucose and fructose, independent of thiamine, by all three fungal species tested. Growth of *U. maydis* and *U. scitaminea* was apparently not affected by thiamine when glucose or fructose was used as the sole carbon source. By contrast, *U. esculenta* was incapable of utilizing glucose and fructose in the absence of thiamine. Addition of thiamine into a synthetic medium drastically enhanced the growth of *U. esculenta*. In all cases, *Ustilago* species preferentially utilized glucose prior to fructose. Fructose uptake in *U. esculenta* exhibited a saturated kinetic, indicative of carrier protein-mediated process. The uptake of fructose by *U. esculenta* was highly influenced by the amounts of glucose, and was likely via., a noncompetitive mode. Taken together, the results strongly indicate that thiamine plays a key role for glucose and fructose metabolisms and energy production by *U. esculenta*. *(Research Journal of Microbiology 4 (4): 178-185, 2009;* ***doi:*** *10.3923/jm.2009.178.185)*

Sero Diagnosis of *Bluetongue virus* Infection and Isolation of Virus in Embryonated Chicken Egg and BHK-21 Cell Line

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Isolation of *Bluetongue virus* from blood samples of sheep and goat was carried out in the present study. Out of one fifty blood samples screened for

seroprevalance of BTV antibodies by Agarose Gel Precipitation Test (AGPT) 42 gave positive results. The overall percentage of virus isolation was 28% from Embryonated Chicken Eggs (ECE). The identities of the isolates were confirmed by cytopathogenicity. All the isolates were passaged twenty one times in embryonated chicken eggs and further passaged in BHK-21 cell lines. The viral isolates adapted well to the cell culture system and produced cytopathic change like grouping of cells, polycaryon, syncytica formation, acidophilic and intracytoplasmic inclusion bodies in BHK-21 cells. This study confirms the BTV incidence in the tested blood sample with a possible means showing that the virus can easily adapt to ECE and BHK-21 cell line. *(Research Journal of Microbiology 4 (5): 186-193, 2009;* ***doi:*** *10.3923/jm.2009.186.193)*

Bacterial Isolates from Ethiopian Soda Lake Producers of Alkaline-Active β-Glucanases Resistant to Chelating and Surfactant Compounds

M. Minig, D. Walker, P. Ledesma, M. Alejandra Martínez and Javier D. Breccia

β-glucanase activities were screened from isolated bacteria of the Ethiopian Shala Lake. Five isolates were selected according to the highest production of alkaline-active β-glucanase. By sequence analysis of 16S rDNA and physiological tests, four strains (SES01, SES22, SES4 and SES05) were related to *Bacillus halodurans* specie and the strain SES33 was identified up to genus level as *Bacillus* sp. Intergenic spacer regions fingerprinting showed different patterns among selected strains, having DNA amplicons of high molecular weight characteristic of alkaliphilic *Bacillus*. Herein, *B. halodurans* SES01 produced a highly stable β-glucanase in presence of surfactant and chelating compounds (sodium lauryl sulphate, Triton X-100 and EDTA) indicating its potential as additive for laundry technologies. *(Research Journal of Microbiology 4 (5): 194-201, 2009;* ***doi:*** *10.3923/jm.2009.194.201)*

Effect of Yeast Extract Supplementation on Curdlan Production from Condensed Corn Distillers Solubles

Thomas P. West

The effect of yeast extract supplementation on bacterial curdlan production using a medium containing corn syrup and the corn-based ethanol coproduct condensed corn distillers solubles was determined. Curdlan was produced by

Agrobacterium sp. ATCC 31749 on a medium containing selected solubles concentrations as a source of nitrogen and corn syrup as a carbon source. The presence of yeast extract in the medium was found to enhance bacterial curdlan production at all three concentrations of solubles tested after 120 h of growth. Bacterial biomass production was also noted to be higher after 120 h when the cells were supplemented with yeast extract. It was concluded that the observed increase in curdlan production by the yeast extract-supplemented ATCC 31749 cells was due to the yeast extract stimulating biomass formation. *(Research Journal of Microbiology 4 (5): 202-207, 2009;* ***doi:*** *10.3923/jm.2009.202.207)*

Date-Palm Fruit Spoilage and Seed-Borne Fungi of Saudi Arabia

Hashem Al-Sheikh

The seeds and fruits of different date palm varieties were collected from local market and brought to the laboratory of the Department of Biology, College of Science, King Faisal University, in Al-Hassa, Saudi Arabia, where further experiments for isolation of fruit spoilage and seed-borne fungi were conducted by using common technique of wet blotter method. A total number of 100 seeds and 100 cubes (1 cm^3) obtained from the fruits (10 pieces per plate) were put on wet filter paper and incubated at 25°C to allow the growth of fungi for a period of 1 week. Fungal species developed on seeds and fruit pieces were isolated on potato dextrose agar for identification. This study was carried out during year from May 2007 to April 2008. Twenty species from 14 genera of fungi have been isolated from 13 different varieties of date-palm as seed-borne fungi while 39 species of 16 genera of fungi were isolated as fruit spoilage fungi. *Alternaria alternata*, *Aspergillus flavus*, *A. niger*, *Fusarium oxysporum* and *F. solani* were the predominant species in both seed-borne and fruit spoilage fungi. *(Research Journal of Microbiology 4 (5): 208-213, 2009;* ***doi:*** *10.3923/jm.2009.208.213)*

Screening of the Efficacy of Some Commonly Used Antibiotics in Ghana

G.K. Helegbe, L.Y. Anyidoho and F.N. Gyang

The objective of this study was to screen some commonly used antibiotics in Ghana for their efficacy in treating diseases so as to select sensitive organisms that

can be used to design an assay in assessing their biological activity. The disc susceptibility test was used to screen stock antibiotics such as ampicilline, chloramphenicol, kanamycin and penicillin based antibiotics from different manufacturers (both local and foreign) which were obtained from different pharmacy shops against some bacteria species such as *Salmonella typhi*, *Staphyloccus auresus* and six strains of *Escherichia coli*. It was observed that both stock and field antibiotics (Antibiotics obtained from pharmacy shops for study) zone of inhibition were similar and compared with literature values. J916 (an *E. coli* isolate) and *Salmonella typhi* were found to be less sensitive to the penicillin-based antibiotics similar to literature values for both stock and pharmacy shop samples. This study revealed that the antibiotics produced by local and foreign pharmaceutical companies appear to be effective. In as much as this study demonstrate that, local and foreign pharmaceutical industries appear to be producing quality drugs, further studies are needed to substantiate this claim observed by this study, which was on a small scale. *(Research Journal of Microbiology 4 (6): 214-221, 2009;* ***doi:*** *10.3923/jm.2009.214.221)*

Captive Dogs as Reservoirs of Some Zoonotic Bacteria

Maha A. Sabry

The present study is an attempt to clarify the role of captive dogs as a source of some zoonotic bacteria to their contacts or vise versa. Bacteriological examination of fecal swabs evidenced infection by 3 enteric bacteria in attendants, puppies and dogs. *Salmonella* (20, 33.3 and 41.67%), *Campylobacter* (13.33, 33.3 and 33.3%) and *Enteroinvasive E. coli* (46.66, 46.67 and 58.33%). Serotyping of these bacteria revealed presence of *S. typhimurium* in dogs (60%) and attendants (66.67%), *S. enteritidis* in one of the worker as well as four untyped strains. Two serotypes of *Campylobacter* as *C. jejuni* in two workers and four dogs, *C. coli* in three dogs, while two untyped isolates were recorded in dogs. Three serotypes of *E. coli* (O 26, O 76 and O 55) and two untyped strains were isolated from workers and dogs. Moreover two isolates (O 5 and O 111) were diagnosed from dogs only. The isolates showed high sensitivity for *Gentamycin* (10 μg) and *Tetracyclin* (30 μg). The study recommended some precautionary measures to minimize the role of captive dogs as a potential source of zoonotic pathogens. *(Research Journal of Microbiology 4 (6): 222-228, 2009;* ***doi:*** *10.3923/jm.2009.222.228)*

