National Symposium on Future of Functional Genomics
(Transdisciplinary Genomics – I)

13 - 14th October 2017

Abstracts

TransDisciplinary University (TDU), 74/2, Jarakabande Kaval,
Attur Post, Via Yelahanka, Bengaluru-560064, India.

Program Schedule – 13.10.2017			
8:30-9:30	Registration		
9:30-9:45	Welcome address by Dr. Malali Gowda/ Dr. Balakrishna Pisupati		
9:45-10:00	Inauguration by Delegates		
Time	**Speakers**	**Research theme**	**Title of the talk**
10:10-10:30	**Dr. Balakrishna Pisupati**	Policy and regulations	Emerging Paradigm in policy regulation in genomics
10:30-11:00	**Dr. Mitali Mukerji**	Human/Ayurveda	Ayurgenomics for an integrative approach towards precision medicine
11:00-11:15	High tea		
11:15-11:35	**Dr. Sanjay Ghosh**	Technology/Human	Knockdown of long non-coding RNAs by CRISPR interference
11:35-11:55	**Dr. K Thangaraj**	Human population	Burden of recessive diseases in South Asia: population and genomic perspectives
11:55-12:15	**Dr. K.P. Ramesha**	Animals	Whey proteome variation at different stages of lactation in Maland Gidda (Bosindicus) - dwarf cattle of Western Ghats, India
12:15-12:35	**Dr. Upendra Nongthomba**	Insect/Fish	Functional genomics using small non mammalian model organisms, Danio and Drosophila
12:35-12:55	**Dr. Arun Kumar K.P**	Insect	Molecular basis of polyphagy and insecticide resistance in agriculture pests
12:55-1:15	**Dr. Keshava Prasad**	Technology/Plant	Functional proteometabolomics approaches to discover biomolecular networks
1:15-2:00	Lunch		
2:00-2:20	**Dr. Sheshshayee M.S**	Plants	Stable isotopes as powerful surrogates for phenotyping drought adaptive traits: Case study of developing Rice cultivar for semi-irrigated aerobic cultivation
2:20-2:50	**Dr. Dinesh A. Nagegowda**	Plants	Genomics-based exploration of specialized metabolism in medicinal and aromatic plants
2:50-3:10	**Dr. LekhaPazhamala**	Plants	Next generation sequencing based transcriptomic studies for pigeonpea improvement
3:10-3:30	**Dr. Srikrishna Subramanian**	Technology/ Microbiome	The long and the short of microbial genomics and metagenomics
3:30-3:50	**Dr. Malali Gowda**	Microbiome	Cultureomics of microbiome identified limonoids from neem endophytes
3:50-4:10	**Dr. Reety Arora**	Microbiome	Merkel cell polyomavirus – a functional genomics success story
4:10-5:00	Tea Break and Plenary Discussions		

Program Schedule – 14.10.2017		
8:00-8:30	Breakfast	
Time	**Speakers**	**Title of the talk**
8:30-9:00	Dr Malali Gowda, TDU	Introduction to Next Generation Sequencing (NGS) and Bioinformatics
9:00-9:15	Dr Abhishek Singh, Premas/Illumina	Next Generation Sequencing: Pushing The Envelope.
9:15-9:30	Ms Roopalakhsmi, Bengaluru Genomics Centre	"HLA - High resolution typing, allele calling and registry affiliation"
9:30-9:45	Mr Avid Hussain, Senior Scientist, Agilent technologies	Agilent Technologies: One stop solution for Genomics Research
9:45-10:00	Dr Anupama Gaur, ThermoFisher	Introduction to ION- TORRENT
10:10-10:15	Tea break	
Demo on sequencing		
10:15-11:00	Dr Abhishek Singh, Premas/Illumina	Demo 1- ILLUMINA
11:00-11:45	Mr Vinay, Bengaluru Genomics Centre	Demo 3- NANOPORE
11:45-12:30	Mr Avid Hussain, Senior Scientist, Agilent technologies	Demo 4- AGILENT
Bioinformatics analysis		
12:30-1:00	Dr Vivekshanbhag TDU	Using UBUNTU for Bioinformatic analysis: An Introduction
1:00-1:30	Lunch	
1:30-2:30	Mr Shivaram K R, Curl Analytics	Gene Mapping by sequencing analysis
2:30-4:00	Mr. Ananth, The Institute of Rubberwood and Dr Mahesh H B	
4:00	Valedictory	

Ayurgenomics for an integrative approach towards precision medicine

Dr. Mitali Mukerji
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The last decade, primarily propelled by the next generation sequencing technologies has revealed an unanticipated extent of variability in the human genome. This contributes to the immense phenotypic and genetic heterogeneity observed between and within populations amongst both healthy and diseased individuals. The aim of the precision medicine is to have a system's level understanding of human individuality for providing tailored solutions that takes into account the individual's variability along with the environmental and life style modifiers.

