# Meta genomics

## Empowering Agricultural Research through

**25 – 28 June 2019**
Faculty of Science
Kasetsart University, Bangkok

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The Gene School
empowering agricultural research

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| 08:00-08:40 | Registration |
| 08:40-08:45 | Celebration of Coronation of King Rama X |
| 08:45-09:05 | Welcome and opening ceremony<br>Group Photo |
| 09:05-09:15 | Overview of the Empowering agricultural research through (meta)genomics workshop<br>***Alexie Papanicolaou*** |
| 09:15-10:15 | **Plenary Talk:** "Genomes, Genes, Allele and Mechanisms."<br>***Roger Hellens*** |
| 10:15:10:30 | Tea break and networking (Room 352) |
| 10:30-10:50 | **Invited Talk:** High Resolution Profiling of Bacterial Communities using Full-Length 16S rRNA Sequence Data from PacBio SMRT Sequencing System<br>***Wirulda Pootakarm*** |
| 10:50-11:10 | **Invited Talk:** From Omics to System Biology: Impact on Industrial Biotechnology and Human Health<br>***Wanwipa Vongsangnak*** |
| 11:10-11:50 | **Invited talk:** Functional Genomics of Tropical Plants<br>***Goh Hoe Han*** |
| 11:50-13:00 | Lunch and Product presentation by Sponsors (Room 352)<br><br>12:00-12.20: Advancing metagenomics research with Illumina solutions - ***Cara Lim***<br><br>12:20-12:40: QIAGEN's Innovative Solutions for Advancing Microbiome Research from Challenging Samples to Insight with Confidence - ***Wasin Sakulkoo*** |

| **June 25, 2019** (Room 341) | |
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| | 12:40-12:50: Sequencing solution on DNBSEQ platform - ***Honghong Liu*** |
| 13:00:13:40 | **Invited talk:** Triggers and outcomes of genome instability in potato<br>***Kirk Amudson*** |
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| 14:00-14:40 | **Invited talk**: Molecular characterisation of four *Rattus* species using mitochondrial cytochrome oxidase I (*COI*) region<br>***Mani Challappen*** |
| 14:40-15:00 | **Talk:** The main olfactory system is possibly involved in the detection of the male effect pheromone in female goats (*Capra hircus*)<br>***Josh Elisha Octura*** |
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| 15:40-16:00 | **Talk:** Identifying Candidate Differentially Expressed Genes in Response to Lethal Heat Stress in *Ceratitis capitata* (Mediterranean Fruit Fly)<br>***Kay Anantanawat*** |
| 16:00-16:40 | **Invited talk:** Microbial community structure and dynamic response to disturbance in Singapore coastal waters<br>**Federico Lauro** |
| 16:40-17:20 | **Invited talk:** PCR-DCGE Analysis for Identification of Intestinal Bacteria in the Black Soldier Fly Larvae, *Hermetia Illucens*<br>***Nguyen Bao Quoc*** |

## WORKSHOP SCHEDULE

**June 26, 2019**

| | *For Biologists* **(Room 308)** | *For Bioinformaticians* **(Room 307)** |
|---|---|---|
| 09:00-10:00 | 16S rRNA library preparation<br>***Kay Anantanawat*** | Shotgun metagenome analysis<br>***Aaron Darling*** |
| 10:00-10:15 | Tea break and networking (Room 352) | |
| 10:15-12:15 | 16S rRNA library preparation<br>***Kay Anantanawat*** | Shotgun metagenome analysis<br>***Aaron Darling*** |
| 12:15–13:00 | Lunch and networking (Room 352) | |
| 13:00-16:00 | Microbial ecology: Theory and practice for 16S rRNA amplicon biodiversity analyses<br>***Thomas Jeffries*** | Shotgun metagenome analysis<br>***Aaron Darling*** |
| 16:00-17:00 | Participants networking (Room 352) | |

**June 27, 2019**

| | *For Biologists* **(Room 308)** | *For Bioinformaticians* **(Room 307)** |
|---|---|---|
| 09:00-10:00 | Microbial ecology: Theory and practice for 16S rRNA amplicon biodiversity analyses<br>***Thomas Jeffries*** | Transcriptome bioinformatic approaches for eukaryotic species of agricultural interest<br>***Alexie Papanicolaou*** |
| 10:00-10:15 | Tea break and networking (Room 352) | |
| 10:15-12:15 | Concept of transcriptome data analysis<br>***Passorn Wonnapinij*** | Transcriptome bioinformatic approaches for eukaryotic species of agricultural interest<br>***Alexie Papanicolaou*** |

| | June 27, 2019 | |
|---|---|---|
| | *For Biologists* ***(Room 308)*** | *For Bioinformaticians* ***(Room 307)*** |
| 12:15–13:00 | Lunch and networking (Room 352) | |
| 13:00-14:00 | Practical transcriptome data analysis<br><br>***Kay Anantanawat*** | Transcriptome bioinformatic approaches for eukaryotic species of agricultural interest<br>***Alexie Papanicolaou*** |
| 14:00-14:15 | Tea break and networking (Room 352) | |
| 14:15-17:15 | Practical transcriptome data analysis<br><br>***Kay Anantanawat*** | Transcriptome bioinformatic approaches for eukaryotic species of agricultural interest<br>***Alexie Papanicolaou*** |
| 18:00-20:00 | Dinner and networking (Room 352) | |

| **June 28, 2019** (Room 352) | |
|---|---|
| 09:00-10:00 | Experimental Design: the alpha and the omega of good science<br>***Alexie Papanicolaou*** |
| 10:00-10:15 | Tea break and networking (Room 352) |
| 10:15-12:15 | Group activity: Building communication skills and teamwork |
| 12:15–13:00 | Lunch and networking (Room 352) |
| 13:00-14:00 | Group activity: Design an experiment with your team |
| 14:00-16:00 | Group activity: Pitching your group project, asking and receiving feedback |

# OPENING SPEECH

Chongrak Wachrinrat
Acting President of Kasetsart University

It is my great pleasure to preside over the "Empowering Agricultural Research through (Meta) genomics 2019", which is held at Faculty of Science, Kasetsart University, Bangkok, Thailand during 25th – 28th of June, 2019. This is a project and activities on the auspicious occasion of the Coronation of King Rama X in this year of 2019 for promoting Science and Technology. On behalf of Kasetsart University, I would like to welcome all participants who came here to exchange experience and develop collaborative networks.

The aims of this workshop are to bring together international and national researchers as well as young scholars and to provide them an opportunity to share recent bioinformatic skills for agricultural research applications. The main program includes 1-day lecture and 3-day genomics and computational laboratory practices. There is also networking session among the participants. I truly believe that this workshop will make a valuable contribution to agricultural research in Thailand.

I would like to thank all supports from various organizations, mainly for Australian Government, Hawkesbury Institute for the Environment, Western Sydney University, Genetics Society of Thailand, the Faculty of Science, Kasetsart University, the Organizing Committee of this workshop, invited speakers, distinguished guests and all participants for supporting this congress. I am now delighted to declare the "Empowering Agricultural Research through (Meta) genomics Workshop 2019" open and I wish you all the greatest success in all your endeavors.

