Poster 1:

Interaction with V2 enhances the ATPase activity of C4 protein of *Cotton leaf curl Kokhran virus* – Dabawali

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Cotton leaf curl Kokhran virus – Dabawali is a mono-partite begomovirus containing single stranded DNA genome about 2.7 kb in size. The genome contains two genes in the virion sense namely V1 and V2, and four in the complementary sense viz. C1, C2, C3 and C4. The V2 and C4 genes were cloned in appropriate vectors to give the corresponding proteins hexa-histidine and GST tag respectively, overexpressed in bacteria and purified by affinity chromatography. The identity of the purified proteins was confirmed by Western blotting using anti-V2 antibodies for His-V2 and anti-GST antibodies for GST-C4. The purified GST-C4 protein was found to possess ATPase activity. On the other hand, His-V2 did not exhibit ATPase activity. Further, the His-V2 and GST-C4 proteins were shown to interact with each other by ELISA. Interaction with His-V2 increased the V_{max} for the ATPase activity of GST-C4 by 2.6 fold whereas the K_m for the same remained practically unaffected. This represents a beautiful example of two proteins interacting in a manner so as to modulate the enzymatic activity of one of the interacting partners. Both V2 and C4 have been implicated to be involved in the cell-to-cell movement of mono-partite begomoviruses. The ATPase activity of GST-C4 and its enhancement upon interaction with V2 may be important for the movement function of these proteins.

Poster 2:

Role of Earthworm Lectin in Root Nodule Formation in *Pisum sativum*

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The present study was done to study the effect of earthworm lectins- coelomic fluid lectin and vermilectin on the nodulation and growth of roots of pea plant grown hydroponically using garden soil solution which contained the already existing strains of Rhizobium. Infection with rhizobium and subsequent root nodulation is important as it provides the pea plant with essential Nitrogen. This in turn effects the growth of the pea plant and subsequent fruiting, which is agronomically important.

In this study we found that earthworm lectins initiated profuse rooting in the pea plant. Vermilectin of concentration 50µL per 200 ml of nutrient solution also induced nodulation on the fourth day of culture. This has thrown a light on the significance of vermilectins in nodulation and its future use in agriculture to obtain high quality yields of pea, since vermilectin can be isolated from a cheap source of vermicompost. Induction of rooting in *in-vitro* cultured pea plants and even in other plants is very difficult. Induction of profuse rooting by coelomic fluid lectin and vermilectin can help in induction of rooting in *in-vitro* cultured plants. However, the exact mechanism by which earthworm lectins affect rooting and nodulation were not studied. Studies at molecular level as well as biochemical level need to be done to understand the exact interaction and mechanism by which lectins influence rooting and nodulation.

Poster 3:

Delineating the Mechanism of Neutralization of Abrin Cytotoxicity by the Monoclonal Antibody D6F10

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Abrin, a plant ribosome inactivating protein (RIP) is extremely lethal and considered as a potential agent for use in biological warfare. Currently, there are no antidotes available for abrin poisoning. The only known neutralizing mAb against abrin A chain, namely, D6F10, was generated in our laboratory and was shown to rescue cells and mice from abrin intoxication. The epitope corresponding to the mAb was shown to be close to the active site of the A-chain, thus inhibiting the substrate binding. Further, the antibody was shown to get internalized in cells when bound to abrin. Therefore, studies were carried out to delineate the mechanism of intracellular neutralization of abrin by the mAb D6F10. We observed significant reduction in binding and delay in abrin internalization in the presence of the neutralizing mAb D6F10. Considering that the majority of the abrin after internalization is removed by lysosomal degradation, we studied the fate of abrin in the presence of mAb D6F10. Confocal images did not show any difference in the distribution of abrin in the lysosomes in the absence or presence of antibody. However, the antibody remained persistently co-localized with abrin even up to 8 h in the cells, suggesting that the antibody might inhibit enzymatic activity of abrin at its cellular site of action, thus rescues cells from toxicity.