Epidemiology of Dermatophytes in the Eastern Province of Saudi Arabia

Hashem Al Sheikh

This study was conducted for one year period during March 2008 to February 2009 in the Eastern Province of Saudi Arabia. Out of a total 250 samples collected during this period 178 (71.54%) were found positive. The dermatophytes causing different types Tinea were *Epidermatophyton floccossum*, *Microsporum canis*, *M. gypseum*, *Trihophyton mentagrophytes*, *T. rubrum*, *T. schoelneinii*, *T. soudanense*, *T. violaceum* and *T. verrucossum*. Besides these non-dermatophytes fungi *Candida albicans*, *C. krusei*, *C. tropicalis* and *Fusarium solani* were also isolated causing infection at different sites of human body. Samples from females yielded higher percentage of dermatophytes as compared to males. The percentage of infection of T. capitis and T. corporis were found to be higher in the age group of 0-15 years, while, T. pedis and T. cruris dominates in the age group of 16-30 years. Orychomycosis was dominated among the age group of 31-45 followed by 46-60 years. While, above 60 years yielded very low percentage of dermatophytes. Present study showed that more females were affected by dermatophytes (almost double in number) than males. Result of present study clearly indicates that the epidemiology of dermatophytes significantly differs from other regions of Saudi Arabia. *(Research Journal of Microbiology 4 (6): 229-234, 2009;* ***doi:*** *10.3923/jm.2009.229.234)*

Rock Phosphate Solubilization by Two Isolates of *Aspergillus niger* and *Penicillium* sp. and their Promotion to Mung Bean Plants

W.I.A. Saber, K.M. Ghanem and M.S. El-Hersh

Isolation and identification of rock phosphate (RP) solubilizing fungi were studied under laboratory conditions. Fungal isolates that displayed the highest ratio of clear zone/colony diameter on plates of phosphate solubilization medium, were selected and identified as *Aspergillus niger* and *Penicillium* sp. The optimum condition for RP solubilization were found to be at the 6th (*A. niger*) and 7th (*Penicillium* sp.) day of incubation with shaking (150 rpm) at 30°C and pH ranging from 5.6 to 6.0. Glucose followed by fructose and xylose supported the RP solubilization process in the presence of 2.5 g L^{-1} RP as the optimum concentration. The overall soluble P after optimization studies on RP were 99.7 (*A. niger*) and 77.5 mg L^{-1} (*Penicillium* sp.). During the fermentation process, there was remarkable

reduction in the final culture pH. The titratable acidity was positively correlated with RP solubilization. Under NaCl salt stress both fungi were able to solubilize RP, in which, *A. niger* was more tolerant than *Penicillium* sp. The dual and individual cultures of fungi solubilized sources of phosphate commonly exist in soil and also, possessed phytase activity. Under *in vivo* conditions, the inoculation of mung bean seeds with *A. niger* and/or *Penicillium* sp. in the presence of RP or calcium superphosphate (CSP), increased significantly the growth (except for branches No. plant^{-1}), seed yield and P-uptake, as well as, improved the nodulation status and population of total and phosphate dissolving fungi in the rhizospheric soil of mung bean. These inoculations saved about 1/3 phosphate fertilizer dose. Hereby, these combined effects encourage the potential use of the isolated fungi in the biosolubilization of RP in soil plant system. *(Research Journal of Microbiology 4 (7): 235-250, 2009;* ***doi:*** *10.3923/jm.2009.235.250)*

Optimization of Media and Cultivation Conditions for Alkaline Protease Production by Alkaliphilic *Bacillus halodurans*

Abdelnasser S.S. Ibrahim and Ali A. Al-Salamah

Media and cultivation conditions were investigated to optimize alkaline protease production by alkaliphilic *Bacillus halodurans*. This includes different carbon, nitrogen and metals sources in addition to different pH, incubation temperature and aeration level. The specific enzyme activity was increased by about 48.8 fold by optimizing different nutrient sources and cultivation conditions. The maximum specific enzyme activity was obtained in a medium containing 15 g L^{-1} lactose as the carbon source, 6 g L^{-1} soybean as the nitrogen source and a 5 mM mixture of Mg, Mn and Ca as trace elements, fermentation for 48 h at 37°C and agitation at 200 rpm. This study indicated the significance of nutrient source and cultivation conditions on the alkaline enzyme production by *Bacillus halodurans*. *(Research Journal of Microbiology 4 (7): 251-259, 2009;* ***doi:*** *10.3923/jm.2009.251.259)*

Isolation of Extreme Halotolerant Bacteria from Asian Desert Dust; Molecular Phylogeny and Growth Properties of their Cells

H. Sasaki, E. Iwata, A. Oshima, A. Ishida and S. Nagata

We tried the isolation of halophilic bacteria from Asian desert dust falls in Japan and growth property of these bacteria and their molecular phylogeny were analyzed. Two Gram-positive bacteria designated as IMU-1 and IMU-2 were

isolated from Asian desert dust. These two strains were adapted with 0-3 and 0-4 M NaCl under nutrient medium culture conditions, respectively, showing the properties of halotolerance. Under the Davis minimal medium culture condition, IMU-1 attained to the similar level of growth as that of nutrient medium culture and growth was observed at 0-2.5 M NaCl. On the other hand, IMU-2 showed the different growth as that of nutrient medium culture condition and growth was observed at 0-1.2 M NaCl. Phylogenetic analysis using 16S rRNA gene sequences revealed that IMU-1 and IMU-2 belong to *Bacillus licheniformis* and *B. megaterium*, respectively. It was first study about the isolation of *B. licheniformis* as the halophilic bacteria in Japan. *(Research Journal of Microbiology 4 (7): 260-268, 2009;* ***doi:*** *10.3923/jm.2009.260.268)*

Antimicrobial Activity of the Methanolic and Crude Alkaloid Extracts of *Acalypha wilkesiana* cv. *macafeeana* Copper Leaf

C.N. Ezekiel, C.P. Anokwuru, E. Nsofor, O.A. Odusanya and O. Adebanjo

The antimicrobial activity of methanolic leaf extracts and crude alkaloid extracts of *A. wilkesiana* cv. *macafeeana* was evaluated after a preliminary phytochemical screening of the leaf extracts. The standard agar well diffusion method was used in the bioassay involving test bacteria and yeast isolates, while percentage inhibition of extracts on radial growths of the molds was determined. The Minimal Inhibitory Concentrations (MIC) and Minimal Bactericidal Concentrations (MBC) were also determined by the broth microdilution assay technique. The microorganisms used were clinical strains of *Escherichia coli*, *Salmonella typhi*, *Streptococcus pyogenes*, *Strept. pneumonia*, Methicillin-Resistant *Staphylococcus aureus* (MRSA), non-methicillin resistant *Staph. aureus*, *Candida albicans*, *Aspergillus fumigatus* and *A. flavus*. Alkaloids, tannins, terpenoids and cardiac glycosides were extracted by the methanol solvent. The crude alkaloid extracts inhibited only the Gram-negative bacteria with mean inhibition zones of 10.0±0.00 to 12.3±0.03 mm while the methanol extracts inhibited all other test organisms, a broad spectrum activity. The water extracts had no activity against the non-MRSA strains. The MIC was 0.4 mg mL^{-1} for all unicells except strains of *C. albicans* which both had MICs of 0.8 mg mL^{-1}. The MBC was 0.4 mg mL^{-1} for tested isolates except the non-MRSA and *C. albicans* which had MBCs of >12.0 mg mL^{-1} and 1.0 mg mL^{-1}, respectively. The methanolic extract totally inhibited all tested aspergilli while the water extract

had a varying inhibitory effect (63.0±2.50 to 81.0±2.90%) on the tested fungi strains. The alkaloid had no effect on the molds. *(Research Journal of Microbiology 4 (7): 269-277, 2009;* ***doi:*** *10.3923/jm.2009.269.277)*