# WELCOME SPEECH

Apisit Songsasen
Dean of Faculty of Science
Kasetsart University

On behalf of the Faculty of Science, it is our great pleasure to welcome you to the Empowering Agricultural Research through (Meta) genomics 2019 being held at Kasetsart University, Thailand during 24th – 28th of June, 2019. This is a project and activities on the auspicious occasion of the Coronation of King Rama X in this year of 2019 for promoting Science and Technology.

This workshop is organized jointly by the Faculty of Science, Kasetsart University, the Genetics Society of Thailand and Hawkesbury Institute for the Environment, Western Sydney University, Australia. It provides both theory and bioinformatic practices for understanding agricultural systems. The workshop also aims internationally to create academic networks among overseas experts and Thai researchers.

I believe that this workshop will serve as a good base to enlighten us as to the numerous possibilities and benefits of the bioinformatic skills. I shall therefore appeal to all of you present here, to participate fully and to take maximum advantages of our experts during this four-day workshop. It is a unique opportunity for you to acquire as much knowledge and information as possible on this technology, which is definitely useful for agricultural research in the very near future.

With these words, I wish you all a very fruitful workshop.

## REPORTING SPEECH

Arinthip Thamchaipenet
Associate Dean for Research and International Affairs
Faculty of Science, Kasetsart University
Co-Chair of Organizing Committee

On behalf of the Organizing Committee, I would like to welcome all of you to the international workshop of Empowering Agricultural Research through Metagenomics 2019 at the Faculty of Science, Kasetsart University during 25th – 28th of June, 2019. This workshop is jointly organized by Faculty of Science, Kasetsart University, Genetics Society of Thailand, and Hawkesbury Institute for the Environment, Western Sydney University, Australia.

This workshop provides a good opportunity for biologists and bioinformaticians to learn practical works on wet laboratory and computational analysis using omics data, and to build collaborative network among them. Over 65 participants from various countries, including Australia, Hong Kong, India, New Zealand, Philippines, Singapore, Taiwan, Vietnam, Malaysia, USA, and Thailand are participating in this event.

I would like to take this opportunity to thank our kind sponsors, Kasetsart University, Western Sydney University, Australian Government, and private sectors to promptly support this workshop. I wish to express my sincere appreciation to our invited speakers and all members of the Organizing Committee for their kind contributions. Last but not least, I am grateful to all participants for attending and contributing to the overall success of this memorable event.

# WELCOME SPEECH

Alexie Papanicolaou
Assistant Professor, Hawkesbury Institute for the Environment
Western Sydney University, Australia
Co-Chair of Organizing Committee

On behalf of the joint organising committee of the Kasetsart and the Western Sydney universities, we welcome you to the second Asia-Pacific workshop on "Empowering Agricultural Research through (meta)genomics. We are proud to welcome you here at Bangkok and the Kasetsart University Campus. Whether you are an early, mid, or senior career researcher, you are a member of a diverse and impactful community. With the first Drosophila whole-genome shotgun-sequencing project published less than two decades ago, I believe that there is no other biological discipline - or perhaps any other science discipline - which has achieved so much in such a little time. Supported by rapid technological advances and cross-discipline interaction, genomics is at the forefront of impactful science and you are its drivers.

Our advantages of diversity and technological advances, however, present us with greater challenges as our community has grown: there is still no established way of training our next generation without asking them to devote 5-7 years of study and even then, what they learn will be made obsolete within a couple of years. Second, the fact that the vast majority of genomic resources will always be centered around biomedicine rather than agriculture limits our efforts in coordinating genomics in the agricultural domain. Third, we know that one-size-fits-all, and one-off traditional workshop training does not work for genomics: the diversity is too great, the techniques are readily made obsolete, and computational skills are lost if not maintained. Fourth, agricultural genomics is such a diverse discipline - involving laboratory assays, physiology, field work, algorithm development, data management etc. that it is counter-productive to teach highly

specialised researchers such as yourselves the basic skill sets you may only use once. Fifth, while every country faces the same challenges, each is trying to solve it independently.

This workshop started out of the need for finding a way to overcome these challenges. Can we raise awareness that we need to train for people's careers, not the project of the day? Can we ensure that the science questions are not drowned out by the fancy technique of the year (or month)? Can we foster mutually beneficial collaborations between disciplines so that science is done accurately, punctually, and effectively? Can we ensure the diversity of both the disciplines and the people is respected and supported? Can we use new online technologies and networks of countries to deliver these solutions more sustainably?

This is exactly what we are working towards this week. First, 65 of you will be exposed to new a cross-discipline model of teaching involving Next Gen wet laboratory techniques, the interpretation of computational genomic figures, the best practices for computational biologists processing genomics data, and fostering collaborations between biologists and computer scientists. But also thanks to the support of the Australian Government, we have invited experts from Australia, India, Malaysia, Philippines, USA, Singapore, Thailand, and Vietnam to help us build a long-term solution. More than 100 of us will listen to their research seminars and how they used Next Gen to deliver real innovations for agriculture. More importantly, however, we are all here to build this initial community of nations and disciplines. This workshop is to be delivered in a different country each year, but it is indeed just a pebble in the ocean. Inspired by Software Carpentry, we formed a non-profit (the Gene School) to help this pebble form ripples across the Pacific Ocean. You are invited to not only be its members but take a leadership role to deliver hands-on and online content to your home nations. I look forward to meeting each one of you and see how we can help our agricultural science community prosper.

## FUNDING SUPPORT

The workshop was organised with funding provided by the Australian Academy of Science, on behalf of the Department of Industry, Innovation and Science. The Regional Collaborations Programme is supported by the Australian Government under the National Innovation and Science Agenda.

The teaching material was created using funding provided by the Gene School LTD, the Australian Academy of Science (on behalf of the Department of Industry, Innovation and Science, the Regional Collaborations Programme is supported by the Australian Government under the National Innovation and Science Agenda), Kasetsart University, University Technology Sydney, and the Hawkesbury Institute for the Environment, Western Sydney University.

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# Advancing Microbiome Research

## From challenging samples to insight with confidence

Microbiome research encompasses sample types as diverse as the human gut, Antarctic soil, ocean water and acidic hot spring biofilms. These samples are challenging because they are difficult to lyse, with some microbes containing a tough extracellular matrix. Incomplete lysis of a microbial community results in an inaccurate representation of the microbial content of the sample. Additionally, PCR inhibitors present in these samples, especially humic acids, polysaccharides, polyphenolics, lipids and heavy metals, result in inaccurate quantification of nucleic acids and may inhibit downstream applications such as qPCR and next-generation sequencing.