Poster 4:

Exploring the Inhibitory Potential of Herbal Ligands towards the Drug Resistant Gene Products of MDR Isolates from Cauvery River, Karnataka, India

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River Cauvery, Ganga of South India, has become a dumping ground for domestic and industrial wastes which lead to the destruction of natural status and prevalence of multidrug resistant (MDR) organisms. The current study aimed to check the prevalence of such MDR isolates and screen natural inhibitors against the MDR genes by computer based virtual screening. Water samples were collected during 2011-12 from ten hotspots as reported by Karnataka State Pollution Control Board. The preliminary study showed high BOD values (5.86 mg/l), bacterial count (2.47 X 10^5 CFU/ml) and fecal coliform (>2400 MPN/100 ml) from most of the hot spots which were statistically found to be significant ($p<0.01$). This study characterized bacterial coli forms and most of them showed resistance to 48 conventional antibiotics, including the carbapenems. Prevalence of *Shigella sonnei, Escherichia coli, Pseudomonas trivialis* strain, *Enterobacter cloacae* etc. were identified by 16S rRNA sequencing. Molecular characterization of selected MDR genes from these bacteria revealed the presence of bla_{TEM} (β-lactamase resistance) and dihydrofolate reductase (*dhfr*), (trimethoprim resistance). The current study further demonstrated the utility of computer aided screening to design novel herbal leads against these virulent targets. The 3-D structures of these genes were modeled homology modeling. Further, 100 ligands were computationally screened for their drug likeliness and pharmacophoric features. The binding efficiency of best ligands and targets were studied by molecular docking. Afzelin and gallocathecin were identified as best inhibitors. Afzelin bind with bla_{TEM} and *dhfr* with energy of -7.44kcal/mol and -9.46kcal/mol respectively. Similarly, gallocatechin bind to bla_{TEM} and *dhfr* with energy of -6.36kcal/mol and -7.17kcal/mol respectively. The best docked poses were validated in comparison with experimental binding energies of the selected receptors with their normal ligands. The present study provide insights for designing novel inhibitors against many MDR gene products there by provide a therapeutic remedy for MDR pathogens.

Poster 5:

Evaluation of Immuno-diagnostic Assay for the Exposure of Filarial Infection

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There is an imperative need to develop sensitive immunoassays that can be used for diagnosis of clinical cases, primary surveillance and detection of early infection for monitoring chemotherapeutic programs to eradicate lymphatic filariasis in developing countries. Towards this, we selected two highly expressed genes of *Brugia malayi,* designated Venom allergen homologue (VAH) and Abundant Larval Transcript-2 (ALT-2) earlier identified in the third larval stage. Both antigens have been detected in sera of endemic normals and those with pre-patient infection. Therefore these antigens were proposed for the establishment of immunodiagnostics and were also compared with microfilariae (mf) specific candidate SXP-1. The recombinant proteins were expressed in *E. coli* and purified for the establishment of hybridoma. The *mf*- specific antigen capture assay was standardized and evaluated with different clinical groups. Of the 230 samples tested, VAH assay detected circulating antigen in 97.91% of bancroftian and 100% of brugian microfilaraemic (MF), comparable to the earlier reported SXP-1 assay. However, the combination capture ELISA was found to be more robust, detecting 100% of MF individuals and with higher binding values. In order to assess stage-specific infection, ALT-2 capture assay was evaluated with high, low infection endemic area clinical samples and compared with the VAH, SXP-1 capture assays. Of the 632 samples tested, ALT-2 and VAH capture assays detected circulating antigen in 57% and 52% of HIA-EN individuals, respectively. The described capture assays can be useful for the detection of early and stage-specific filarial infections in endemic regions of developing countries.