Human individuality is an outcome of coupled systems that evolves and adapts itself over time. Variability in the interactants can lead to emergence and evolution of new couplings for adaptation in spatio-temporal scales. A major challenge we face now is threading the variability across different functional hierarchies from cellular to phenome level in the context of the background population and environment.

A systems' level approach to disease and the tenets of precision medicine have been documented and practiced in Ayurveda for over 5000 years. In Ayurveda, individuals are phenotypically stratified into different constitution types "Prakriti" which remains invariant throughout lifetime and are assessed through examination of multiple system attributes. Context of the ethnic background, geography, age, temporal variations as well as heritability aspects are also factored. Prakriti of an individual is invariant throughout lifetime and can also be ascertained in diseased states. Prakriti types also differ with respect to the susceptibility to diseases as well as response to environment and therapy.

We have built an Ayurgenomics framework to integrate the phenotyping principles of Ayurveda with genomics for understanding inter-individual variability. We have set up extensive efforts through a TRISUTRA Ayurgenomics consortium wherein we have phenotypically stratified healthy individuals from genetically homogeneous populations on the basis of Prakriti and explored their molecular correlates at multiomic level. Some of the interesting insights that highlight the potential of Ayurgenomics in precision medicine will be presented.

Suggested references

1. Prasher B et al **.** Ayurgenomics for stratified medicine: TRISUTRA consortium initiative across ethnically and geographically diverse Indian populations. J Ethnopharmacol. 2016 Jul 22. pii: S0378-8741(16)30488-3.
2. Bhavana Prasher, Greg Gibson, **Mitali Mukerji** (2015) Genomic insights into Ayurvedic and Western approaches to personalized medicine Journal of Genetic J Genet. 2016 Mar;95(1):209-28
3. Aggarwal S et al (2015). Combined genetic effects of EGLN1 and VWF modulate thrombotic outcome in hypoxia revealed by Ayurgenomics approach. J Transl Med. 2015 Jun 6;13:184.
4. Tav Pritesh Sethi, Bhavana Prasher, **Mitali Mukerji** (2011) Ayurgenomics: a new way of threading molecular variability for stratified medicine. ACS Chem Biol 6: 9. 875-880 Sep
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Knockdown of long non-coding RNAs by CRISPR interference

Dr Sanjay Ghosh
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Recent transcriptome analyses have revealed numerous transcripts that originate from the non-protein-coding part of the genome in metazoa. Long noncoding RNAs (lncRNAs) constitute a subset of these transcripts and have emerged as master regulators of gene expression. Although linked to several human pathologies, it remains difficult to establish lncRNA function as current loss-of-function analysis techniques are inefficient, non-specific and/or non-programmable. Therefore, it is desirable to develop new experimental strategies that would circumvent these limitations and enable functional characterization. Using the CRISP interference (CRISPRi) system, I developed genetic tools to abolish the transcription of *Drosophila* lncRNAs in cell lines and *in vivo*. Co-expression of the catalytically inactive Cas9 (dCas9) protein and guide RNAs targeting the endogenous *roX* locus results in a robust and specific knockdown of RNAs demonstrating effective gene silencing by transcription suppression. This approach does not introduce changes in the DNA and is suitable for transcription vs. transcript analyses. The single transfection vector designed for cell-based assays in this study allows quick evaluation of the effectiveness of several guide RNAs before their use in *in vivo* studies ranging from scaling to silencing of gene expression as well as promoter and/or enhancer mapping experiments in a high-throughput manner. In combination with traditional knock down approaches, CRISPRi allows functional and mechanistic exploration of lncRNAs, thus shedding light on these enigmatic transcripts that constitute the "dark matter" of the genome.

Burden of recessive diseases in South Asia: population and genomic perspectives

K. Thangaraj

CSIR-Centre for Cellular and Molecular Biology, Hyderabad, India

South Asia is inhabited by about 5,000 anthropologically well-defined populations, many of which are endogamous communities with significant barriers to gene flow due to sociological, linguistic and cultural factors that restrict inter-population marriage. To understand the impact of endogamy, we have analysed samples from more than 2,800 individuals from over 275 distinct South Asian groups from India, Pakistan, Nepal, Sri Lanka, and Bangladesh using about 600,000 genome-wide markers. We found that 81 out of 263 unique South Asian groups, including 14 groups with estimated census sizes of over a million, have a strong founder event than the one that occurred in both Finns and Ashkenazi Jews in the West – these are founder groups known to have large numbers of recessive diseases. We identified multiple examples of recessive diseases in South Asia that are the result of such founder events. Our study provides opportunity for discovering population-specific disease causing genes in communities known to have strong founder events. Mapping of mutations that are responsible for population-specific disease would help in developing strategies for diagnosis, counseling, management and modifying the clinical course of these disorders and to reduce the disease burden among South Asians.