Klebsiella pneumoniae Producing CTX-M-15 Genes from Neonatal Intensive Care Unit in Saudi Arabia

M.H.M. Al-Agamy, A.M. Shibl, A.F. Tawfik and A.R. Elbannai

Reports on outbreak of extended-spectrum β-lactamases (ESβLs) by Enterobacteraiceae and especially *Klebsiella pneumoniae*, are few in Saudi Arabia. This study was therefore devoted to describe the outbreak which occurred by ESβL-producing *K. pneumoniae*. Sixteen *K. pneumoniae* isolates were isolated from 16 neonatal patients hospitalized from September 2007 to December 2007 in the neonatal intensive care unit during the outbreak in Al-Qatif Hospital, Eastern Province, Saudi Arabia. These isolates were sent to microbiological laboratories, College of Pharmacy, King Saud University, for investigation. *Klebsiella pneumoniae* strains were found to produce antibiotic resistance and produce extended spectrum beta-lactamase. Genotypic characterization of extended spectrum beta-lactamase producing *K. pneumoniae* showed that all isolates carried TEM-1, SHV-1 and CTX-M-15 genes. Matting out assay revealed that all third generation cephalosporins were located on transferable plasmid. An outbreak which occurred in neonatal intensive care unit was due to CTX-M-15-producing *K. pneumoniae* isolates either single or in multiple clones. This is the first report of $bla_{\text{CTX-M-15}}$ gene in Saudi Arabia from *K. pneumoniae* and the first outbreak in Saudi hospitals due to CTX-M-15 producing *K. pneumoniae. (Research Journal of Microbiology 4 (7): 278-285, 2009;* ***doi:*** *10.3923/jm.2009.278.285)*

Synergistic effect of *Trichoderma* and *Rhizobium* on Both Biocontrol of Chocolate Spot Disease and Induction of Nodulation, Physiological Activities and Productivity of *Vicia faba*

W.I.A. Saber, K.M. Abd El-Hai and K.M. Ghoneem

Experiments were carried out to correlate the biochemical features of *Trichoderma* species and *Rhizobium leguminosarum* to both biocontrol of *Botrytis fabae* and improving the productivity of faba bean. Of several

Trichoderma species, isolated from phyllosphere of faba bean, six isolates, which grew considerably faster than *B. fabae* and have moderate to very good antagonism against this pathogen, were selected. *Trichoderma*'s growth inhibiting properties of *B. fabae* were due to the combined action of non-volatile and volatile metabolites (with antibiotic nature) and the secretion of cell-wall degrading enzymes. *Trichoderma viride* (tag3 and tag4) and *T. harzianum* tag7 have shown to be efficient mycoparasites on *B. fabae* (in which the mycelium appeared to be fragmented hyphae, vacuolated and disrupted as a result of *Trichoderma* parasitism). These three *Trichoderma* isolates were further applied in field of faba bean combined with *R. leguminosarum* which, the chromatographical analysis of its supernatant showed activity in growth promoter substances. The dual inoculation of seeds with a mixture of *R. leguminosarum* and *T. viride* tag4 then foliar spraying of the developed plants with the spore suspension of the same *T. viride* tag4 at the 35th and 55th day from sowing reduced chocolate spot disease and enhanced nodulation, nitrogenase activity and nitrogen fixing bacterial population in the rhizosphere. In addition to the improvements in the physiological activities (photosynthetic pigments, total phenol and polyphenol oxidase), plant growth and yield. On average, this treatment recorded about 57% reduction in chocolate spot disease and 23% increase in faba bean yield, compared to control plants. Therefore, a commercial production of an inoculum based on a mixture of *Rhizobium* and *Trichoderma* is very encouraged. *(Research Journal of Microbiology 4 (8): 286-300, 2009;* ***doi:*** *10.3923/jm.2009.286.300)*

Single Cell Oil Production by an Oleaginous Yeast Strain in a Low Cost Cultivation Medium

Husain A. El-Fadaly, Noura El-Ahmady El-Naggar and El-Sayed M. Marwan

An oleaginous yeast strain, *Cryptococcus curvatus* NRRLY-1511 was used for the production of single cell oil (SCO) using a low cost cultivation medium containing beet molasses and corn gluten meal as carbon and nitrogen sources. Obtained results showed that 125 and 0.130 g L^{-1} showed to be the optimum concentrations for carbon and nitrogen, respectively. In addition, 28°C, 72 h, 5.5, 200 rpm were the favorable values of growth temperature, incubation period, pH value of cultivation medium and agitation speed, respectively. The extracted lipids were mainly 30.68% linoleic acid (C18:2), 22.66% oleic acid (C18:1) and 16.74% palmitic acid (C_{16}:0). Furthermore, the GC analysis also showed that the total saturated fatty acids (n = 9) represented 41.96% while the value of the total unsaturated fatty acids (n = 6) was 58.04%. These results giving possibility to use

such this yeast strain to produce SCO in a low cost medium from economic point of view. *(Research Journal of Microbiology 4 (8): 301-313, 2009; **doi:** 10.3923/jm.2009.301.313)*

Antibacterial Activity of Seagrass Species Against Biofilm Forming Bacteria

P. Mayavu, S. Sugesh and V.J. Ravindran

The present study was carried out on antimicrobial properties of seagrass species against biofilm forming bacteria's from boat hull during the period April 2008 to March 2009. Seagrass species have a very potential groups were producing several secondary metabolites. The bioactive potential of two different seagrass species viz., *Cymodocea serrulata* and *Syringodium isoetifolium* occurring commonly along the Tuticorin coastal area were selected and preliminary effort has been made against the marine biofilm forming bacteria's *Pseudomonas aeruginosa*, *Bacillus cereus*, *Proteus vulgaris*, *P. mirabilis*, *E. coli*, *Listeria monocytogenes*, *Salmonella enteritidis*, *Staphylococus aureus* and *Vibrio paraheamolyticus*, which also the human pathogens. The seagrasses of *C. serrulata* and *S. isoetifolium* were extracted with four different solvents such as ethanol, methanol, acetone and dichloroethane. Ethanol and methanol extracts of *S. isoetifolium* was inhibited the biofilm forming bacteria such as *E. coli* (14 mm), *P. aeruginosa* (8 mm) and *V. paraheamolyticus* (7 mm) and it showing Minimum activity against *S. aureus* (2 mm). The crude extract of ethanol and methanol of *C. serrulata* was inhibited the growth of all the 9 species of the biofilm forming microbes. The results of present study were concluded that seagrasses have potential bioactivity against marine biofilm forming microorganisms. *(Research Journal of Microbiology 4 (8): 314-319, 2009; **doi:** 10.3923/jm.2009.314.319)*

In vitro Susceptibility of *Naegleria fowleri* Trophozoites to Amphotericin B-combined Chlorpromazine

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The objective of this study was to study the susceptibility of *Naegleria fowleri* trophozoites to Amphotericin B-combined chlorpromazine investigated the activities of single drugs used in combination with amphotericin B compared to those of each drug alone *in vitro*. The 50% inhibitory concentrations (IC_{50}) and 100% minimal concentrations (MIC_{100}) were calculated for single drugs and the

drugs combination with fixed combination ratios of IC_{50} of amphotericin B. Single drugs, amphotericin B had the best IC_{50} and MIC_{100} scores against *N. fowleri* trophozoites. chlorpromaxine, Artesunate and azitromycin had following IC_{50} and MIC_{100} scores against trophozoites. However, we found that chlorpromazine in combination with amphotericin B was the best synergistic drug against *N. fowleri* trophozoites. According to single drugs, chlorpromaxine, artesunate and azitromycin plus amphotericin B had also been synergistic drugs against *N. fowleri* trophozoites. It was suggested that the combined use of these agents may be beneficial in treating Primary amoebic meningoencephalitis. *(Research Journal of Microbiology 4 (9): 320-333, 2009;* ***doi****: 10.3923/jm.2009.320.333)*