Poster 7:

Monoclonal Antibodies to Envelope Proteins of Hepatitis C Virus that Inhibit Virus Binding and Entry in Hepatocytes

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Hepatitis C virus (HCV) is the major cause of chronic viral hepatitis worldwide. HCV infection may result in liver fibrosis, leading to cirrhosis or hepatocellular carcinoma. Till date a protective vaccine is not available and the current therapy is limited to the administration of PEG-ylated interferon alone or in combination with ribavirin. The glycosylated envelope proteins (E1-E2) of HCV are considered to be important for binding to host cell receptors leading to viral entry, therefore, monoclonal antibodies (mabs) were generated to E1-E2 and tested for inhibiting virus binding to hepatocytes. To evaluate the efficacy of the mabs to block virus binding, HCV virus like particles (HCV-LP) developed using the *Baculovirus* expression system and human hepatocarcinoma cells, HuH7 were used as the model system. Three mabs (A10F2, E3D8 and H6D3) specific for the E2 region of the envelope protein were found to inhibit the HCV-LP binding to Huh7 cells, as studied by flow cytometry and by assessing infection of hepatocytes by HCV. The epitopic regions corresponding to the mabs have been delineated. The epitopes recognized by two of the antibodies were found to be highly conserved across different genotypes of HCV.

Poster 9:

Towards Establishment of Human Ectopic Liver Tissue in Mice as a Model for Hepatitis C Virus Studies

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Infection with Hepatitis C virus (HCV) is associated with inflammation of the liver leading to cirrhosis and could lead to cancer. Apart from understanding the disease condition, research on HCV is largely aimed at identifying anti-viral reagents as therapeutics. Presently there is no animal model that can be utilized to screen *in vivo* anti-HCV regents. Thus our project aims at establishing human ectopic livers in nude/ SCID mice by tissue engineering approach that can support HCV infection. In this study the PEG-Alginate-Gelatin cryogel developed for the purpose was found to support HepG2 and Huh7 cell growth and their 3D cultures showed an almost 10 fold higher cell number by the end of the 13 day study. In our endeavor to mimic the liver structure and develop a functional ectopic liver, co-cultures of HepG2, HUVEC and NIH3T3 were evaluated. The hepatocytes formed comparably large aggregates surrounded by NIH 3T3 fibroblast cells and the HUVEC cells formed capillary like structures with closely interacting and growing hepatocytes. A month long *in vivo* study was carried out, wherein the scaffold seeded with and without the Huh7 hepatoblastoma cells was transplanted in the intraperitoneal cavity of nude mice. The biochemical parameters tested showed no abnormality in the test mice suggesting that the scaffold material is biocompatible and can be used as a polymeric support for the development of the liver tissue and its transplant in the mice to fulfill the aim of the study.

Poster 10:

A Monoclonal Antibody to an Abrin Chimera Neutralizes Abrin Toxicity *In-Vivo*

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Abrin, obtained from the seeds of *Abrus precatorius* plant, is a potent toxin belonging to the family of class II ribosome-inactivating proteins. Previously, a recombinant vaccine consisting of A subunit of abrin and its homologue *Abrus precatorius* agglutinin (APA) was constructed in our laboratory and was demonstrated to protect mice from abrin lethality. Towards identifying neutralizing epitopes recognized during this response, we generated monoclonal antibodies against the proposed vaccine candidate. One such monoclonal antibody, namely A7C4 prevented abrin cytotoxicity in vitro and more importantly, neutralized the toxicity *in vivo*. Subsequently, we mapped the functional epitope of A7C4 which was found to be away from the active site. This observation was confirmed by the inability of the antibody to inhibit the enzymatic activity of the A subunit *in vitro*. To our knowledge this is the first report of an epitope on the A subunit of abrin which is not positioned close to the active site cleft and yet confers protection from abrin toxicity.