Whey proteome variation at different stages of lactation in Maland Gidda (*Bos indicus*) - dwarf cattle of Western Ghats, India

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Lactation stages have marked influence on milk yield and composition in cows. In this study, we carried out high-resolution mass spectrometry-based quantitative proteomics of bovine whey proteins at early, mid and late lactation stages of Malnad Gidda (*Bos indicus*) cows. A total of 564 proteins were identified, out of which, 403 proteins were found to differentially abundant at different lactation stages. As is expected of any body fluid proteome, 51% of the proteins identified in the milk were found to have signal peptides. Gene ontology analyses were carried out to categorize proteins altered across different lactation stages based on biological process and molecular function; which enabled us to correlate their significance in each lactation stage. We also investigated the potential pathways enriched in different lactation stages using bioinformatics pathway analysis tools. To the best of our knowledge, this study represents the first and largest inventory of bovine milk proteins identified to date for an Indian breed. We believe that, the current study of the temporal expression of milk proteins during lactation stages will help to enrich the existing knowledge of bovine milk during different stages of lactation in cattle.

Keywords: bRPLC, complement and coagulation, ultracentrifugation, isoelectric precipitation, TMT labeling

Functional genomics using small non mammalian model organisms, *Danio* and *Drosophila*

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Indian Institute of Science, Bangalore

The drawback in studying human genetic disorders is that one may map and study the phenotype of a genetic disease, but may not completely understand how the disease phenotype is achieved. Model organisms have been useful in addressing such genotype-phenotype correlations. How do we confirm that a SNV or SNP is just a silent change or a mutation responsible for the final disease phenotype/condition? In the first part of my talk, I will present how we have utilized *Danio rerio* (Zebrafish), a tropical, fresh water, cyprinid fish native to South-East Asia as a model system to validate and decipher molecular mechanisms of a mutation in the WDR8 gene which causes isolated microspherophakia in humans.

In the second part of my talk, I will discuss how we have used the genetically amenable organism *Drosophila melanogaster* (the humble fruit fly), to dissect the etiology of human nemaline and cardio-myopathies. Most of the structural proteins that assemble into muscle structural unit, i.e. the "sarcomere", exhibits high structural/residue conservation and performs similar functions in humans and other species. Therefore, we used a group of flight muscles called the "indirect flight muscles" as a model to trace the etiology of these human muscle diseases. The availability of multiple mutations and amenability of making transgenic animals at "will" allowed us to address the development of complex traits. Many of the Drosophila mutants, like in the human disease, may not exhibit obvious phenotypes in the heterozygote state. However, when brought in a trans-heterozygote combination with other mutants it may generate appropriate synthetic phenotypes allowing us to understand and test how certain diseases develop in a family. This approach to studying muscle diseases may not be feasible in any other model organisms.

Finally, I will discuss how these small non mammalian model organisms can be used extensively to study function genomics.

Molecular basis of polyphagy and insecticide resistance in agriculture pests

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Several agriculture pests have acquired the ability to feed on a variety of crop plants and also developed resistance to high doses of pesticides through continued genetic changes to their highly plastic genomes. This is intriguing as different plants produce different kinds of secondary metabolites that are toxic to insects but the polyphagous insects have evolved mechanisms to neutralize these metabolites and successfully feed on the plants. Though a lot of studies have been carried out to understand the molecular basis of this behavior there is no clear evidence to show the reason(s) behind this extraordinary ability of pests. Our recent work on the noctuid pest *Spodoptera litura* has provided novel insights into this long-standing enigma. Our results through RAD-seq based genetic linkage mapping suggested that because of massive expansions of certain class of genes these pests have been able to tolerate the toxins either coming from the crops they feed on or as pesticide spray. The linkage analysis resulted in a map of length 3971.7cM, with a mean distance of 0.652cM between markers. The comparative analysis of this pest with that of the domesticated silkworm *Bombyx mori* provided deeper insights into the genetic makeup of this pest that is responsible for their extraordinary abilities to survive under extremely unfavorable conditions such as high doses of pesticides. This finding paves way for a number of further studies in designing new pesticide molecules to efficiently combat the highly polyphagous and pesticide resistant serious insect pests in agriculture.

Functional proteometabolomics approaches to discover biomolecular networks

Prof. T. S. Keshava Prasad

Professor and Deputy Director, YU-IOB Center for Systems Biology and Molecular Medicine, Yenepoya Research Center, Yenepoya University, Mangalore
Faculty Scientist, Institute of Bioinformatics, Bangalore

It is reasonable to assume phenotype is often a collective expression of dynamics of networks of proteome and metabolome. Although genome represents the blueprint of most living creatures, proteins and metabolites are closer to the phenotype. Therefore, proteomic or metabolomic approaches will provide a platform for discovery of novel biomolecular networks, which can drive any specific biological conditions, including potential biomarkers and therapeutic targets for human diseases or plant kinases which may increase the flowering and yield. Recent improvements in mass spectrometry allow researchers to accomplish unbiased analysis of the entire protein complement of cells or body fluids in a single experiment. Proteomic studies to identify differentially regulated proteins between disease and normal conditions are now common. Quantitative proteomic strategies such as SILAC (stable isotope labeling with amino acids in cell culture) and iTRAQ (isobaric tags for relative and absolute quantitation) have contributed immensely to our ability to accurately determine relative protein levels. In addition to protein profiling studies, mass spectrometry is also being employed in the quantitative analysis of post-translational modifications such as phosphorylation, which can be used to dissect signaling pathways driving human diseases. I will briefly introduce mass spectrometry based proteomics and metabolomics technologies and describe our efforts to identify potential biomarkers, therapeutic targets and biological pathways involved in infection and virulence in tuberculosis and malaria using proteomic and metabolomic approaches using high-resolution mass spectrometer.