Sample to Insight

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# Tech-Solution Services

## 1 RNA Sequencing

1.1 Transcriptome
- Eukaryotic Transcriptome de novo Assembly
- Eukaryotic Transcriptome Resequencing
- Prokaryotic RNA-seq

1.2 RNA-SEQ (Quantification)
- RNA-SEQ (Quantification)
- MicroRNA-mRNA Integrated Analysis

1.3 RNA-SEQ (Quantification)
- Eukaryotic Small RNA sequencing
- Eukaryotic long non-coding RNA (lncRNA) sequencing

## 2 DNA Sequencing

2.1 DNA Resequencing
- Whole Genome Resequencing (Human/Plant/Animal)
- Target Region Resequencing (Human/Plant/Animal)
- Reduced-Representation Genomics Sequencing

2.2 DE NOVO
- Genome Survey
- Plant and animal de novo sequencing
- Chloroplast and Mitochondrial Genomes Sequencing
- Fungal and bacterial de novo sequencing

2.3 Epigenomics
- Whole Genome Bisulfite Sequencing (WGBS)
- ChIP Sequencing
- Target region bisulfite Sequencing (TBS)

2.4 Genotyping
- Microarray-Based Genotyping (Non-Standard)
- SNP Genotyping by MassARRAY

## 3 Metagenomic Sequencing

3.1 Metagenomic Survey

3.2 16s/18s/ITS Amplicon Sequencing

## 4 Proteomics

4.1 Protein Identification

4.2 Quantitative Proteomics

4.3 Targeted Proteomics

4.4 Modification Proteomics

## 5 Single cell

5.1 BGI 10X Genomics High Throughput Single Cell RNA-seq (Beta) Product

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## Dr Alexie Papanicolaou

Alexie is an Assistant Professor / Senior Lecturer in Bioinformatics from the Hawkesbury Institute for the Environment (Western Sydney University); he is a computational genomicist who works (mainly) on insect genomics. He assembled and annotated multiple insect genomes (published in Nature, Molecular Ecology, Genome Biology, Nature Ecology and Evolution, and others). He led the i5k manual curation team which designed the first protocols and workflows for genome annotation and curation.

## Professor Roger Hellens

Prof Roger Hellens is a life scientist with 30 years' experience leading and conducting industry-oriented research in molecular biology, bioinformatics, genetics and cell biology. Roger is currently a Professor of Agricultural Biotechnology and the Deputy Executive Director of the Institute for Future Environments (IFE) at QUT. Since joining QUT in 2014, Roger has worked as a researcher in the Centre for Tropical Crops and Biocommodities and held several leadership positions within IFE and the Science and Engineering Faculty. From 2000 to 2014, Roger worked for the New Zealand government research institute Plant & Food

Research (formerly HortResearch), where he held several senior roles, including leading the institute's genomics research program and more recently its $10M p.a. kiwifruit breeding research program. Roger's research interests at Plant & Food Research included developing red-fleshed apple and kiwifruit varieties and exploiting Next Generation Sequencing (NGS) techniques. He also maintained a keen interest in post-transcriptional gene regulation, which has become relevant in work to understand the regulation of vitamin C. Before moving to New Zealand, Roger worked at the John Innes Centre in Norwich, UK, where he developed the first genetic maps of peas (including his PhD on the molecular basis of Mendel's white flower phenotype). He also developed the pGreen plant transformation vector and studied gene silencing (RNAi) in petunia. Roger received a PhD in Molecular Genetics from the University of East Anglia in 1995 and a Bachelor of Science from the University of Liverpool in 1989.

## Dr Wirulda (Nik) Pootakham

Dr Wirulda (Nik) Pootakham studied Plant Molecular Biology at Cornell University and graduated summa cum laude in 2002. She went on to pursue a PhD in Cell and Molecular Biology at Stanford University, studying a signal transduction pathway that regulates nutrient starvation responses in Chlamydomonas, a single-cell green alga. After obtaining a doctorate degree, she joined

the Genomic Research Lab at the National Center for Genetic Engineering and Biotechnology (BIOTEC) and is now the Head of the Genomics Research Lab. Her work focuses on the identification of molecular markers linked to important agronomic traits for plant crop. Nik also has been involved in projects that address environmental issues of coral bleaching in the Gulf of Thailand and Andaman Sea. She received a Young Technologist Award from the Foundation for the Promotion of Science and Technology under the Patronage of His Majesty the King in 2017, the UNESCO-L'Oréal For Women in Science Fellowship in 2018, and was elected to serve as a Young Affiliate for The World Academy of Science (TWAS) from 2017 to 2022.

## Associate Professor Wanwipa Vongsangnak

Associate Prof Dr Wanwipa Vongsangnak completed her PhD at Chalmers University of Technology, Sweden in 2009 before joining the Soochow University as an associate professor at the Center for Systems Biology in 2011. Presently, she is an associate professor at Department of Zoology, Faculty of Science, Kasetsart University, leading the Bioinformatics and Systems Biology Unit. Her research focuses on the development of new bioinformatics and systems biology approaches to study the use of microorganisms in industrial biotechnology.

## Associate Professor Goh Hoe Han

Hoe-Han Goh, a plant molecular biologist, graduated from the University of Sheffield, UK. He started his first academic position at the Institute of Systems Biology, Universiti Kebangsaan (National University) Malaysia in Nov 2011. He was further trained on NGS applications through workshops, such as EMBO course at BGI-Shenzhen and SeqAhead@TGAC-UK. Since then, he established a Plant Functional Genomics Research Group which focuses on crop improvement and molecular exploration of tropical plant species using functional genomic approaches. In Jun 2014, he was appointed as the Head of Plant Biotechnology Centre before becoming the Head of Bioinformatics Research Centre in 2016 and promoted as an Associate Professor in Jan 2019. Since 2012, he has successfully mentored a total of 11 MSc and 6 PhD graduates with 47 peer-reviewed articles. He has also conducted various seminars and workshops on qPCR, transcriptomics, and proteomics analysis.

## Kirk Amundson

Kirk is a fourth-year PhD student in the lab of Prof. Luca Comai at the University of California, Davis. Using potato as a model system, he studies two processes that trigger genome instability in plants: first, uniparental genome elimination, a process by which sexual progeny inherit the genome of only

one parent, and second, regeneration of whole plants from single cells. Although both techniques are routine in plant biology, the underlying mechanism leading to genome destabilization in both contexts remains elusive. He uses a combination of genomic and cytogenetic analyses to document the incidence and types of chromosomal restructuring events that occur in either process. By understanding how chromosomes break, his goal is to provide a framework for either preserving or engineering plant genomes.

## Professor Luca Comai

Dr Luca Comai is Professor of Plant Biology at the Genome Center of the University of California at Davis. He has B.S. equivalent from the Universita' di Bologna, Italy, and a PhD in plant pathology from UC Davis. In his career, he has worked on bacterial plasmid genetics, plant biotechnology (glyphosate resistance via alteration of EPSP synthase), and genetics and genomics of polyploidy. He co-developed TILLING, a method to identify targeted mutations. Since joining UC Davis in 2006, he has focused on function and regulation of chromosomes in polyploid genomes and on stress-induced genome instability. Dr Comai teaches the foundation genetics course at UCD using a flipped approach. He has authored over 130 publications, has an H impact factor of 72, is a Fellow of the American Association for the Advancement

of Science. In 2017 he received the ASPB Innovation Prize for Agricultural Technology.