Poster 12:

Bioactive Molecules from Amphibian Skin

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Frog's skin secretions are known to present peculiar characteristics involving an arsenal of bioactive molecules. These organisms, in response to stress, injury or predator attack, release a viscous toxic secretion through granular glands containing biogenic amines, alkaloids, steroids, proteins and also peptides. Among such compounds, the antimicrobial peptides (AMPs) are responsible to play an important role in amphibian first-line defense against pathogenic microorganisms such as Gram-negative and positive bacteria, fungi and virus. In amphibians, AMPs have been isolated from different species and functionally studied, presenting not only antimicrobial but also antitumor, antifungal, anti-protozoa and spermicidal activities. However, a large number of AMPs have also shown cytotoxic activities against mammalian cells. In order to develop novel anti-infective drugs with low side effects, recent research has also been done to describe novel frog AMPs with different structural patterns. In this context, this review will focus on the antimicrobial activities of nine recently discovered amphibian AMPs including phylloseptins, nigrocins, japonicins, palustrins, parkerins, jingdongins, medusins, limnonectins and hylaranins. The biochemical properties will be discussed, as well as their possible applications in human health as new alternatives to conventional medicines.

Poster 13:

Fever and Metabolic Rate in the Toad: *Bufo marinus*

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Toads injected with the pyrogen LPS had a higher metabolic rate than control toads injected with saline if the toads were held at the febrile temperature of 32°C but not if they were held at the normothermic temperature of 25°C. We suggest that the elevated metabolic rate of febrile toads reflects the cost of the stimulation of the immune system. We discuss the evolutionary implications of these data, and suggest that the febrile response of vertebrate ectotherms and endotherms is homologous.

Poster 15:

Applications in Enzyme Technology

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In medicine, the assay of blood plasma enzyme can be extremely useful in helping to establish a diagnosis in a sick patient. In some instances, it may be possible to obtain a prenatal diagnosis. Enzyme are becoming increasingly important I the treatment of inborn errors and other diseases. Enzymes assay has played a significant role in forensic science in detecting body fluid and indicating genetic individuality, and enzyme-catalyzed reactions are involved in the key technique of DNA fingerprinting. Enzymes are widely used as reagents in clinical chemistry, forensic science and industry. Enzyme may be immobilized by being bound to an insoluble carrier (by physical adsorption, or ionic binding or covalent binding), by being entrapped in a gel or membrane, or by being cross-linked (often, but not always, in addition to being bound or entrapped. Immobilization can affect the stability, pH optimum. Immobilized enzymes are easily removed from a reaction mixture, thereby stopping the reaction and making the enzyme available to use again. They are ideal for incorporation into continuous processes. Enzymes have been used for centuries in the baking and brewing industries, as components of yeast cells and malt. More frequently, many applications have found in these and other industries for purified enzymes.

Poster 16:

MMA measurement in Vitamin B12 deficient individuals

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Prediction of vitamin B12 deficiency using total vitamin B12 measurement is difficult because several factors affect the measurement. There is a debate about the use of total vitamin B12 measurement as an indicator of deficiency for methodological and biological reasons. Holotranscobalamin (holoTC) reflects physiologically active form of vitamin B12 level and is popular for this reason. However, these measurements do not reflect tissue response to B12 deficiency. Moreover, measurement of holoTC is not available in many laboratories hence not measured routinely. This can be overcome by the measurement of tHcy(total Homocysteine), but it suffers from being non specific as it is affected by B6, fotate and B2. Vitamin B12 is a cofactor for methylmalonylCoA mutase which catalyzes the conversion of methylmalonyl CoA to succinyl CoA this reaction is specific to B12 deficiency. Advantage of using MMA as a biomarker is that its concentration is not affected by other vitamins. But measurement of MMA is difficult, requires trained people and costly chemicals and instruments like LCMS. We standardized MMA assay on LCMS and present result in B12 deficient population. We processed some samples on which we have already measured vitamin B12, holoTC and tHcy.

Subject and Method: Thirty volunteers from a know B12 deficient population participated in a B12 challenge study. We collected basal fasting blood and urine samples and after three doses x 10µg of vitamin B12 were given orally every 6hr. Vitamin B12, total homocystine, holotranscobalamin, were measured on plasma samples. MMA was measured using Agilent 6460A triple quadrupole LC-MS/MS tandem mass spectrometer under *–ve* ion mode at 1300 V. MMA was extracted from 50 µl plasma samples using 200µl acetonitrile (ACN). The ACN extract was used for estimation of MMA. The separation was done using gradient chromatography. The mobile phases used for this method consists of solvent A: 5mM Ammonium acetate + 0.1% Formic Acid and solvent B: Acetonitrile. The method was developed with 5 min run time on Obelisc C18, 4.6 x 150 mm, 5µ column. The injection volume for the sample was 5 µl. The MRM transitions were recorded for 117 to 73 and 120 to 76 for normal and D3-MMA in plasma samples.