Enteric Bacteria Associated with Farmed Freshwater Fish and its Culture Environment in Kerala, India

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A study was designed to investigate the enteric bacterial population associated with farmed freshwater fish and its environment, limnological quality of carp farm and the existing association between these parameters. Enteric indicator bacterial counts were determined following the United States Food and Drug Administration (USFDA) methods and the physico-chemical parameters according to the standard methods of American Public Health Association (APHA). Fish samples yielded mean microbiological counts in the range of 4.19 to 4.85 log CFU g^{-1}, sediment in the range of 5.18±0.01 to 6.34±0.01 log CFU g^{-1}, pond water in the range of 3.64±0.03 to 6.10±0.04log CFU mL^{-1}. Fish and feeder canal water showed higher count for all indicator bacterial count. Sediment showed 2 log cycle higher count of sulphite reducing *clostridia*. Emerging pathogen *E. coli* O157:H7 were absent in all the samples analyzed. *Aeromonas* (26.2%) followed by *Enterobacter* (24.6%) were the dominant flora recovered. *Escherichia*, *Klebsiella*, *Serratia*, *Hafnia*, *Plesiomonas*, *Shigella*, *Salmonella*, *Morganella* and *Yersinia* were the other opportunistic enteric bacterial pathogens detected from this system. The rearing practices such as natural fertilization and feeding could have influenced the enteric flora. Study on the various physico-chemical parameters of pond water revealed that they were within the suitable range for the freshwater fish culture throughout farming phase. Correlation analysis revealed a significant positive correlation between physico-chemical parameters such as total organic carbon (TOC), Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD) with that of Total Plate Count (TPC), Total *Enterobacteriaceae* Count (TEC), Total Coliforms (TC), Faecal Coliforms (FC) and *E. coli* (EC). Presence of bacteria of

public health significance in the aquaculture ponds envisages a strict hygienic handling and processing of fish from the culture systems for ensuring public health safety. *(Research Journal of Microbiology 4 (9): 334-344, 2009;* ***doi:*** *10.3923/jm.2009.334.344)*

Swimming Motility in *Agrobacterium tumefaciens* is Controlled by Quorum Sensing and Inhibited by Garlic Bulb Extract

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Bacteria can produce and sense signal molecules, allowing the whole population to initiate a concerted action once a critical concentration (corresponding to a particular population density) of the signal has been reached; a phenomenon known as Quorum Sensing (QS). The current study was conducted to examine the possible role of QS in the regulation of swimming motility of *Agrobacterium tumefaciens*. In addition, we investigated the anti-QS or Quorum-Quenching (QQ) activity of garlic bulb and *Salvadora persica* extracts. We found that treatment of *A. tumefaciens* culture with different exogenous QS compounds induced swimming motility. C4 AHL, C6 AHL, C7 AHL, C8AHL, C10 AHL and C14 AHL induced bacterial swimming motility by about 3.5, 4, 4.5, 4.5, 3.5 and 4 fold, respectively, providing strong evidence that quorum sensing in *A. tumefaciens* controls cell motility, or at least plays a major role in its regulation. We also found that different QS compounds affect the bacterial phenotype, including the colony pattern and morphology. In addition, garlic bulb and *Salvadora persica* extracts were investigated for their QQ activity. While *S. persica* extract did not show any significant QQ activity, garlic bulb extract showed QQ activity against C4 AHL, C8 AHL, C10 AHL and C14 AHL, repressing the *A. tumefaciens* swimming motility induced by these QS compounds. To the best of our knowledge, this is the first report of a possible role for QS in the regulation of swimming motility in A. tumefaciens. *(Research Journal of Microbiology 4 (9): 345-354, 2009;* ***doi:*** *10.3923/jm.2009.345.354)*

Microbiological Evaluation of the Quality of Tap Water Distributed at Khartoum State

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This study was aimed to evaluate the microbial quality of drinking water distributed at Khartoum state- the capital of the Sudan. Water distributed at piped system was investigated using two different standard methods (MPN and chromogenic media- based techniques), 47.5-90% showed positive isolation of bacteria. The results revealed isolation of faecal coliform (*E. coli*), coliform group (*Klebsiella*

sp., *Citrobacter* sp., *Enteriobacter* sp.), some pathogenic and potential pathogenic bacteria (*Staphylococcus aureus*, *Salmonella* sp., *Yersienia enteriocolitica*, *Proteus* sp., *Bacillus* sp. and *Pseudomonas aeruginosa*) were isolated. Other bacteria with significant importance were detected. The quality of drinking water, types and number of isolated bacteria were evaluated and discussed according to seasons and locations. *(Research Journal of Microbiology 4 (10): 355-360, 2009; **doi:** 10.3923/jm.2009.355.360)*

Viability of Antifungal Metabolite Producing *Pseudomonas* Bacteria

M.S. Shathele and A. Fadlelmula

The objectives of this study were to determine the suitability of transport medium (ice jells) and estimate the duration of viability of *Pseudomonas* in the transport medium. Bacteria of the genus *Pseudomonas* comprise a large group of the active biocontrol strains as a result of their general ability to produce a diverse array of potent antifungal metabolites. These include simple metabolites such as 2,4-diacetylphloroglucinol, phenazine-1-carboxylic acid and pyrrolnitrin [3-chloro-4-(2-nitro-3-chlorophenyl)-pyrrole], as well as the complex macrocyclic lactone, 2, 3-de-epoxy-2, 3-didehydro-rhizoxin. Pyrrolnitrin is active against *Rhizoctonia* sp., *Fusarium* sp. and other pathogenic fungi and it has been used as a lead structure in the development of a new phenylpyrrole fungicide. The survival rates of four different pseudomonad strains after continuous incubation for 4 h in the cold temperature (4°C) were: 94.8% for *P. putida* strain CBD, 94.5% for *P. aeruginosa* No. BRCH and 62.1% for *Pseudomomas* species *(fluorescent)* with lowest survival rate of 33.5% for *P. aeruginosa* strain H. Since, there were no drastic reductions in the survival rates, the study findings suggest that the transport medium would be generally suitable for these cold-sensitive bacteria. *(Research Journal of Microbiology 4 (10): 361-365, 2009; **doi:** 10.3923/jm.2009.361.365)*

In vitro Activity of Some Antimicrobial Agents against *Staphylococcus aureus* and Methicillin-Resistant *Staphylococcus aureus* in Khartoum, Sudan

H.A. Saeed and W.B. Ahmed

Staphylococcus aureus is a causative agent of many types of diseases throughout the world. Patients hospitalized for long period of time usually are predisposed to infection by methicillin resistant *S. aureus* (MRSA). The objectives of the present study were to evaluate the efficacy of some antimicrobial agents against *S. aureus* and MRSA and to select the most effective antibiotic. Clinical specimens were

collected from patients with wounds and/or urinary tract infections. The specimens were proceeded for isolation of the pathogens. Identification was done by conventional methods. Antimicrobial sensitivity test was carried out using modified Kirby-Bauer Disc Diffusion Technique in accordance with National Committee on Clinical Laboratory Standards (NCCLS). Of 163 *S. aureus* recovered, 15 (9.2%) isolates were MRSA. The most effective antimicrobial agent against both *S. aureus* and MRSA was vancomycin (99%). The activity of the rest antimicrobial agents was cephalexin, 92%, methicillin, 90%, cloxacillin 33%, penicillin 14% and amoxicillin 10%. It is concluded that vancomycin may be an alternative antibiotic for patients with wound and/or urinary tract infections caused by *S. aureus* or MRSA. *(Research Journal of Microbiology 4 (10): 366-369, 2009;* ***doi:*** *10.3923/jm.2009.366.369)*

Cyclodextrin Glycosyltransferase Production by the *Bacillus* sp., Subgroup *alcalophilus* using a Central Composite Design