## Professor Mani Chellappan

Professor Mani Chellappan is the head of the Entomology department at the Kerala Agricultural University. As an insect physiologist, he works on understanding the effects of nutrition and toxicity of a number of insects, in particular bees. His recent research involves understanding how the gut microbiome of invasive insects and higher vertebrates contributes to their species range and ecological adaptation.

## Dr Jorge Gil Angeles

Dr Jorge Gil Angeles completed his PhD in Molecular Biology with a Minor in Bioinformatics at New Mexico State University, USA. He then worked as a post-doctoral researcher at the University of California San Francisco (UCSF) and the Northern California Institute for Research and the Education/Veterans Health Research Institute (NCIRE). Recently, he joined the Philippine Genome Center the University of Los Banos, Philippines as the assistant leader of the functional Epigenetics and Phenomics group. His research interests include promoter research, molecular biology, epigenetics and codon usage analysis. His instruction endeavors are on the graduate-level courses

on biocomputing and epigenetics. He is also a member of the graduate advisory committee of several MS students pursuing the fields of Molecular Biology and Biotechnology, Statistics and Information Technology. Dr Angeles was a visiting fellow at the Environmental Epigenetics Laboratory, Hawkesbury Institute for the Environment, Western Sydney University - Hawkesbury in 2018.

**Dr Kay Anantanawat**

Kay is the Next Generation Sequencing facility manager for the University of Technology Sydney. After a career as a molecular entomologist, Kay is working with Aaron on creating new protocols for biodiversity assessment and running the NGS facility. Since her PhD, Kay has been extensively involved in creating and analysing NGS data. Her expertise includes creating Next Generation Sequencing libraries (RNA, DNA, and amplicon) using both commercial kits and custom methods she develops. She has hands-on experience working with sequencing instruments including 454 Roche sequencing, Illumina MiSeq and HiSeq2500.

## Professor Federico Lauro

Professor Federico Lauro graduated from University of Padua and went on to obtain his PhD at Scripps Institute of Oceanography (SIO) at the University of California in San Diego. His driving interest is to identify how microorganisms evolve and adapt and how their functions can drive the ecological processes that are critical for sustaining the health of marine environments. His research covers both the experimental and the computational sciences – in particular, deep-sea microbiology, and the latest "omic" technologies that are essential for deriving a clear and thorough understanding of microbial communities and ecosystem function.

## Dr Nguyen Bao Quoc

Dr Nguyen Bao Quoc is a lecturer at the Research Institute for Biotechnology and Environment, Nong Lam University (RIBE-NLU), Ho Chi Minh City, Vietnam. He leads molecular microbiology and pathogenesis group in RIBE-NLU and is the head of Human Pathogenesis Lab. His main research interests are microbial pathogenesis and molecular diagnostics of microbial pathogens using innovative approaches such as LAMP, RPA, real-time and PCR. He has published more than 38 papers in peer-reviewed international and national journals, book chapters and

patents. He has successfully supervised over 20 Master and is mentoring 3 PhD candidates. He is the PI/co-PI of many national and international projects (JSPS, ICGEB, LAL, EP-NUFFIC, UNU-IAS, MOST, MOET, FIRST, eASIA, NLU). He is also organizer/co-organizer of many national and international training courses/conferences in Vietnam.

**Professor Aaron Darling**

Aaron is a distinguished academic known for the development of cutting edge computational and molecular techniques for the study of microbial genomes.

He is a Professor of Computational Genomics and Bioinformatics at the ithree institute, University of Technology Sydney, with an interest in computational metagenomics. At UTS he leads a team of computational scientists and molecular biologists. His research is widely cited and creates real-world impact; his software packages for genome assembly and comparison (A5-miseq, Mauve) are used by thousands of academic researchers as well as a range of private industries, public health (Centers for Disease Control), government and environmental organisations. His academic publications have been highly cited, with over 20000 citations to date and an H-index of 38. Aaron currently holds the title of President of the Australian Bioinformatics and Computational Biology Society (ABACBS). He is on the editorial board of PLOS

Computational Biology and formerly on the editorial board of BMC Bioinformatics.
In addition to his academic work, Aaron is the co-founder and CSO of startup Longas Technologies Pty Ltd. Longas has developed the Morphoseq virtual long read technology. Morphoseq enables short read sequencers to generate high accuracy 8-10kbp reads at low cost.
Aaron holds a PhD in Computational Biology from the University of Wisconsin, Madison, US

**Dr Krithika Arumugam**

Dr.Arumugam is a Bioinformatician at the Singapore Centre for Environmental Life Sciences Engineering (SCELSE), NTU, Singapore. With interests in metagenomics and distributed computing, she designs pipelines and analyses high throughput next generation sequencing data in high-performance computing environments. Her primary research focuses on genome recovery from metagenome assembled genomes.

**Dr Thomas Jeffries**

Dr Tom Jeffries is a Lecturer in Microbiology at Western Sydney University and an emerging expert in the application of computational biology and ecology tools to microbial genomics datasets. His work focuses on determining the ecological drivers of microbial diversity in a variety of habitats spanning the ocean, dryland soils and the human body. His research has

produced 47 peer-reviewed publications and has been cited 954 times. He is the Director of Joint Academic Microbiology Seminars, a committee member of the Australian Society for Microbiology and a Director of The Gene School.

## Dr Passorn Wonnapinij

Dr Wonnapinij is a computational biologist in the faculty of Sciences. Her work focuses on mitochondrial genetics: new models of mtDNA heteroplasmy inheritance, the evolution of social amoebas, fireflies, and studying the evolution of pig through bones found in archeological sites.

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# Invited Talks and Talks

PLENARY TALK

# Genomes, genes, allele and mechanisms

Roger P. Hellens
Institute for Future Environments, QUT, Brisbane, Australia

The ability to assemble genomes has benefited greatly from technological advances in DNA sequencing. Genome annotation continues to improve as more genomes are sequenced and gene models are more clearly defined. Functional characterisation remains one of the more time-consuming steps in genome analysis but, as genome resequencing become more commonplace, the subtle sequence difference in the alleles of gene provide clues to the underlying molecular mechanism that leads to phenotypic diversity. In our work, we have sequenced the genomes of strawberry, apple, kiwifruit and banana and the Australian native Nicotiana benthamiana. Where genome sequence is not available, we have used synterney between crops with genome sequence. We have used this information to annotate and characterise genes involved in the biosynthesis and regulation of vitamin C and anthocyanins (the red, purple and blue pigment commonly found in the flower and fruit of plants). We have then explored the allelic diversity that exists for these genes in germplasm, breeding and mapping populations, and use these sequence difference to understand how the DNA changes have resulted in phenotypes such as the red and white flower character use by Mendel to discover inheritance, red-flesh apple and high vitamin C kiwifruit. Studying these alleles has given insight into the molecular mechanisms that allow is to better understand the biology, physiology and biochemistry of these phenotypes. This information can be used by researches to deepen the fundamental understanding of biological processes. We are also able to help breeders to better understand the parents and parental combinations that are needed to create new cultivars that are more

productive, resilient to environmental change or more nutritious to eat.