Results: 53% of participants were vitamin B12 deficient (<150 pM), and all had low holoTC (<35pM), 38% had hyperhomocysteinemia (>15 µM). Median plasma MMA concentration is 0.67 µM (range 0.14-3.60); and 90% had MMA concentration >0.26 µM which is a

conventional cut-point used to diagnose vitamin B12 deficiency. Plasma MMA was significantly correlated with plasma B12 (rs = -0.400, p=<0.028), and to holoTC (rs=-0.457, p=0.011) but not to folate concentration. Within 24h of supplementation, basal plasma MMA were significantly related to post dose plasma MMA (p=0.000). In conclusion our result demonstrates that MMA measurements and an international cut-point of >0.26 μM diagnosed many individuals with B12 deficiency who were classified normal by total B12, holoTC and tHcy assays. This suggests the superiority of MMA measurement to diagnose B12 deficiency and allows physiological studies of B12. The MMA assay that has been standardized in our lab is presently being used to explore the association of Vitamin B12 deficiency with Diabetes.

Poster 17:

Low Metabolic Rate in Scorpions

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Scorpions are abundant in arid areas, where their population biomass may exceed that of vertebrates. Since scorpions are predators of small arthropods and feed infrequently across multi-year lifespans, a parsimonious explanation for their observed, anomalously high biomass may be a depressed metabolic rate (MR). The hypothesis that scorpion MR is significantly depressed compared with that of other arthropods, and also measured the temperature-dependence of the MR of scorpions to quantify the interaction between large seasonal variations in desert temperatures and MR and, thus, long-term metabolic expenditure. Scorpion MR increased markedly with temperature with considerable inter-individual variation. At 25 °C, the MRs of scorpions from two genera were less than 24% of those of typical terrestrial arthropods (spiders, mites, solpugids and insects) of the same mass. It is likely, therefore, that the low MR of scorpions contributes to their high biomass in arid areas. The combination of high biomass and high production efficiency associated with low MR may also favor a density-dependent 'trans-generational energy storage' strategy, whereby juveniles are harvested by cannibalistic adults that may be closely related to their juvenile prey

Poster 18:

Modulation of Octadecanoid Pathway during Induction of Systemic Resistance by Trichoderma Elicitors in Pearl Millet against Downy Mildew Pathogen

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Trichoderma sp. were isolated from root rhizosphere soil samples and screened for their potential in inducing the resistance in pearl millet against downy mildew (DM) pathogen. Totally, 91 species were collected from 10 different states in India. Species isolated from Delhi (DL-81), Maharashtra (MH-50) followed by Haryana (HR-73) and Uttar Pradesh (UP-91) significantly enhanced the seedling vigor and reduced DM disease incidence. Modulation of octadecanoid pathway by different elicitors like oligosaccharides, protein and sphingolipids from potent *Trichoderma sp*. was studied. At the biochemical level, sphingolipid extract from *T. brevicompactum* (UP-91) showed increased activity of defense-related enzymes such as lipoxygenase and allene oxide synthase along with accumulation of signaling molecules Jasmonic acid and methyl jasmonate. Under field conditions, seeds treated with *T. asperellum* (DL-81) and *T. atroviride* (HR-73) in spore suspension form and sphingolipid extract from *T. brevicompactum* (UP-91) provided significant protection against the DM disease over other treatments and this was correlated with modulation of octadecanoid pathway.