Stable isotopes as powerful surrogates for phenotyping drought adaptive traits: Case study of developing Rice cultivar for semi-irrigated aerobic cultivation

Dr. Sheshshayee M.S
University of Agricultural Sciences, Bengaluru

Among all constraints that affect crop growth and productivity, drought is the most overriding stress. While the ensuing climate change is expected to cause water scarcity in rainfed Agri ecosystems, civic and industrial demands for fresh water from the burgeoning human population is expected to reduce water resources even in irrigated systems. Thus, saving water and sustaining productivity under water limiting conditions deserve maximum research intervention, especially in India, to remain food and nutrient secure.

Significant progress in genomics and the global initiatives in drought research led to the discovery of a large number of genes and QTL governing drought tolerance and yield under stress. However, deploying these genomic resources in improving productivity has only met with limited success. Improper enumeration of drought adaptive traits and inaccurate phenotyping for capturing genetic variability in drought adaptive traits appear to contribute for the lack of success in breeding to improve drought adaptation in crop plants.

For a comprehensive improvement in drought adaptation, a trait based breeding strategy is being adopted. This necessitates development of suitable methodology for phenotyping large populations. Thus, drought research is strongly phonotype centric.

Stable isotopes form a powerful option for high throughput phenotyping for drought adoptive traits like WUE and roots. Plants discriminate against the heavy isotopes of carbon during photosynthesis and enrichment of oxygen isotopes during transpiration. A dual isotope strategy was developed and adopted to screen diverse rice germplasm to identify specific trait donor genotypes. Progeny were screened using stable isotope ratios to identify trait introgressed lines. One such line has now been released for Farmer's cultivation under semi-irrigated aerobic condition. This cultivar has a yield potential of around 6 t/ha and saves 50% of irrigation water.

Next generation sequencing based transcriptomic studies for pigeonpea improvement

Dr. Lekha T Pazhamala
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Legumes play an important role in providing food and nutrition security, in addition to improving soil quality through nitrogen fixation in the semi-arid tropics. In recent years, advancements in next-generation sequencing (NGS) have led to speedy developments in legume genomics. Various sequencing and resequencing projects had been initiated at ICRISAT for different legumes for identifying genomic regions for domestication and agronomic traits. Reference genome sequences for pigeonpea has provided useful insights into the protein coding regions, gene functions and clues to biological processes. However, this information was mainly based on the homology and de novo gene predictions. In order to correlate and complement the genome information with the gene expression that modulate agronomic traits, RNA-Seq based gene expression atlas has been developed (CcGEA). Coexpression network has been used for predicting functional roles of individual genes at a system-wide scale. Transcriptome based network predictions have been found useful for identifying candidate genes involved in agronomic traits for instance, pollen fertility and seed setting in pigeonpea. In a different study, a coherent NGS-based anther transcriptome analysis supported by physiological and cytological data have led to the identification of a thermo-sensitive male sterile (TSMS) line for a possible two-line hybrid breeding system in pigeonpea. It has been found that day temperature precisely determined the male fertility of the line with a critical threshold temperature of 25°C. Transcriptomic data deciphered a possible molecular mechanism underlying thermo-sensitive male sterility. In summary, NGS based transcriptomic approach could accelerate the on-going efforts for pigeonpea improvement.

The long and the short of microbial genomics and metagenomics

Dr. Srikrishna Subramanian
Institute of Microbial Technology, Chandigarh

It is estimated that several billion genes/proteins exist among the various living organisms on Earth. This diversity is the consequence of evolution from a small number of ancestral sequences that over a few billion years have diverged to perform a wide variety of functions. Cost-effective large-scale sequencing of DNA are helping us elucidate the genomic content and protein repertoire of life forms from diverse niches. Such high-throughput data generation techniques are capable of revealing not only the entire genomic information of individual life forms but also of all organisms present in a given niche. These technologies termed genomics and metagenomics have assumed a central role in biological research and find application in exploring the diversity of life forms that inhabit various ecological and biomedically-relevant niches. During my talk, I will cover the progress of microbial genomics and metagenomics in the last five years and talk about the various workflows we have developed and optimized for microbial genomics at CSIR-IMTECH, including the ability to obtain the complete genome of a microbe in the presence of contaminating reads. Close to 100 microbes have been sequenced, assembled, annotated and taxonomically classified using our workflow. These include several entamopathogenic fungi, probiotic bacteria and yeast, human pathogens and the soil-dwelling myxobacteria. I will conclude my talk with our exploration of the Indian gut metagenomics data and my thoughts on where the field of computational biology and genomics is heading in an increasingly data-driven scientific and technological landscape.