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Cyclodextrin glycosyltransferase (CGTase) activity was produced by the *Bacillus* sp., subgroup *alcalophilus* in a culture medium containing cassava starch. A central composite design and response surface methodology were used to study the influence of carbon source (cassava starch), nitrogen sources (yeast extract and tryptone) and sodium carbonate in the production medium. Assays were performed in 300 mL Erlenmeyer flasks containing 100 mL of production medium maintained in a shaker at 150 rpm at 35±1°C for 72 h of fermentation. The independent variables [0.75% cassava starch, nitrogen sources (0.375% yeast extract and 0.375% tryptone) and 1% Na_2CO_3] produced an enzyme activity of 96.07 U mL^{-1}. *(Research Journal of Microbiology 4 (11): 450-459, 2009;* ***doi:*** *10.3923/jm.2009.450.459)*

Enumeration and Identification of Pathogenic Pollution Indicators in Cauvery River, South India

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This study was aimed to estimate current levels of pollution indicator as well as many groups of human pathogenic bacteria and their seasonal variations in different locations of Cauvery river, South India. The samples were collected from 16 different sites along river from Stanley reservoir to estuary regions (Bay of Bengal). The microbiological scrutiny was performed during monsoon (2007), Winter, Summer and Spring (2008) seasons taken for the bacterial analysis of Total Viable

Counts (TVC), Total Coliform counts (TC), Total Streptococci counts (TS) and also four different types of pathogenic bacterial load were counts, which are indicator organisms of pollution studies. Total viable counts were found in the range of 6.2-26.0 ($\times 10^4$) mL^{-1} in monsoon, 5.2-20.0 ($\times 10^4$) mL^{-1} in summer, 4.0-17.9 ($\times 10^4$) mL^{-1} in winter and 3.3-15.5 ($\times 10^4$) mL^{-1} in spring. The TC was found in the range of 4.1-21.0 ($\times 10^3$) mL^{-1}, 3.6-17.0($\times 10^3$) mL^{-1}, 2.9-14.1 ($\times 10^3$) mL^{-1} and 2.3-12.0 ($\times 10^3$) mL^{-1}, for TS, it was 4.3-18.0 ($\times 10^2$) mL^{-1}, 3.2-13.0 ($\times 10^2$) mL^{-1}, 2.6-11.0 ($\times 10^2$) mL^{-1} and 2.0-9.6 ($\times 10^2$) mL^{-1} during monsoon, summer, winter and spring, respectively. Counts of EC, SA/SH, SF and PA were in the range of 300-3700 mL^{-1}, 20-280, 20-270 and 40-490 mL^{-1}, respectively. The Cauvery river basin has been facing severe anthropogenic activities, mostly due to religious belief, dense population, municipal sewage and industrial waste confluences etc. A huge bacterial gene pool was obtained after this study which was indicative of immense bacterial diversity in the region. *(Research Journal of Microbiology 4 (12): 540-549, 2009; **doi:** 10.3923/jm.2009.540.549)*

Studies on Acid Stress Tolerant Proteins of Cyanobacterium

C.V. Karthikeyan and G. Gopalaswamy

Cyanobacterial cultures isolated from diverse environment of acidic condition were studied for their tolerance mechanism. The identified predominant genera of *Anabaena*, *Westiellopsis* and *Nostoc* were taken for study. Among the acid tolerant cyanobacterial cultures, a protein of 15.7 kDa was identified in the cyanobacterium *Nostoc* sp. governing for acid tolerance mechanism. The N terminal sequencing of the desired protein were done to construct a suitable primer for the identification of desired gene. This may serve as a tool in engineering them onto suitable saline tolerant effective nitrogen fixers making them a good candidature for all type of soils thus directly influencing the productivity of rice. *(International Journal of Biological Chemistry 3 (1): 1-10, 2009; **doi:** 10.3923/ijbc.2009.1.10)*

Cytoprotective Activity of Chemical Constituents Isolated from *Streptomyces* sp.

T. Taechowisan, N. Chuaychot, S. Chanaphat, A. Wanbanjob and Y. Shen

Four flavonoids including kaempferol (1), isoscutellarin (2), umbelliferone (3) and cichoriin (4) have been isolated from *Streptomyces* sp. Tc052, an endophyte in the root tissue of *Alpinia galanga* Swartz. The evaluation for protective effect of compounds 1-4 against glutamate-induced cytotoxicity in hippocampal HT22 cell line was conducted. Compounds 1 and 2 showed significant effective protection

ratios of 62.4+2.8 and 55.3+3.4%, respectively, at a concentration of 100 µM, whereas compounds 3-4 were inactive. Compounds 1 and 2 showed potent scavenging effects on DPPH radical exhibiting IC_{50} value of 60.74 and 75.65 µM, respectively. These results suggest that compounds 1 and 2 may possess the neuroprotective activity against oxidative cellular injuries. *(International Journal of Biological Chemistry 3 (1): 11-17, 2009;* ***doi:*** *10.3923/ijbc.2009.11.17)*

A Novel Method for Detection of Glycoproteins on Sodium Dodecyl Sulphate Polyacrylamide Gel Using Radio-Iodinated Tyrosine

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The aim of this study is to develop a novel method for detection of glycoproteins on polyacrylamide gel. In this method, radio-iodinated-tyrosine (^{125}I-tyrosine) was conjugated to glycoprotein by schiff's base mechanism on the sodium dodecyl sulfate-polyacrylamide gel. Ovalbumin and Concanavalin A (Con A) were used as a glycosylated and a non-glycosylated model proteins, respectively. The proteins were separated in SDS-PAGE and oligosaccharides on the glycoprotein were oxidised using periodic acid to produce aldehydes than ^{125}I-tyroine was conjugated to aldehyde groups without using reducing agent like Sodium Metabisulfite. The radio-iodinated glycoprotein on gel was scanned using a Multi-Photon Detection (MPD) scanner. The electrophoretic analysis of ovalbumin and Con A were performed and stained with Coomassie brilliant blue to identify total proteins, while MPD detection of glycoproteins using ^{125}I-tyrosine selectively detected ovalbumin. Present results showed that MPD enhanced glycoprotein detection method can be used as a sensitive tool for the detection of glycoproteins on polyacrylamide gel. *(International Journal of Biological Chemistry 3 (1): 18-24, 2009;* ***doi:*** *10.3923/ijbc.2009.18.24)*

The Influence of Alcohol Kolanut Constituents on Liver Aspartate Amino Transferase and Alanine Amino Transferase Enzyme Activities on Albino Wistar Rats

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The effect of alcohol-kolanut (*Cola accuminata*) co-administration in dietary regimen was investigated in some liver function enzymes (Aspartate Amino transferase and Alanine amino transferase) using Standard Kits and Spectrophotometric methods. A 4:1 ratio of livestock feed and kolanut as well as

20% v/v alcohol in H_2O produced no significant change in the relative liver weights when compared to the control. However, the average body weight of the treated group were lowered (230.75±22.9 g) when compared to the control (370.50±25.17 g). In the liver homogenate (WH) and sub fractions (pms and cytosol), the activities of AST and ALT of the co-treated groups showed significant difference ($p \leq 0.05$) when compared with the control. The results indicate that the opposing effect of kolanut alcohol on the liver enzymes (AST, ALT) can be related to the neuronal function and liver function enzymes. The mechanism of the triggering effect of the constituents of alcohol kolanut on the activities of these two important liver enzymes is discussed through out this research. *(International Journal of Biological Chemistry 3 (1): 25-29, 2009; **doi:** 10.3923/ijbc.2009.25.29)*

The Influencing Aspects of Atorvastatin on C-Reactive Protein and Lipid Profile in Patients with Stroke