Poster 20:

Role of miRNA over protein complex forming ability in human

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Most cellular processes are carried out by proteins present in multi-protein complexes. The identification and analysis of the components of these multi-protein complexes provide insights into the understanding of the protein complexes. In higher eukaryotes, micro-RNAs (miRNAs) have emerged as an abundant class of regulatory genes that control protein expression. But till date it remains unclear how miRNAs regulate the expression of genes present in protein complexes. Our study reveals that a significant negative correlation exists between protein complexity (protein's presence in protein complex) and number of miRNA targeting per gene, confirming that proteins present in protein complexes are less likely to be regulated by miRNAs. The same trend was also observed in proteins present in single complex as well in proteins that are part of multiple complexes. Moreover, our studies also highlight the fact that this negative correlation between protein complexity and miRNA targeting is not due to the mutual exclusive relationship between protein connectivity and protein complexity or due to the influence of other factors like gene duplicability, proteins disorderness etc. Abundance of co-expressed proteins as well as shortening of 3'UTR length of proteins present in protein complex also support the avoidance of miRNA targeting in complex proteins.

Poster 21:

Pharmacokinetic Studies of Coq10 Complex in Rodents by HPLC Method

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The purpose of this investigation was to compare the bioavailability of standard powdered coenzyme (CoQ-10) with that of the CoQ10 complex. Albino Wistar rats weighing between 150-180 g were divided into two groups of 16 each and received 0.42mg/kg body weight of the standard powder and CoQ10 complex respectively. On day 0 after administration of the standard powder and CoQ10 complex, pooled blood samples were collected at different time intervals (0, 1, 2, 4, 6, 8, 24 and 36 hours) and plasma CoQ10 concentrations were determined by high performance liquid chromatography method. The same single dose of both (powder and CoQ10 complex) was administered to animals till 21 days and blood sample analysis was done on 0^{th}, 7^{th}, 14^{th} and 21^{st} day respectively for both the groups. Results showed that there was an increase in the bioavailability of CoQ10 complex (two fold) compared to the standard powder. C_{max} of the CoQ10 complex and standard powder was found to be 29.343mcg/ml and 19.06mcg/ml at 6^{th} and 4^{th} hour respectively. A steady state concentration of the CoQ10 complex was maintained from day 1 for a period of 21 days. It can therefore be concluded that bioavailability of CoQ10 complex is proved to be better than the standard powder.

Poster 22:

In-silico Enzymology

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MACiE, Mechanism, Annotation and Classification in Enzymes, is a collaborative project between the Thornton Group at the European Bioinformatics Institute and the Mitchell Group at the University of St Andrews (initially within the Unilever Centre for Molecular Informatics part of the University of Cambridge). The current version of MACiE (Version 3.0) contains 335 fully annotated enzyme reaction mechanisms, which comprise 321 EC numbers (182 EC sub-subclasses) and 372 distinct CATH codes. **HMDB**–THE HUMAN METABOLOME DATABASE is a freely available electronic database containing detailed information about small molecule metabolites found in the human body. It is intended to be used for applications in metabolomics, clinical chemistry, biomarker discovery and general education. Database is designed to contain or link three kinds of data-chemical data, clinical data and molecular biology/biochemistry data. The database contains 41,818 metabolites entries including both water-soluble and lipid soluble metabolites. It is 5,688 protein sequences are linked to these metabolite entries. Many data fields are hyperlinked to other databases and a variety of structure and pathway viewing applets. The HMDB supports extensive texts, sequence, chemical structure and relational query searches. **HCS-**a classification of hydrolases catalytic sites based on hierarchical organization. The web-accessible database provides information on the catalytic sites, protein folds, EC numbers and source organisms of the enzyme and includes software allowing for analysis and visualization of the relations between them.