Cultureomics of microbiome identified limonoids from neem endophytes

Dr. Malali Gowda
TransDisciplinary University, Bengaluru.

Endophytes are the microorganisms that persist within or between tissues of plants without causing any negative effect on plant's growth and development. Endophytes are mutally coevolved with host plant species and have been reported to produce secondary metabolites similar to host plants. In this study, we developed unique neem media to isolate neem endophytic bacteria and fungi (361 fungi and 80 bacteria) from various neem explants including leaf, flower, seed, bark, cortex and root. This is the first of its kind to demonstrate culturing of endophytic bacteria and fungi on a selective neem media without using any external nutrients. Out of total 361 fungal and 80 bacterial endophytes, only 10 fungi and 3 bacteria were inhibiting the growth of Rice blast fungi, *Magnaporthe oryzae*. The two fungi namely *Fusarium sp., st.*Ai.67A and *Neocosmospora ramose* Ai.51D and bacteria *Pantoea sp*. Ai.A2 and *Bacillus sp*. Ai.C5 were further selected to check for secondary metabolite production. *Pantoea sp* Ai.A2, *Bacillus sp*. Ai.C5 and *Fusarium sp.* Ai.67A produced 3 neem metabolites as Epoxy/hydroxyl-azadiradione, Nimbin and Salanin wherein the maximum production of neem metabolites was observed in bacteria as compared to fungi. These endophytes will have great potential applications to control pests in agriculture, medicine and bio-energy.

Merkel cell polyomavirus – a functional genomics success story

Dr. Reety Arora
Prof. Sudhir Krishna Group, National Centre for Biological Sciences (NCBS), GKVK-UAS Campus, Bengaluru.

Merkel cell polyomavirus or MCV is the seventh known human tumor virus that causes a rare, lethal skin cancer occurring in specialized mechanoreceptor Merkel cells. MCV was discovered integrated in the host genome via a process called digital transcriptome sequencing. Briefly, cDNA from tumor tissue was sequenced, computationally subtracted for human sequences following which the unique viral sequence identified was extended using PCR. Since its discovery, enough evidence has justified MCV's classification as a WHO IARC Group 2A carcinogen.

Over the past few years NGS based studies have substantially contributed to our understanding of this virally caused neoplasm. Whole genome sequencing studies of MCC tumors have shown distinct mutation patterns in MCV positive (~80%) vs negative cases. The samples that looked identical to a pathologist can now be categorized into two different diseases based on UV-specific mutation marks.

Further, we used ChIP-sequencing to elucidate MCV small T antigen (sT) oncoprotein mediated gene regulation in Merkel tumor cells. We found that MCV small T antigen upregulates the MYC homolog (L-MYC), and then binds to the protein expressed recruiting it to the EP400 chromatin-remodeling complex. RNA-seq data and a genome-wide CRISPR-Cas9 screen further validated the requirement of MYCL and its interaction with sT in MCV positive MCC cells.

In my talk, I will describe how the application of genomics to a rare disease not only identified its' causative agent but continues to expand our knowledge and guide diagnosis and therapy.

Next Generation Sequencing: Pushing The Envelope.

Dr Abhishek Singh
Premas/Illumina

Next Generation Sequencing (NGS) helps us to discover more. More possibilities. More insights. And More breakthroughs. NGS make us to ask more ambitious questions and go further than we ever have before. Over the past few years, next-generation sequencing (NGS) technologies have continued to evolve, and have incorporated revolutionary innovations to tackle the complexities of genomes. These tools have provided unprecedented insights into the Basic Molecular Biology, Oncology, Microbial Genomics, Genomics In Drug development, Complex Disease Genomics, Agrigenomics, Forensic Genomics, RGH etc. With population sequencing and precision medicine initiatives, goals of sequencing tens of thousands of genomes have unearthed interesting variations. Rapid and low-cost sequencing is providing world with the tools needed to translate genomic information into actionable results.

HLA - High resolution typing, allele calling and registry affiliation

Ms Roopalakhsmi
Bengaluru Genomics Centre

Human leukocyte antigen (HLA) genes are involved in regulation of immunity. It is also associated with hundreds of different disease and extensively applied in human organ transplantation. HLA loci are clustered on chromosome 6 and their size varies from 3.5 to 20 Kb. In addition, HLA loci are highly polymorphic region in the human genome. High resolution HLA typing is crucial for successful outcome of organ transplantation. The previously established methods for HLA typing can only detect known alleles, which only cover partial HLA gene (exon 2 and 3 of class I, and exon 2 of class II) with high cost per sample and less survival rate after transplant. However high resolution HLA typing technology was not available for the registry due to technical complexities. With the advent of second and third next generation sequencing (NGS) approaches, HLA typing with full-length gene can be achieved with high resolution, high accuracy and high throughput at reduced cost per sample and it is more sensitive in detecting recessive/rare alleles. (Gowda et al. 2016). We have successfully developed a method to capture and sequencing full-length HLA gene, allele calling and software tool. For large scale HLA cataloguing, we have established HLA registry (Yenepoya-BGC registry) which will be useful to people across India and world. We encourage organizations and individuals to make use of HLA registry.