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The present study was designed to determine the effects of atorvastatin on C-Reactive Protein (CRP) and lipid profile in patients with stroke, since their anti-inflammatory properties have been investigated recently. Ninety five patients with or without stroke were recruited for the study, of which 60 belongs to control (untreated) and 35 were test group (treated) and received daily with 10 mg day^{-1} of atorvastatin. The patients were followed for over a period of 3 months. For entire study population, CRP along with lipid profile, Serum Glutamic Oxaloacetic Transaminase (SGOT) and Serum Glutamic Pyruvic Transaminase (SGPT) were measured 1st day and at the end of 3rd month of the treatment. Treatment with atorvastatin decreases both inflammatory activity and atherogenic lipoproteins. The results of this study will provide important information on how to maximize the therapeutic benefits of atorvastatin in a broader range of patients at risk for cerebrovascular morbidity and mortality. *(International Journal of Biological Chemistry 3 (1): 30-34, 2009; **doi:** 10.3923/ijbc.2009.30.34)*

Eukaryotic Release Factor 1 Affects +1 and -1 Ribosomal Frameshifting in HeLa Cells

M.A. Hossain and S.W. Peltz

The aim of this study is to identify trans-acting element(s) that alter ribosome frameshifting event(s) result a polyprotein coded by two Open Reading Frames

(ORFs) separated by a recoding signal. The hypothesis is that a depletion of eRF1 leads to inefficient recognition of stop codon or stem loop structure will force ribosomes to scan past the structure with altered efficiency resulting production of poly-protein using +1 or -1 frameshifting. The hypothesis is tested in eRF1 depleted HeLa cells using a reporter based assay system containing HIV-1 or antizyme frameshifting signaling sequence. Small interference RNA (siRNA) specific to eRF1 were transfected into cells and reporter activity was measured. The results revealed that depletion of eRF1 increased +1 frameshifting about 1.8 folds, whereas, decreased -1 frameshifting by 50%. These findings indicate that eRF1 is involved in recoding events. Particularly, alteration of -1 frameshifting in HIV-1 would be a target for drug development against the AIDS, because -1 frameshifting is the most decisive events in HIV replication that controls gag and poly-protein gag-pol ratio. (*International Journal of Biological Chemistry 3 (1): 35-41, 2009;* ***doi:*** *10.3923/ijbc.2009.35.41)*

Chemical Constituents and Hemolytic Activity of *Macrotyloma uniflorum* L.

S.M.A. Kawsar, G. Mostafa, E. Huq, N. Nahar and Y. Ozeki

The bioactivity guided separation of the dichloromethane extract of the aerial parts of *Macrotyloma uniflorum* Linn. resulted in the isolation of methyl ester of hexadecanoic and ethyl ester of hexadecanoic acid mixture (I) and n-hexadecanoic acid (II). The structures of the isolated compounds were elucidated by spectroscopic analysis, including UV, IR, ^{1}H-NMR, ^{13}C-NMR and mass spectroscopy. In addition, the fractionated crude extract of 1-butanol exhibited the significant hemolytic activity by using mouse erythrocytes. *(International Journal of Biological Chemistry 3 (1): 42-48, 2009;* ***doi:*** *10.3923/ijbc.2009.42.48)*

The Effect of Solvent Extracts of *Parimari microphylla* on Metabolites of Alloxan-Induced Diabetic Rats

A.P. Igbakin and I.G. Adanlawo

The effect of solvent extracts (ethanol and normal saline) of the seed of *Parimari microphylla* was investigated on alloxan induced diabetic rats. Rats were administered orally with 500 mg kg^{-1} body weight of the extracts over a period of five weeks. Plasma glucose, total lipids and cholesterol levels of the animals

were monitored throughout the period of the experiment. Also determined were the tissue cholesterol, total lipids, pyruvate and glycogen. The results shows high level of plasma glucose, total lipids and cholesterol i.e., hyperglycemia, hypperlipidemia and hypercholesterolemia, respectively in all the diabetic treated rats except those administered with the ethanolic extract of *P. microphylla* (EEPM) which reduced the plasma glucose, total lipids and cholesterol of the diabetic animals significantly ($p<0.05$). Also the elevated tissue pyruvate, total lipids and cholesterol in the diabetic rats were equally reduced significantly ($p<0.05$) by EEPM and their reduction is favourably compared to that of the control. EEPM increased the reduced tissue glycogen significantly ($p<0.05$). These results show that the EEPM possesses antidiabetic properties which can normalise all the alterations associated with metabolites in diabetic animals. *(International Journal of Biological Chemistry 3 (2): 49-55, 2009;* ***doi:*** *10.3923/ijbc.2009.49.55)*

Allelopathic Effects of Rice Cultivars on the Growth Parameters of Different Rice Cultivars

Saeid Ghahari and Mohammad Miransari

Eight and ten rice cultivars were tested in laboratory and greenhouse bioassay, respectively to evaluate the allelopathic effects of rice (*Oryza sativa* L.) hull extracts on the growth parameters of different rice cultivars. Extracts of rice hulls at different concentrations including 0, 5, 10 and 15% were produced and used to treat seeds of different rice cultivars. The growth parameters of germinated rice seeds after incubation for 12 days in the germinator were evaluated. In the greenhouse experiment, the treated seedlings were harvested at 21 days after planting and growth parameters determined. Treatments were combined factorially in both experiments, which were performed on the basis of a complete randomized design. Both positive and adverse effects of rice extracts on the growth of different rice cultivars in both experiments were observed. Although, the growth of genetically modified cultivars were greatly superior to the local ones, the inhibitory effects of their extracts were very much clear on the growth of different cultivars. This indicates that more modification of these cultivars, with respect to the related rice phytotoxicity potential and the response of rice cultivars to phytotoxins is necessary. Thus, there are some kinds of interactions between different rice cultivars, greatly influencing their production efficiency. *(International Journal of Biological Chemistry 3 (2): 56-70, 2009;* ***doi:*** *10.3923/ijbc.2009.56.70)*

Synthesis and Evaluation of N'-((Substituted Phenyl) Methylidene)-2-(3-Methyl-2-oxoquinoxalin-1 (2*H*)-yl) Acetohydrazide for Possible Antibacterial and Antifungal Activities

G.K. Rao, R.B. Kotnal and P.N.S. Pai

A novel synthetic methodology of Schiff's bases incorporating 3-methylquinoxalin-2(1H)-one is described. The title compounds were prepared by condensation of substituted aromatic aldehydes and 2-(3-methyl-2-oxoquinoxalin-1(2*H*)-yl) acetic acid hydrazide. Structures of all these compounds were confirmed by their spectral studies. These compounds were screened for *in vitro* antitubercular, antibacterial and antifungal activities. From the biological studies, it was possible to observe that some of the substituent on the phenyl ring of quinoxalinone hydrazones influenced the activity. Among synthesized compounds (4f, 4g, 4i and 4j), have shown good anti tubercular activity (25 μg mL^{-1}) when compared to reference drug. Compounds (4g and 4j) showed moderate to good antimicrobial activity at low concentration. The MICs (Minimum Inhibitory Concentration) against gram positive, gram negative and some species of fungi are in the range 2-4 μg mL^{-1} when compared to standard drug. In conclusion, the antimicrobial testing results revealed that the compounds possess broad spectrum of *in vitro* antimicrobial activity at low concentration. The ambient conditions, excellent product yields and easy workup procedures make this methodology a better protocol for the synthesis of newer derivatives. *(International Journal of Biological Chemistry 3 (2): 71-77, 2009;* ***doi:*** *10.3923/ijbc.2009.71.77)*

Effect of Methyl Alcohol on Conformational Structure and Thermal Behavior of Eri (*Philosamia ricini*) Silk Fibroin Film

Prasong Srihanam

This study aims to prepare silk fibroin film of Eri (*Philosamia ricini*) and investigate their conformational structure and thermal properties after treating with methyl alcohol. The Eri silk fibroin solution was obtained by dissolving the Eri cocoons with 9 M $Ca(NO_3)_2$. The silk fibroin hydrolysate was dialyzed to remove the concentrated salt against distilled water. It was then concentrated to give 2% silk fibroin by weight. Ten milliliter diluted silk fibroin to 0.5% weight per polystyrene plate was used to cast film at room temperature for 2 days. The obtained films were then treated with 80% methyl alcohol with different times. They were subjected to investigate using Fourier transform infrared spectroscopy