Poster 23:

Comparative Study of Influence of Plant Lectins & Animal Lectins on the Lateral Branching of Roots in *Cicer arietinum*

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Lectins are carbohydrate-binding proteins, macromolecules that are highly specific for sugar moieties. Lectins have been implicated as playing an important role in mediating recognition and specificity in the Rhizobium–legume nitrogen-fixing symbiosis which is still a topic of controversy. The aim of this study is to compare the effect of partially purified plant lectins & animal lectins on the lateral branching of roots of *Cicer arietinum* grown hydroponically using garden soil solution containing the already existing strains of Rhizobium, which directly affects the nodulation. Lectins were partially purified from shoots of *Cicer arietinum* and coelomic fluid of *Eudrilus eugeniae.* It was found that both plant lectins & earthworm lectins initiated profuse rooting in the *Cicer arietinum*; however with the higher contribution of plant lectin. Induction of rooting in *in vitro* cultured legumes and even in other plants is very difficult. Induction of profuse rooting using earthworm coelomic fluid lectins and plant lectins can help in induction of rooting in *in-vitro* cultured plants. Increase in lateral branching helps in the uptake of nutrients of low diffusivity in soil; also it provides a larger surface area for the attachment of rhizobium for nitrogen fixation. Infection with Rhizobium and subsequent root nodulation is important as it provides the plant with essential Nitrogen which is recognized as the limiting factor for reduction in the yield. This in turn effects the growth of the plant, subsequent fruiting with increased nutrient content, which is agronomically important. However, the exact biochemical mechanism by which these lectins affect rooting and nodulation were not studied.

Poster 24:

Modulation of NIa-Protease Activity by Intrinsically Disordered Domain of Vpg from Pepper Vein Banding Virus: Understanding Interaction Networks of Protease

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Nuclear Inclusion protein A- protease (NIa-pro) is a protease involved in processing of *Pepper vein banding virus* (PVBV) poly-protein to generate various intermediates and mature proteins at different stages of the viral life-cycle. NIa-Pro has two domains- N-terminal viral protein genome linked (VPg) and the C-terminal protease domain (Pro). The cleavage site between VPg and Pro is sub-optimal. VPg belongs to the group of proteins that are intrinsically disordered, but attain stable structures upon interaction with other globular proteins. Such protein- protein interactions have a regulatory role on the function of the interacting partners.

In order to delineate the domain of VPg which could be involved in the modulation of Pro activity, several deletion mutants of VPg were constructed. It was observed that deletion of 22 residues from N-terminus of VPg modulates both the structure and function of Pro in *cis* and in *trans* suggesting that VPg might interact with Pro via these residues which was then confirmed by techniques like SPR.

Based on modeling and biochemical studies it was proposed that Ser 129 and Trp 143 of Pro domain might interact with VPg and relay the conformational changes to the active site catalytic triad (His 46, Asp 81 and Cys 151) leading to activation. Further it was hypothesized that other interaction networks could exist to relay the information from the VPg interaction interface to the catalytic site

In this study, two residues of the Protease domain, viz., His 142 and His 167 were identified to play a pivotal role in the interaction networks that were observed to interact with Trp 143 and Cys 151 respectively in the modelled structure of Pro. A drastic reduction in the activity of Pro was observed upon mutation of these residues. In order to delineate the interaction pathway and to understand the dynamics of the proteins, simulation studies were performed. Although, different interaction networks between W143 and Cys 151 could be traced in the mutants they were not the shortest pathway and such alterations in the network of interaction could be responsible for the loss of activity.

Poster 25:

Small Scale Large animal trials of Heat Shock Protein Inhibitors for the treatment of Trypanosomosis

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Trypanosomosis is a fatal disease caused by Trypanosoma species which affects both human and animal population and poses a major threat to the developing countries. The incidence of animal trypanosomosis is on a rise. Surra, caused by Trypanosoma evansi, has been included in priority list B of significance by World Organization of Animal Health (OIE). Control of Surra has been a challenge due to the lack of effective drugs, vaccines and drug resistance problem. These factors call for substantial efforts to develop newer therapeutic strategies against Surra. SurraMed, a Hsp90 inhibitor has shown promising results in the elimination of Trypanosoma infections in animals. This is supported by data from small scale trials in small as well as large animals performed in our lab. The small scale trials were done in mice and the large scale trials were done in horses.