INVITED TALK

# High resolution profiling of bacterial communities using full-length 16S rRNA sequence data from PacBio SMRT sequencing system

Wirulda Pootakham
National Center for Genetic Engineering and Biotechnology (BIOTEC), National Science and Technology Development Agency (NSTDA), Pathum Thani, Thailand

Over the past decade, the use of Sanger sequencing in bacterial community surveys has gradually been replaced by various next generation sequencing platforms due to their economy of scale and magnitude orders higher sequencing throughput. The majority of microbial profiling studies utilizes the short-read amplicons instead of the full-length 16S rRNA sequences in environmental community surveys. The advance in throughput has, however, come at the cost of read length, and this tradeoff has inevitably resulted in less accurate classification of partial 16S sequences, especially at the genus or species level. Recently, Pacific Biosciences (PacBio) has developed a single molecule real time (SMRT) DNA sequencing system that is able to generate raw reads with an average length of longer than 10 kb. With the high-throughput PacBio CCS technology, the cost of obtaining full-length 16S sequence data is considerably lower than that of the gold standard Sanger approach, making PacBio an attractive sequencing system for microbial diversity studies. The use of barcodes enables multiplexing of different samples into a single SMRTbell library, further reducing the overall sequencing cost. We took advantage of the PacBio circular consensus sequencing (CCS) technology to investigate the diversity and structure of coral-associated microbiome and demonstrated the superior performance of full-length 16S rRNA sequences in

resolving taxonomic uncertainty of coral associates at the species level.

INVITED TALK

# From Omics to Systems Biology: Impact on Industrial Biotechnology and Human Health

Wanwipa Vongsangnak
Department of Zoology, Faculty of Science, Kasetsart University, Thailand

Omics technologies over the past decade have resulted in a wealth of big data describing the functions, expressions and interactions of different molecules within the cell. Rapid development of bioinformatics and systems biology, there are several studies on the behavior of the complete biological systems. Genome-scale metabolic models (GEMs) have become a popular tool for systems biology. This has allowed to predict whole-cell effects of environmental or genetic changes, as well as to perform integrated data analysis. In this talk, the GEMs of industrial microbes and their applications for industrial biotechnology are introduced. For impact on human health, metagenomics is further presented to investigate role of microbiome in gut health and diseases. In summary, omics and systems biology can be a key driver for addressing the grand challenges in context of sustainable production of required materials and ensuring a better health for long life.

INVITED TALK

# Functional Genomics of Tropical Plants

Hoe-Han Goh
Plant Functional Genomics Research Group, Institute of Systems Biology, Universiti Kebangsaan Malaysia

Malaysia is rich in biodiversity with many tropical plants remain to be explored for human wellbeing. The advent of sequencing and mass spectrometry technology allowed unprecedented opportunity for transcriptome-wide study and biomolecular discovery. For the past seven years, we have been applying omics approach on the study of various plants, including rice, papaya, mangosteen, *Arabidopsis*, *Nepenthes*, *Rafflesia cantleyi*, *Persicaria minor*, and *Mitragyna speciosa*. In this talk, I will be using examples from some of these plants to describe how we utilised omics approaches to understand the molecular regulation of plant physiology, such as seed germination, flower development, and biotic stress response from elicitation. Transcriptome-wide effects of gene perturbation were also studied for fundamental understanding and biotechnological applications. Transcriptomic profiling has been useful for elucidating biosynthesis pathways of beneficial compounds of interest, such as phenylpropanoids, flavonoids, and alkaloids. Lastly, a systems approach was also taken to understand plant carnivory and hybridisation on molecular expression of tropical pitcher plants through comparative omics approach.

INVITED TALK

# Triggers and outcomes of genome instability in potato

**Kirk Amundson[1,2],** Benny Ordonez[1,2], Michelle Fossi[1,2,3], Monica Santayana[4], Sundaram Kuppu[1], Ek Han Tan[5], Isabelle Henry[1,2], Awais Khan[4,6], Merideth Bonierbale[4], Anne Britt[1], and Luca Comai[1,2]
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Maintaining the right balance between genome integrity and flexibility is essential for survival of organisms and their offspring. Various endogenous processes can precipitate genome instability, an outcome whose nature and consequences are not clearly understood. Due to flexible development patterns, lack of a predetermined germline, and frequent polyploidy, plants are generally tolerant to genome instability, and may therefore be more likely to exhibit and retain novel karyotypes than other systems. Using the model plant *Arabidopsis thaliana* and cultivated potato (*Solanum tuberosum* L.), we are investigating genome instability resulting from various stimuli. In Arabidopsis, we have shown that various modifications to centromeric histone H3 (CENH3), the major epigenetic determinant of centromere function, lead to postzygotic centromere malfunction and subsequent missegregation, restructuring and/or loss of chromosomes derived from the parent encoding modified CENH3. In extreme cases, the entire chromosome set of one parent, the haploid inducer, is lost, which yields haploid plants. Similarly, in potato, certain intraspecific crosses are known to yield uniparental dihaploids. We have resequenced the genomes of over 1000 putative potato dihaploids, and report the incidence and type of progeny with partially eliminated haploid inducer genomes. Plant regeneration is another well-documented source of genome instability under the umbrella term "somaclonal variation", though the causes remain uncertain. We have observed frequent and pervasive rearrangements in both "off type" and plants indistinguishable from wild-type. This instability affects one to several chromosomes and can persist in regenerated plants. To unravel underlying mechanisms, we are focusing on specific events that appear in

independent regenerated lines. Our results suggest underlying similarities between these processes and provide a framework for understanding and engineering plant genomes.

TALK

# Corals host unique and abundant microbial communities

Lindsey Deignan
The Singapore Centre for Environmental Life Sciences Engineering (SCELSE),
Nanyang Technological University

Corals host unique and abundant microbial communities. This microbial community, or microbiome, plays an essential role coral holobiont functioning, including nutrient cycling (particularly carbon, nitrogen, phosphorus and sulfur), protection via the production of antibiotics, and maintaining homeostasis. My research investigates the potential for the microbiome to help corals become more resilient to environmental stressors. I examined the response of the coral microbiome to targeted stressors, particularly interaction with algae. Coral microbiomes responded differently to algal interactions depending on the specific reef within Singapore from which the coral was collected, underpinning the variable and complex nature of the coral microbiomes in response to their environments. We are now monitoring the variability and stability of coral microbiomes across Singapore and correlating it with environmental parameters. As coral reefs face increasing environmental impacts, particularly that of climate change, it is becoming increasingly valuable to understand the extent to which the microbiome provides an adaptive resilience to the coral against environmental impacts.