(FTIR) and thermogravimetric analyzer for conformational and thermal studies, respectively. The FT-IR spectra showed that Eri silk films were composed of both α-helix and β-sheet structures before exposure to alcohol and changed from low content of β-sheet structure to higher ratio when immersed in alcohol. In addition, the β-sheet structure gradually increased according to the increase of treating time used. The relative data were obtained from thermal investigation since the decomposition temperatures of Eri silk fibroin films were increased as follow by the increase of methyl alcohol treating time. It is promising that methyl alcohol can be affected to change both conformation and thermal behavior of the Eri silk fibroin film. *(International Journal of Biological Chemistry 3 (2): 78-83, 2009; **doi:** 10.3923/ijbc.2009.78.83)*

Preliminary Phytochemical Analysis, Elemental Determination and Antibacterial Screening of *Codium decorticatum*-A Marine Green Algae

J. Anbu Jeba Sunilson, R. Suraj, K. Anandarajagopal, G. Rejitha, M. Vignesh and P. Promwichit

In this study, petroleum ether, chloroform and methanol extracts of *Codium decorticatum* showed the presence of amino acids, carbohydrates, saponins, phytosterols, alkaloids and glycosides. The antibacterial activity against Gram-positive bacteria, such as *Streptococcus pneumoniae*, *Staphylococcus aureus* and Gram-negative bacteria, *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella typhi*, *Pseudomonas aeruginosa* was carried out using cup-plate method. All the extracts showed good zone of inhibition against *S. pneumoniae* and *K. pneumoniae* at the concentrations of 25, 50 and 100 mg mL^{-1} compared with standard drugs, gentamicin and ampicillin (30 μg mL^{-1}). To standardize the algae, elemental analysis was also carried out on *C. decorticatum* powder which revealed the presence of various elements. The present findings show the importance of *C. decorticatum* in producing new compounds having antibacterial activity. *(International Journal of Biological Chemistry 3 (2): 84-89, 2009; **doi:** 10.3923/ijbc.2009.84.89)*

Anti-Bacterial Activity of *Cryptolepis buchanani* Aqueous Extract

C. Sittiwet and D. Puangpronpitag

The aqueous extract of *Cryptolepis buchanani* leaves was tested against food-borne pathogen bacteria (*S. aureus* ATCC 25923, *E. coli* ATCC 25922 and *S. typhimurium* ATCC 14028), nosocomial infection bacteria

(*K. pneumoniae* ATCC 10031, *P. vulgaris* ATCC 13315 and *Ps. aeruginosa* ATCC 9721) and normal flora bacteria (*L. plantarum* ATCC 14917 and *S. epidermidis* ATCC 12228). The plant aqueous extract showed inhibitory effect against *S. aureus* ATCC 25923, *E. coli* ATCC 25922, *S. typhimurium* ATCC 14028, *K. pneumoniae* ATCC 10031, *P. vulgaris* ATCC 13315, *B. subtilis* ATCC 6633, *L. plantarum* ATCC 14917 and *S. epidermidis* ATCC 12228. The MICs (Minimal Inhibitory Concentrations) and MBCs (Minimal Bactericidal Concentrations) of this plant against all tested bacteria are in the range of 1-16 and 2-32 g L^{-1}, respectively. In conclusion, *C. buchanani* leaves aqueous extract showed broad-spectrum antimicrobial activity against food-borne pathogen +bacteria, nosocomial infection bacteria and some normal flora bacteria at low concentration. This may supported the used of *C. buchanani* aqueous extract as food-borne pathogen bacterial growth control additive and nosocomial infections treatment remedy. *(International Journal of Biological Chemistry 3 (2): 90-94, 2009;* ***doi:*** *10.3923/ijbc.2009.90.94)*

Antimicrobial Activity of *Acanthus ebracteatus* Vahl. Aqueous Extract: The Potential for Skin Infection Treatment

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The *A. ebrateutus* was extracted in boiling water with 0.7-1.3% yields. The antimicrobial activity of *A. ebrateutus* aqueous extract has been screened using agar diffusion method. *A. ebracteutus* aqueous extract showed inhibitory effect on growth of *S. aureus* ATCC 25923, *S. epidermidis* ATCC 12228, *L. plantarum* ATCC 14917, *K. pneumoniae* ATCC 10031 and *P. vulgaris* ATCC13315. The MICs and MBCs of *A. ebracteutus* has been evaluated using agar dilution and broth macro dilution methods. The MICs and MBCs of *A. ebracteutus* aqueous extract are in the range of 1-2 and 2-4 g L^{-1}, respectively. In conclusion, *A. ebracteutus* aqueous extract showed good antimicrobial activity against nosocomial pathogen and skin infection bacteria at low concentrations. This might supported the used of *A. ebracteutus* to treat nosocomial infection and skin infections. *(International Journal of Biological Chemistry 3 (2): 95-98, 2009;* ***doi:*** *10.3923/ijbc.2009.95.98)*

Antioxidant and Hepatoprotective Activity of Leaf Extract of *Justicia gendarussa* Burm

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The ancient literature reports *Justicia gendarussa* (JG) for its various uses in folk medicine including its hepatoprotective activity. The methanolic extract of air dried

leaf was prepared by soxhlet extraction method and marc remained was further extracted with double distilled water by hot percolation. Preliminary phytochemical studies were carried out and total phenolic and flavonoid contents were determined. Both the extracts were evaluated for their antioxidant activity using DPPH free radical scavenging, hydrogen peroxide scavenging and reduction of ferric ion in presence and absence of EDTA. Methanolic extract has more phenolic and flavonoid content and shows good antioxidant activity. The methanolic extract was further studied for its *in vivo* hepatoprotective activity using CCl_4 induced hepatotoxicity in albino rats. The various biochemical parameters were evaluated to asses its hepatoprotective nature. The extract showed significant hepatoprotective activity at 300 mg kg^{-1} body weight. Interestingly its hepatoprotective activity decreases as the dose increases. This study concludes that, leaf extract of JG has moderate hepatoprotective activity, which may be due to its antioxidant and free radical scavenging potential. High total phenolic content and flavonoid content are responsible for its antioxidant and hepatoprotective activity. Further studies are required to elucidate the exact phytochemical (s) and their mechanism responsible for hepatoprotective potential of JG. *(International Journal of Biological Chemistry 3 (3): 99-110, 2009;* ***doi:*** *10.3923/ijbc.2009.99.110)*

Hemorrhoid Therapy with Medicinal Plants: Astringency and Inhibition of Lipid Peroxidation as Key Factors

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Free radicals are generated in ano-rectal diseases and stress process results in pain, inflammation, swelling, itching and tenderness. The present study investigates the benefit of astringent herbs in hemorrhoid therapy. Astringent herbs used locally in the treatment of hemorrhoids [*Achyranthes aspera* Linn. (Amarantheceae), *Adansonia digitata* Linn. (Bombacaceae), *Dialium guineense* Willd (Leguminosae), *Harungana madagascariensis*, *Kigelia africana* (Lam.) Benth. (Bignoniaceae), *Newbouldia leavis* Seem. (Bignoniaceae) and *Spondias mombin* Linn. (Anacardiaceae)] were subjected to assay. Astringency was measured as the amount of tannin precipitated by a standard protein Bovine Serum Albumin (BSA) using ferric chloride blue chromophore at an absorbance maximum at 510 nm. The effects of these plant extracts on *Scomber japonicum* Houttuyn (Scombridae) lipid peroxidation was accessed by thiobarbituric acid reactivity method measured at UV 532 nm and expressed as MDA equivalent/mg of tissue. Total phenol and flavonoid contents were determined as gallic acid and rutin equivalents,

respectively. Astringency of extracts was in the order of Spondias leaves>Dialium seeds>Dialium leaves>Newboldia leaves>Kigelia fruit>Spondias fruit>Kigelia bark>Harungana bark>Newboldia bark>Harungana leaves >Adansonia leaves>Achyranthes leaves. Astringency correlated positively with total phenols ($r^2 = 0.7944$), inhibition of lipid peroxidation ($r^2 = 0.6596$ with raw homogenate and 0.9220 with cooked homogenate), low correlation with flavonoid ($r^2 = 0.059$) and no correlation between total phenol and flavonoid content ($r^2 = -0.0387$). It is proposed that these astringent herbs accomplish haemorrhoid therapy by inhibiting lipid peroxidation and plugging up minute leaks and holes in the veins and capillaries thereby promoting vein elasticity and acting as vasoconstrictors in the perianal area. *(International Journal of Biological Chemistry 3 (3): 111-118, 2009;* ***doi:*** *10.3923/ijbc.2009.111.118)*

Anti-Oxidant Activity of *Piper longum* Linn.