Method: Horse serum collected at various time points (Time points for analysis in hrs 0.083, 0.5, 1, 1.5, 2.5, 3.5, 4.5, 5.5, 6.5, 24 hours) after administration of SurraMed. The plasma samples were deprotinated, evaporated to dryness and reconstituted in the mobile phase A. 10µL of each sample was injected into LC-QTOF. A calibration curve was plotted for increasing concentrations of SurraMed using the response of the fragment of SurraMed for quantification. Further a rate of elimination analysis was done by single ion monitoring of the fragment. Other pharmacokinetic parameters such as t1/2, Tmax, Cmax, AUC were calculated as non-compartment model using Winon-Lin 2.0.

Results: Two different concentrations of SurraMed were injected in two different horses and the rate of elimination followed the same pattern with the average t1/2 being 0.31 hours. There was a marked decrease in the parasite load in treated animals after five days of treatment with SurraMed whereas the untreated animals died by the 5th day. Also calculations indicated that repeated administration of SurraMed did not cause either accumulation or relevant modifications of the PK profile. Large scale animal trials need to be conducted to validate other relevant parameters.

Poster 26:

Functional Characterization of Interactions of *Sesbania mosaic virus* (SeMV) RdRp and P10

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Sesbania mosaic virus (SeMV) is a single-stranded RNA (4149 nucleotides) virus, belonging to the genus Sobemovirus. The viruses of this genus infect both mono and dicotyledonous plants. The viral genome is compact and encodes for 3 overlapping reading frames. The 5' and 3' proximal open reading frames (ORF) encode for movement protein (MP) and coat protein (CP) respectively. The central ORF encodes for two poly-proteins 2a and 2ab and they have a domain arrangement of Membrane anchor-Protease (pro) – viral protein genome linked (VPg) – p10 – p8 and Membrane anchor pro-VPg-RdRp respectively, the latter is translated by -1 ribosomal frame shifting mechanism. We have shown earlier that RdRp interacts with P10 by using Yeast Two Hybrid system. Hence, to determine the effect of such interaction on the activity of RdRp, RdRp and P10 were purified separately and also as RdRp P10 complex when co expressed together. It was observed that the purified RdRp P10 complex had a 10 fold higher activity when compared to the activity when the proteins added in *Trans*. Disordered domains play an important role in mediating protein– protein interactions and modulating the function of other interacting partners. The C-terminal domain of RdRp is predicted to be disordered therefore; the activity and interaction studies were carried out with the C-terminal deletion mutants of RdRp (CΔ15, CΔ29, CΔ43, CΔ65, CΔ72, and CΔ85 with and without P10). It was observed that the deletion of 43 residues from C terminal of RdRp results in almost 10 fold increase in activity when compared with activity of RdRp. The deletion of 65 and 72 residues from C terminal of RdRp results in lesser activity when compared to CΔ43, and further deletion of 85 residues leads to complete loss of activity which shows that residues between the 72 and 85 is essential for activity of protein Interestingly the interaction with P10 was reduced but not lost in these deletion mutants. To understand the oligomeric state of these proteins FPLC analysis was carried out which showed that the protein occurs in two forms i.e., dimeric and monomeric form. The results suggest that the interaction of P10 with the C-terminal disordered domain prevents aggregation of RdRp thereby increasing its activity.

Poster 28:

UPLC-ESI-MS based Metabolite Analysis of *Memecylon talbotianum* Brandis. and its Bioactive potential

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Memecylon talbotianum, hitherto unexplored endemic plant of Western Ghats was subjected to UPLC-PDA-ESI/HDMS metabolite profiling. Fourteen metabolites were tentatively identified in the methanol extract of the leaves. A broad spectrum of antibacterial activity of Gram positive pathogenic bacteria was demonstrated as evident from lysis of bacterial cells, loss of cell viability and inhibition of biofilm formation (MIC = 54 µg / mL; Gram-positive bacteria) at 24 h incubation was reported, resulting in nearly a 4 log10 CFU / mL drop in cell viability at 1.6 X MIC (Gram-positive) for this extract. The extract at two fold MIC inhibited the bacterial biofilm formation and at 8 X MIC eradicated biofilms. However, a higher concentration of this extract was required in this study for a similar effect on Gram-negative bacteria.