INVITED TALK

# Molecular characterization of four Rattus species using mitochondrial cytochrome oxidase I (COI) region

Mani Challappen
Kerala Agricultural University, Kerala, India

Identification of rodents are very difficult due to their highest similarity and complex taxonomy. It requires specialized skills to identify. The result generated by the morphological observation is not always easy. In this study we used DNA barcoding as a successful tool for the identification of four Rattus species such as R. *ranjiniae, R. rattus, R. norvegicus,* and *R. wroughtoni* at molecular level. DNA barcoding by sequencing of a short fragment of the mitochondrial cytochrome oxidase subunit (COI) region has been standardized for these species. The total genomic DNA has been isolated from the tail of the species using DNA extraction kit. The mtCOI region got amplified by polymerase chain reaction from genomic DNA using universal barcode primers and carried out the DNA sequencing. The sequences generated from the study were analyzed for sequence homology using nucleotide BLAST, NCBI. The specimen details and sequences generated as the result of the study were submitted to BOLD database. A barcode was generated for the species *R. ranjiniae, R. rattus, R. norvegicus,* and *R. wroughtoni.* The limitations in the inherent morphology based identification system of rodents can be improved by using this novel microgenomic method.

TALK

# The main olfactory system is possibly involved in the detection of the male effect pheromone in female goats (Capra hircus)

**Josh Elisha R. Octura**[1,2,3], Keiichiro Maeda[1] and Yoshihiro Wakabayashi[2]

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Most mammals have two physiologically distinct olfactory systems namely, the main olfactory system (MOS) and the vomeronasal system (VNS), which function to detect odorants and pheromones, respectively. The neural activity of the gonadotropin-releasing hormone (GnRH) pulse generator, which can be detected as multiple unit activity (MUA), accompanied by pulsatile GnRH / LH secretion in female goats can be stimulated by the male effect pheromone. The detection site of this pheromone, however, remains unclear. In this study, the possible involvement of the MOS in the detection of the male effect pheromone was investigated. Ovariectomized goats implanted with electrodes in the hypothalamic arcuate nucleus for recording the MUA were used. To know if the MOS can detect the pheromone, the MUA of goats were monitored upon exposure to the male goat hair (pheromone source) before and after vomeronasal organ (VNO) occlusion. Furthermore, in situ hybridization (ISH) for vomeronasal type-1 receptors (V1Rs) and GÎ±i2, a V1R-associated G protein and GÎ±i2 immunohistochemistry (IHC) were carried out to examine the presence of VNS-related genes in the MOS. The results of this study showed that the VNO occlusion did not prevent the male hair-induced activation of the MUA in female goats. Moreover, the ISH for V1Rs revealed that a small subset of OSNs expressed V1Rs in the olfactory epithelium. Some of the V1Rs

co-expressed GÎ±i2 suggesting that a functional V1R-GÎ±i2 signal transduction mechanism is employed by a small subset of OSNs in the olfactory epithelium. GÎ±i2 IHC revealed some positive axon terminals in the olfactory bulb indicating that the GÎ±i2-expressing OSNs may functionally project their axons to the olfactory bulb. Taken together, the findings of this study suggest that the MOS might be involved in detecting the male effect pheromone in female goats.

INVITED TALK

# Molecular and Bioinformatic Characterization of Promoters Due to Variations in Sequences, Motifs and Methylation States

Jorge Gil C. Angeles
Philippine Genome Center – Program for Agriculture, Livestock, Forestry and Fisheries, Office of the Vice-Chancellor for Research and Extension, University of the Philippines Los Baños, Los Baños, Laguna, Philippines

Promoters are upstream elements of a gene that control its expression. Promoter efficacy and strength is assessed qualitatively or quantitatively. Promoter activity is influenced by sequence differences, motif variation or via altered epigenetic or methylation states that are investigated through molecular or bioinformatic methods. Molecular and phenotypic examples will be presented.

TALK

# Identifying candidate differentially expressed genes in response to lethal heat stress in Ceratitis capitata (Mediterranean fruit fly)

**Kay Anantanawat**[1], Kelly Hill[2], Alexie Papanicolaou[3], Wei Xu[1]
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Lethal stresses such as heat, cold and irradiation have been used in postharvest treatments to control Tephritid fruit flies. However, there is a gap in understanding the cellular biological pathway in response to these treatments in fruit flies. This study investigates the molecular basis of stress pathways in two major invasive tephritids: *Bactrocera tryoni* (Queensland fruit fly) and *Ceratitis capitata* (Mediterranean fruit fly). The stresses in focus during the study are heat, cold and irradiation. The aim is to generate a better understanding of the molecular pathways specific to each stress, as well as those that are shared by all stresses. Consequently, this knowledge will allow for the refinement of post-harvest treatment protocols, potentially including the delivery of lower dosages and/or the use of combined treatments. This current presentation will focus on the study done on 3rd instar larvae of Mediterranean fruit fly in response to lethal heat stress.

Briefly, we first identified the sub-lethal dosage for heat treatment (the dosage that kills 75% of treated insect) through bioassay. Then, we investigated the gene expression profiles from the insect that has been treated right after treatment (T1) and two hours after the treatment (T2) using RNA sequencing. By comparing the gene expression profiles of the treated larvae to the control larvae (larvae that have not been treated; T0, pair-wise comparison), we found more genes are down-regulated compared to up-regulated. Surprisingly, many heat shock proteins were found to be lower express in the treated larvae, compared to the control larvae. The down-regulation of these genes might indicate that the cell is in the process of apoptosis or cell death. Further analysis on RNA seq data using the random forest algorithm also indicates some

key genes involved in cell death and autophagy which might be affected by the treatments. Genes identified through RNA seq experiments are now candidate genes for follow up study using further RNA seq experiments.

INVITED TALK

# Microbial community structure and dynamic response to disturbance in Singapore coastal waters.

**Federico Lauro**
Nanyang Technological University, Singapore

The island of Singapore depends on the surrounding marine environment for ecosystem services such as food production from aquaculture, recreational water-related activities/eco-tourism and drinking water supply (e.g. desalination). The island has undergone a rapid urbanization in the last 30 years resulting in the alteration of biotic and abiotic conditions and perturbing microbial ecosystems from their natural state. A combination of hydrodynamic and anthropogenic forces partitions the coastal waters surrounding the island into defined trophic habitat types. To the North, the Johor Strait is nutrient rich and subjected to a wide range of environmental fluctuations resulting from anthropogenic activities. To the South, the Singapore Strait is more oligotrophic and is subjected to seasonal variability in the biogeochemical environment correlating to seasonal monsoon reversal and oceanographic connectivity. In this talk I will show how this unique configuration provides an unprecedented opportunity for natural experiments aimed to understand how structural and functional changes affect the ecological processes driven by equatorial microbial communities in response to the trophic status of the environment.