Agilent Technologies: One stop solution for Genomics Research

Dr. Avid Hussain
Agilent Technologies

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Ion Torrent: Semiconductor based Next Generation Sequencing technology-Introduction and Applications

Dr. Sreejayan Nambiar
Thermo Fisher Scientific, Bangalore

Next-generation sequencing (NGS) utilizes massively parallel sequencing to generate thousands of mega bases of sequence information per day, opening doors to new research studies that were once difficult to accomplish in a practical manner. Powered by semiconductor chips, Ion Torrent Next Generation Sequencer utilizes the first PostLight™ sequencing technology and has the power to accurately call mutations with greater confidence using Hi-Q chemistry. The instrument offers robust technology to study genes and related mutations thereby helping in understanding genetically linked disorders, expression pattern of various genes and also to study DNA protein interaction studies besides basic sequencing and gene expression applications.

Briefly on the technology front, Ion Torrent NGS sequencing systems are solid state pH measurement based technology. The sequencing reaction takes place on the semiconductor chips where there are millions of micro wells. Each well can hold only one bead (Ion Sphere Particle-ISP) where the library fragments are attached. Nucleotides used here are the natural dNTP's which are flown on to the chip one at a time. Wherever there is an incorporation of nucleotide basing on the complementarity, a hydrogen ion gets released as a byproduct of the reaction. These hydrogen ions from many such additions/reactions over the bead cause localized change in the pH of the particular micro well. This change in pH is being picked up by the sensors under each well which then converts the signal to digital form/voltage and gets recorded. As we flow the nucleotides sequentially over the chip, the signals get recorded after the flow of each type of dNTP, hence, we come to know the sequence of the target.

Ion torrent NGS platforms are by far amongst the fastest in the industry allowing users to perform multiple applications ranging from microbial whole genome sequencing, metagenomics, targeted resequencing for disease specific genes in oncology, inherited and infectious diseases using AmpliSeq technology, whole exome sequencing and whole transcriptome sequencing. With reduced hands-on time and streamlined workflow, this semi-conductor based sequencing technology makes sequencing more accessible to both translational, academic as well as to clinical research labs.

Informatics for RNA-Seq analysis

Dr. Anantharamanan Rajamani
Advanced Centre for Molecular Biology and Biotechnology,
Rubber Research Institute of India, Rubber Board, Government of India, Kottayam - India

High-throughput RNA sequencing (RNA-Seq) technology provides unique insights into the transcriptome of different organisms. Recent advances in high-throughput RNA sequencing and computational algorithms have enabled tremendous leaps forward in our understanding of transcriptome in non-model organisms by performing *de novo* transcriptome assembly. RNA-Seq has a wide variety of functional genomics applications, but the analysis protocols differ widely. Several analysis pipelines and methods have been developed to date, but no single method can be used for accurate and unbiased detection of differential gene expression in non-model organisms. In this talk I will review the state-of-the-art in RNA-Seq data analysis.

Phylogenetic Analysis of Exo-1,4-Beta Glucanase producing Actinomycetes Strains from Western Ghats of Kerala.

Lekshmi K. Edison, Shiburaj S. and Pradeep N. S. *

Microbiology Division, Jawaharlal Nehru Tropical Botanic Garden and Research Institute, Palode, Trivandrum-695 562

Actinomycetes are ubiquitous group of microorganisms, disseminated broadly in natural ecosystems and are outstanding producers of variety of enzymes and secondary metabolites. Unexplored areas of Western Ghats ecosystems facilitate the growth of highly potent microorganisms with diverse enzyme activities. Exo-1,4- β-glucanase (EC3.2.1.91) also referred to as C1 cellulase or cellobiohydrolase, is an enzyme component of cellulolytic enzyme complexes. Five morphologically different actinomycetes strains with potent Exo-1,4- β-glucanase activity were isolated from soil samples of different Western Ghats regions of Kerala. A systematic phylogenetic analysis were performed by isolating genomic DNA from the selected strains, amplification of 16s rRNA regions and finally performed the sequencing of particular 16s amplicons. A neighbour joining and maximum composite likelihood algorithm with topology tree of 16s rRNA were constructed using MEGA7. Based on the observation of comparative sequence analysis with closely related taxa, the phylogenetic diversity of these strains were belongs to genus *Streptomyces*.