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Myocardial ischemia is a serious cardiac problem which should be diagnosed and treated effectively to prevent its complications. Many plant based medicine have been utilized for the effective treatment in cardiac problems. Myocardial ischemia was induced in rats by administration of isoproterenol. Petroleum ether extract of root and piperine from roots of *Piper longum* Linn. were subjected for evaluation of their anti-oxidant activity by DPPH scavenging method. Lipid peroxide and Glutathione values in myocardial ischemic rats have also been estimated by inducing myocardial ischemia by using isoproterenol. It has been found that at 50 mg mL^{-1} concentration pet ether extract and piperine exerts 74.12 and 72.13% of inhibition. Pet ether extract and piperine pretreatment decreases lipid peroxide level and maintain glutathione content to near normal in treated rats. The present study shows that the extract of the root of the plant and piperine exert anti-oxidant activity and are protective in the myocardial ischemic condition. *(International Journal of Biological Chemistry 3 (3): 119-125, 2009;* ***doi:*** *10.3923/ijbc.2009.119.125)*

Effect of Treatment Process and Preservation Method on Shelf Life of Truffles: II. Non-Conventional Methods (Radiation)

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Two varieties of local truffles namely (Zubide and Khlassi) were treated with acetic acid (pH = 3.6) and sodium oxalate (pH = 6.2) to eliminate the microbial

flora from the surface. Seven doses of gamma radiation at the rate of 0, 150, 250, 500, 1000, 2500 and 3000 Gy were used to irradiate the truffles. In both the varieties, irradiated truffles, with or without prior treatment with acid and alkali and stored in refrigerator showed significantly longer shelf life than the control (un-irradiated samples) under the same conditions. However, pretreated samples stored in refrigerator showed comparatively longer shelf life than the control (un-treated samples). In general, the shelf life of truffles was longer under refrigeration as compared to storage at room temperature. Irradiated truffles stored at room temperature showed longer shelf life than the control only when pre-treated with acid and alkali. Without pre-treatment, shelf life of irradiated and un-irradiated truffles stored at room temperature (25°C) was similar except in truffles treated with 1000 and 2500 Gy which showed significantly longer shelf lives. Overall, the shelf life of truffles can be extend by the synergistic effects of pre-treatment with acid and alkali, mild radiation, and low storage temperature. Physical and chemical analysis showed that desert truffles are good source of food components. *(International Journal of Biological Chemistry 3 (3): 126-131, 2009; **doi:** 10.3923/ijbc.2009.126.131)*

Interaction Between Ascorbic Acid and Dopamine D_2 Receptor in the Nucleus Accumbens Shell in Response to Feeding

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The aim of the present study is to evaluate the effects of intra-accumbens administration of Ascorbic Acid (AA) and co-administration of D_2 agonist, bromocriptine (Br) and the D_2 antagonist sulpiride (Su) (8, 16 μg rat^{-1}) with AA on feed intake in adult male rats. The rats (220-300 g) were divided into several groups for intra-accumbens injections: control (intact), sham AA (injected vehicle of ascorbic acid), ascorbic acid (10, 50, 250 μg rat^{-1}), sham Br (injected vehicle of bromocriptine), bromocriptine (12.5, 25, 50 μg rat^{-1}), sham Su (injected vehicle of sulpiride), sulpiride (4 and 16 μg rat^{-1}), AA (50 μg rat^{-1}) + Br (50 μg rat^{-1}) and AA (50 μg rat^{-1}) + Su (16 μg rat^{-1}). After stereotaxic operation and passing one week recovery period, drugs were injected daily (volume = 1 μL) for four days. The intra-accumbens administration of ascorbic acid (10, 50, 250 μg rat^{-1}) decreased feed intake. Intra-accumbens injection of D_2 agonist bromocriptine (25, 50 μg rat^{-1}) decreased feed intake. Co-administration of the AA (50 μg rat^{-1}) also decreased feed intake. Administration of D_2 antagonist sulpiride (8, 16 μg rat^{-1}) increased food intake and co-administration of AA (50 μg rat^{-1}) blocked this effect. These results suggest that AA can act within the Acb to decreases feed intake and it has an

agonistic effect on feeding regulatory effects of D_2 receptor. *(International Journal of Biological Chemistry 3 (4): 132-141, 2009; **doi:** 10.3923/ijbc.2009.132.141)*

Effect of Therapeutic and Toxic Doses of Ivermectin (Mectizan) on Total Serum Proteins and Hepatic Enzymes of Wistar Albino Rats

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The toxicities of the therapeutic and harmful doses of an anthelminitic drug Ivermectin (5-0-demethyl-22, 23-dihydrobenzohydrofuran) are studied in rats. Doses of 0.4 and 4.0 mg kg^{-1} b.wt. of Ivermectin (Mectizan) given at the rate of four repeated doses at daily interval within 21 days to normal rats on a stock mash protein diet produced total serum protein concentration (g L^{-1}) of 72.61±4.54 and 75.51±3.82, respectively, in comparison to 70.94±8.13 for the control thereby showing no significant effect of Ivermectin dosage on serum proteins. The corresponding test values for the activities of the serum enzymes (μ L^{-1}): alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT) and γ-glutamyltranspeptidase (GGT) were 90.22±2.96, 94.20±1.81; 32.08±3.87, 43.98±1.64 and 47.16±2.43, 56.73±4.05; 15.29±1.48, 25.88±0.77 against control values of 84.15±4.34, 28.83±1.66, 2.19±1.64 and 4.58±0.93, clearly indicating Ivermectin induced toxicity in rats which is dose dependent. The significance of the role of Ivermectin in toxicity studies and its relation to serum enzymes is discussed. Short term administration of Ivermectin (Mectizan) at therapeutic and toxic doses to albino rats did not affect total serum proteins but has marked effects on some liver function enzymes such as AST, ALT and GGP. *(International Journal of Biological Chemistry 3 (4): 142-147, 2009; **doi:** 10.3923/ijbc.2009.142.147)*

Natural Cumin Seeds for Wound Healing Activity in Albino Rats

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To screen the wound healing activity of extract and different fractions of cumin seeds on excision, incision, granuloma wound models in albino rats. The cumin seeds obtained from the plant known as Cuminum cyminum were subjected to solvent extraction with 90% ethanol, its successive fractionation by petroleum ether (40-60°) and ethyl acetate. Extract and fractions were screened for wound

healing properties on excision, incision and granuloma wound models in albino rats. The exactly weighed quantity of (250 mg kg^{-1} b.wt.) alcoholic extract and its petroleum ether fraction showed better epithelisation ($p<0.001$) as compared to control in resutured incision and granuloma wound models. It is concluded that, alcoholic extract and its petroleum ether fraction of seeds of cumin showed promoted wound healing activity on excision, incision and granuloma wound models. However, ethyl acetate fraction failed to show significant wound healing activity. *(International Journal of Biological Chemistry 3 (4): 148-152, 2009;* ***doi:*** *10.3923/ijbc.2009.148.152)*