INVITED TALK

# PCR-DCGE Analysis for Identification of Intestinal Bacteria in the Black Soldier Fly Larvae, Hermetia Illucens

**Nguyen Bao Quoc**[1], Nguyen Viet Ngoc Hanh[1], Ho Thi Thu Trang[1], Pham The Hai[2], Nguyen Thi Thu Thuy[2], Nguyen Ngoc Bao Chau[3]
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*Hermetia illucens* so-called the black soldier fly (BSF), has been reported as an important role to play an important role for treating domestic, agricultural or manure wastes. With the ability to live well in the environment of decomposition or waste containing a variety of substances and many types of microorganisms can be related to the intestinal microflora, helping the larvae digest, grow and develop well, even though creating protection against parasites and pathogens. In this study, the intestinal microflora of BSF larvae is used to partly explain the excellent treatment and the ability to live well in the waste environment. DGGE method is used to determine the microbiological composition of the larvae, thereby determining the dominant species in the intestine and having a clearer view of the application of BSF as well as knowledge of intestinal microflora. The results indicated the appearance of 12 dominant bands of larval intestinal microflora belonging to 4 groups: *Clostridium, Dialister, Dysgonomonas* and *Actinomycetales*. Among the 4 groups, the *Clostridium* group is the most dominant compared to other groups and present in all 3 parts of larvae intestine.

# Lecture Summary

LECTURE SUMMARY

# Hands-on session: Create 16S amplicon sequencing libraries

Kay Anantanawat

In this hands-on session, we will learn how to create 16S amplicon NGS library. We will amplify the V3 and V4 regions of 16S rRNA genes in the metagenomic samples. The session will include setting up amplicon PCR and cleaning up PCR reaction using SPRI magnetic beads. The product of the class can then be barcoded and sent for sequencing.

LECTURE SUMMARY

# Microbial ecology: Theory and practice for 16S rRNA amplicon biodiversity analyses

Thomas Jeffries

Participants will be introduced to the theory underpinning microbiome ecology studies In agriculture and workflows for addressing ecological questions using amplicon sequencing (16S rDNA). There will be a focus on the statistical interpretation of the outputs and visualising diversity data. Following a description of the tools, students will work with agricultural microbiomes and solve real-world problems with next-generation sequencing datasets.

LECTURE SUMMARY

# RNA Seq Essential for biologists

Kay Anantanawat
Passorn Wonnapinij

This class will introduce you to RNA sequencing (RNA Seq); the technique uses to study the gene expression profiles in organisms. We will go through the analysis process of RNA seq data, including sequencing read alignments, counting abundance, normalisation, data exploration and differentially expression analysis. We will explore two R packages used for differential expression analysis: edgeR and DESeq2. Finally, we will touch a bit on how to use machine learning methods such as random forest to identify the main genes, in which their expression profiles define the clustering between control and treatment groups.

By the end of the class, you should be able to:

- Understand the normalisation process: why we need to normalise the data? What are the normalisation methods?
- Understand how to interpret the data generated from the analysis, including heatmap, PCA and MDS plots.
- Understand the p-value, False Discovery Rate and adjusted p-value

LECTURE SUMMARY

# Computational metagenomics hands-on workshop

Aaron E. Darling
Krithika Arumugam

This tutorial teaches computational metagenomics from basic principles of study design through to sequencing, assembly, and hypothesis testing. The day begins with a discussion of the principles of microbial communities, microbiome profiling, and metagenome sequencing. We continue by using a particular example system where we would like to test the effect of an agricultural probiotic. Attendees will do hands-on study design and power calculations for metagenomic sequencing in this context. Then, using metagenomic data, we will learn to do basic sample QC and taxonomic profiling (hands-on).

The afternoon will commence with a lecture that outlines some of the limitations of different metagenomic analysis methods. We will learn why taxonomic analysis methods lack the resolution to answer many of the key questions we seek to answer with metagenomics. Current approaches to genome binning, e.g. methods that resolve genomes from metagenomes, will be discussed along with their limitations. We will then proceed with a hands-on workshop to learn how to run metagenome assemblies and reconstruct genomes using time-series binning. We will experiment with different binning strategies to understand what works well and what doesn't.

A final lecture component we will then discuss the principles of advanced techniques such as metagenomic Hi-C. This method can resolve certain problems associated with other metagenomic methods, but comes with its own set of challenges.

We will then have a hands-on component in which we learn how to apply Hi-C data to help resolve additional questions in metagenome analysis.

LECTURE SUMMARY

# Transcriptome bioinformatic approaches for eukaryotic species of agricultural interest

Alexie Papanicolaou

Transcriptomic analysis is essential for the identification of genes and their functional analysis. With a plethora of software and sequencing technologies, bioinformaticians today are swamped with options. In this class, we will discuss these options for species with and without a sequenced genome. We will explore de-novo assembly, differential expression, and biomarker development, genome annotation, focusing on the technical aspects, and when to choose each option. Even though the expertise is on eukaryotic genomes, much of what we talk about would be transferable to bacteria (with caveats). In this class, you will see the results of real large-scale analyses, focus on topics of specific interest to bioinformaticians such as parameterisation, importance of metadata, data management, and how to work with that rare biologists who may believe that RNA-Seq data analysis should be quicker than month-long development and use of a single qPCR marker.

# Poster Abstract

POSTER

# Phylogenomic approach to allergenicity in genetically modified soybeans

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Despite rapid growth of the genetic modified (GM) crops, effective evaluations of genetic modification on allergenicity are still lacking. Gly m Bd 30 K is cross-reactive with cow milk protein casein, Gly m 4, and with birch pollen allergen Bet v 1. Here we compared the allergenicity between GM and non-GM soybeans with respect to the foci Gly m 4 and Gly m Bd 30K. Recombinant allergens of Gly m Bd 30K and Gly m 4 were generated and polyclonal antibodies raised to identify these two allergenic components in soybeans. GM soybean was first PCR-confirmed using 35S-promoter. A total of 20 soybeans (half GM, half non-GM) obtained from food market were used to assess their allergenicity based on IgE-binding and histamine release. The concentrations of Gly m Bd 30K and Gly m 4 in soybeans were then determined. Most soybean-allergic patients (9 out of 10) showed IgE-positive reactions to the allergen of 30kDa in molecular weight. That allergen turned out to be Glycine max Gly m Bd 30K based on LC/MS/MS analyses. Gly m Bd 30K is therefore the major allergen in the soybean. An increase in the transcription of both the Gly m 4 (stress-induced protein SAM22) and Gly m Bd 28K (soybean allergen precursor) was found after GM. The protein concentrations of Gly m 4 and Gly m Bd 30K were no statistically significant differences between Non-GM and GM soybeans. There were also no statistical significances between them in the tests of IgE binding and histamine release. In conclusions, soybeans showed similar concentrations of Gly m Bd 30K and Gly m 4 regardless of genetically modified or not. The allergenicity of both Gly m Bd 30K and Gly m 4 were therefore not altered after genetic modification. Patients showing hypersensitivity to soybeans and

who had pre-existing allergy to birch pollen and cow milk casein might not further increase their allergic reactions following exposures to the GM soybeans.