Identification of Multiple Cancer Biomarkers through Comprehensive Approach: A New Era for Personalized Cancer Therapeutics

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Cancer contributes to be an important global medical and economic burden to humanity. With the expanding cancer population worldwide, whole cancer genome sequence bestows the catalogue of many cancer causing mutations whose clinical relevance has not been clearly understood. Cancer genomic studies have supported the theory of genomic aberrations and alterations as the hallmark of cancer cells. Since genomic instability due to accumulated mutations has resulted in tumor heterogeneity, metastatic progression and therapeutic resistance, we hypothesized that it is highly relevant to understand the crucial gene mutations that are driving the metastatic transformations and tumor recurrence. Based on the information of crucial gene mutations, relevant therapeutic approaches can be implemented as part of treatment regimen. To better understand the tumor genome complexities, a global approach of sequencing whole cancer genome was proposed. Though whole genome sequence provides important insights on mutation spectra, lack of relevant tools and pipelines to clinically analyze and interpret the framework of sequence data has limited the use of sequencing technology for cancer prognosis. As part of this study, we are developing a software tool- Multiple Biomarker Identification for Cancer and Genetic Disorder (MBICGD) for identifying multiple cancer specific biomarkers from whole cancer genome sequencing data using computational methods. Galaxy toolbox, which analyses the mutation spectra from cancer genomes, will be utilized for developing a relevant pipeline that encompasses the application tools for genome assembly, alignment, variation calling and annotation. The developed database will be useful for cancer diagnosis, clinical and translational research and academic studies.

Microbial diversity of cellulase and amylase producing fungal strains in Western Ghats of India

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Fungi are one of the dominant group of microbes present in soil with obvious influence on ecosystem structure. Hence there is a growing interest in assessing soil fungal biodiversity and its biological functioning in production of various significant biomolecules. With the advent of new frontiers in the field of biotechnology, the application level spectrum of cellulase and amylase has expanded in various industries, including food, fermentation, textiles, laundry, pulp, paper, agriculture as well as in research and development. The present study deals with screening of industrially important amylase and cellulase producing fungal strains. The fungal strains were collected from the Western Ghats and initially ones with cellulolytic and amylolytic enzymes production were screened. Thirty two fungal strains were isolated and among those, seven isolates where found to have both cellulolytic and amylolytic activity. Correlation of fungal enzyme production and soil organic matter, p^H, electrical conductivity and organic carbon were also discussed in details.

Ayurvedic nootropics for enhanced cognition and protection from neurodegenerative diseases.

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Optimal development and function of nervous system is vital for quality of life. Enhanced cognition and memory, adequate sensory-motor activities and absence of disease are the indications of healthy nervous system.

Advancements in the scientific research have led to a very good understanding of nervous system structure, function and physiology at cellular and molecular level. However, scientifically studied, effective healthcare solutions for cognition enhancement and management of neurodegenerative disease like Alzheimer's (AD) and Parkinson's (PD) are largely lacking[1]. Thus, we need to look at novel prevention and treatment strategies. Ayurveda has a holistic understanding of the function of the nervous system. It offers various nootropic herbs and formulations called *"Medhyarasayana"*. Ayurvedic nootropics are prescribed not only for enhanced cognition, but also as a part of treatment of AD and PD[2]. However, scientific evidence justifying the efficacy of the herbs and the underlying mechanism of action is largely lacking.

We have used *Caenorhabditis elegans* model system to study mode of action of Ayurvedic nootropics on learning-memory and neurodegeneration. Our data suggests that multiple dosage forms of Brahmi (*Bacopa monnieri*) namely juice, lipid extract and alcoholic extract can enhance short term memory and protect the worms from disease phenotypes (Ab induced paralysis and MPP+ iodide induced neuro-degeneration).

Results from such research will provide scientific evidence for use of *Medhyarasayana* in managing various clinical conditions as well as for promotion of nervous system health and also offer leads to novel nootropic products.

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Genome-wide analysis of transposons in field isolates of Magnaporthe

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Rice, wheat and millets are the most important staple cereals in India. Blast disease caused by *Magnaporthe* species is a major factor affecting productivity of these important food crops. The *Magnaporthe* species complex comprises of many phylogenetic species that cause diseases on over 50 grass species. In this study, we isolated over 100 isolates which infect rice, finger millet and grasses. We generated whole genome sequences for four rice and six non-rice isolates (finger millet, foxtail millet and grasses). This comparative study displayed high genetic variation between rice and non-rice isolates. Genome-wide comparison of transposons revealed high copy number of Pot2, MAGGY, Mg-SINE and MINE in rice isolates than finger millet and grass isolates. This is the first whole genome study on transposons in rice and non-rice *Magnaporthe* filed isolates that widen our understanding the fungal virulence spectrum.

Prevention of mastitis in cattle during dry period using an ayurveda formulation - a pilot study

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Bovine mastitis is one of the important production diseases of dairy animals which affect the economy of the farmers. A dry period of at least 40 days is needed to get optimal milk production. If it is shorter than 40 days, milk production will be reduced. This period allows the lining of the udder to be repaired and restored, so that when lactation starts again, milk production is optimal. Dry cow therapy generally involves use of intra-mammary antibiotics. The present study aimed to assess the efficacy of Ayurveda formulations in the prevention of mastitis during dry period under field conditions in comparison with regular antibiotics therapy.

The study area was the north Bangalore, KMF, India. Group I: A Standard modern veterinary drug preparation (Dry cow therapy) – Ampiclox (Ampicillin+Cloxacillin) in oil base. Group II: Ayurveda formulation and Group III: Control. The Observations included physical changes such as swelling of quarters colour. Odor and consistency of milk. The milk was tested for pH and Somatic cell count (SCC). Collection of milk samples was done on the following days: Day -0(Partial weaning), Day – 15(complete weaning), Day of calving and 5th day after calving.

The pilot study indicated the positive impact of the Ayurvedic herbal intervention in the quality of milk produced. Use of the Natural plant based formulation helped to contain the microbial infection of the udder during the crucial dry period. The data showed a comparatively better activity of the herbal formulation than the regular antibiotic dry cow therapy, in terms of restoring optimal pH and SCC of the milk during early lactation. This finding suggests an elaborate study to confirm the possibility of avoiding antibiotics in dry cow therapy to enable organic milk production.

Genome-wide Analysis of Transposable Elements in rice varieties

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Rice (Oryza sativa L.) is the most important food crop and model monocot plant species. It is the staple food for half of the World human population. Previous study sequenced the whole genome of indica cultivar, HR-12, Co-39, Tetep and Tadukan. Rice genome harbor over 30% of repeats including transposons. In the current study, we explored the transposable elements in the rice genomes and their association with genes. MITEs (Miniature inverted-repeat) are one of the transposons, which shown to associate with genes and regulate gene expression. MITEs like Tourist and Truncator have associated with Pid3/Pikh and Pi37/ Pita R-genes, respectively. This genome study will improve the understanding of indica rice germplasm.
Keywords: Rice, transposons, MITE, Tourist, Truncator

Genetic diversity assessment of *Saraca asoca (Roxb.) Willd.* population using genotyping by sequencing

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The loss of wild plant biodiversity has been increasing due to various reasons such as global climatic changes, overexploitation, pollution, habitat loss and fragmentation. *Saraca asoca (Roxb.) Willd* belonging to *Leguminosae* family, has been extensively used in almost all ayurvedic formulations for treatment of bleeding, gynaecological disorders and has many health benefits such as anti-ulcer, anti-oxytocic, anti-inflammatory, anti-oxidative, anti-bacterial and anti-cancer activities. *S. asoca* is a species of conservation concern and has been assessed as vulnerable (IUCN) and few population of this species occurs in Kolluru, in Udupi district of Karnataka, considered as MPCA (Medicinal Plant Conservation Area). We applied Genotyping By Sequencing (GBS) method to assess the genetic diversity between *S. asoca* samples collected from Kolluru and other places of southern India. Diversity relationship has been established between 50 *S. asoca* samples using the distance matrix. It was observed that *S. asoca* samples collected from specific locations clustered together. The present study reveals that there is no drastic changes in terms of genetic diversity of *Saraca asoca* collected from same locations and is specific to locations. Genetic diversity using genome-wide markers is an important study for the selection of accessions for phenotypic evaluation.

De novo Sequencing of transcriptome of *Karkatashringi* (*Pistacia integerrima*) Galls

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Pistacia integerrima belongs to family Anacardiaceae and a dioecious tree native to Asia widely distributed in East Afghanistan, Pakistan, and North West & West Himalaya. *Pistacia integerrima* is used in ayurveda for the treatment of coughs, appetite, dyspeptic vomiting, phthisis, asthma and dysentery. It has been reported for antimicrobial, antioxidant activity and toxicity studies. Recently the gall formation has been reduced due to environmental factors and also human interference. Therefore the objective of this study was to understand the molecular mechanisms of gall development (Plant-insect). *Pistacia* samples (leaf and gall) were collected from different locations at Uttarakhand. To understand the gall development, we sequenced the transcriptome of gall from *P. integerrima*. RNA was extracted and Illumina sequencing of four samples (stage 3 gall, stage 3 uninfected leaf, stage 2 gall and stage 2 uninfected leaf) yielded ~37.3, 33.1, 39.2 and 33.5 million paired-end reads (151 bp read length) for the four samples, respectively. Differentially expressed genes mapped to plants such as *Schinus molle, Morus notabilis, Lupinus angustifolius, Citrus sinensis and Glycine max.* Phylogenetic analyses and evolutionary relationships of *Pistacia integerrima* based on the chloroplast RbcL gene and RUBISCO activase gene showed that it is closely related to *Pistacia chinenis* and *Prunus persica* respectively in NCBI database .

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