POSTER

# Expression based studies for identifying key regulatory molecules for rheumatoid arthritis

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DNA microarrays are a powerful tool to investigate differential gene expression for thousands of genes simultaneously. Gene expression profiling provides unprecedented opportunities to study patterns of gene expression regulation, for example, in diseases or developmental processes. Rheumatoid arthritis is an autoimmune disease that causes chronic joint inflammation. The dominant local cell populations in joints affected by rheumatoid arthritis are synovial and cartilage cells. Synovial cells can be divided into fibroblast-like and macrophage-like synoviocytes. Synovial fibroblasts are in the pathogenesis of Rheumatoid Arthritis. Since Rheumatoid Arthritis Synovial Fibroblasts (RASFs) mediate most relevant pathways of joint destruction, molecular insights into these cells constitute an important target for novel therapeutic approaches that inhibit the destruction of cartilage and bone in RA. For understanding the differentially expressed genes in synovial Fibroblast for Rheumatoid Arthritis, analysis of gene expression profiling data derived from micro-array technology was done. The dataset was downloaded by the publically available server GEO database, the raw data file was normalized and compared using DChip. The normalized file has a set of 12,558 genes. The differentially expressed genes were identified on the basis of 1.5 fold change. Genes with fold change values ≥ 1.5 were categorized as up-regulated and those genes with fold change values ≤ 1.5 were categorized as down-regulated genes. 2058 genes were found to be up-regulated and 9428 genes were found to be down-regulated. These differentially expressed genes were loaded into Cladist for clustering and correlation analysis. Gene-gene correlation analysis of the individual gene clusters was done and the highly correlated gene interaction where the correlation coefficient was ≥ 0.9 was observed for down regulated and ≤ 0.8 for up regulated. Network was built

using Cytoscape for each cluster and genes which had the relevance Functional characterization gave an insight for developing novel targets for rheumatoid Arthritis.

POSTER

# Meta-barcoding and genomic insights into Mānuka (Lepstospermum scoparium) microorganisms

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Microorganisms are key metabolite producers in numerous biological systems. In plants, they engage in symbiotic or antagonistic interactions, where multiple species coexist and cooperate or compete for substrates. The endemic New Zealand plant *Leptospermum scoparium* (Mānuka) contains dihydroxycetone (DHA) in its nectar, which chemically converts in the honey into methylglyoxal, associated with the antimicrobial and antioxidant properties of Mānuka honey. The origin of this compound is still unclear. Interestingly, bacteria and yeast that are often present in floral nectar have the capability to produce DHA from several types of carbohydrates and other photosynthesis intermediates, suggesting that this compound could have a microbial origin. Pollinators add complexity to this interaction by influencing the microbial composition of nectar and its physicochemical environment. This study aims to determine the diversity and abundance of bacteria and fungi in Mānuka nectar, identify the mechanisms of DHA production, and gain a better understanding of the nectar-microbial interaction and the effect on pollinators on microbial communities. To achieve these aims we cultured bacteria from Mānuka flowers and began their functional characterisation including metabolic capabilities and microbe-microbe interactions. Culturing showed a high abundance of the metabolically versatile bacterium *Pseudomonas fluorescens* in the nectar of Mānuka plants. As Mānuka plants produce minuscule amounts of nectar, we optimized the methods for isolation of metagenomic DNA to determine its microbial community composition using high throughput sequencing. This project will

contribute to understanding the ecology of tri-trophic interactions between plant, microorganisms, and pollinators and their effect on secondary metabolites.

POSTER

# Towards understanding genetic influences on migration timing in bar-tailed godwits

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Describing mechanistic links between genetic variation and behaviour-level variation in free-living animals presents a major scientific challenge. Migratory bar-tailed godwits that breed in Alaska and over-winter in Australasia perform the longest recorded avian non-stop migratory flight, directly from Alaska to New Zealand (N.Z.), 12,000 km away. The control of avian migration timing is believed to involve changes in daylength as perceived by the birds, but this insight alone does not explain the observed inter-individual variation in different departure schedules. It is anticipated that genetic variation is causally and mechanistically associated with such phenotypic variation. To investigate this possibility, we looked for associations between individuals' migration departure time (chronophenotypes) and genotypic variation at loci that form elements of the endogenous circadian clock, of the Hypothalamic-Pituitary-Gonadal axis, of photoreception, and fat metabolism. A reference bar-tailed godwit genome was used to identify the candidate genes and 19 additional genomes which served to prioritize 4,919 SNPs that were genotyped on 265 individuals. 3,412 SNPs (harboring 120 candidate genes) passed our filtering parameters. Our assessment of the genetic architecture underlying migratory departure time together with a population structure analysis suggested that this phenotype has a highly polygenic basis. Although we did not identify any strong association of any specific candidate genes with an 'earlier' or 'later' chronophenotype, we found that 30 SNPs can explain significantly around 36% of it. Whether this finding is principally founded on a smaller number of SNPs is something that deserves further investigation.

POSTER

# Extracellular DNA in Monochloraminated Drinking Water and its Influence on DNA-based Profiling of Microbial Community

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The interaction between biofilms and disinfectant in drinking water distribution systems (DWDS) as well as the role of this interaction in the formation of disinfection by-products has been extensively studied in recent years. In contrast, lysis of cells and/or release of intracellular biomolecules from inactivated/damaged cells and their fate and implications are an overlooked aspect of DWDS. In particular, DNA, once released into DWDS, may persist in water as extracellular DNA (eDNA). In this study, we report for the first time that the total DNA extracted from monochloraminated drinking water contains a high fraction of eDNA. Drinking water samples were obtained from locations 1 (~20-year-old pipeline) and 2 (~7-year-old pipeline) using glass fiber membranes with a nominal pore size of 0.4 µm. At location 1, 85–386 ng of eDNA was found per liter of sampled water, which accounted for 52 ± 12% of total DNA, while at location 2, 33–58 ng of eDNA was found per liter of sampled water, accounting for 42 ± 8% of the total DNA. We further showed that the eDNA was mainly of bacterial and archaeal origin and is expected to contribute to the analysis of the microbial community through the 16S rRNA gene amplicon sequencing approach. Removal of eDNA reduced α diversity, increased community evenness, and changed the relative abundance of detected taxa. Among the top 25 genera detected at each location, *oc32*, *Sphingomonas* and *Piscinibacter* were found to be significantly affected by the DNase treatment at location 1. At location 2, detection of *Methylophilus*, *Curvibacter*, *Caulobacter*, *Algoriphagus*, *Bdellovibrio*, *Hydrogenophaga*, *Azospira*, *Porphyrobacter*, *DSSF69*, *Piscinibacter*, *Nitrosomonas*, *Sphingomonas*, *Hyphomicrobium*, and

*Singulisphaera* was significantly affected. Our findings lead to future research questions about the source, fate, and implications of eDNA in DWDS.