

# INTERNATIONAL PROTEOMICS CONFERENCE 2017

in conjunction with

The 4<sup>th</sup> Conference of Asia Oceania  
Agricultural Proteomics Organization

*Proteomics in  
Biotechnology &  
Life Sciences*

[ipc2017.weebly.com](http://ipc2017.weebly.com)

15<sup>th</sup> – 17<sup>th</sup> August, 2017

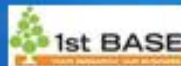
KLTC, Kuala Lumpur, Malaysia

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## *From the Chair*

On behalf of the organizing committee, I would like to extend a warm welcome to all the participants of the International Proteomic Conference 2017 in conjunction with the 4<sup>th</sup> Conference of the Asia Oceania Agricultural Proteomics Organization (AOAPO).

In this conference, the organizing committee has put together a program of keynote and plenary addresses, seminar and poster sessions which are delivered by both experienced and young international experts with the aims of fostering knowledge exchange, networking, information and technology sharing in the field of proteomics. We hope that the sessions will inspire more young researchers to join in the use of this powerful proteomics tool for diverse applications.

I would like to take this opportunity to thank The University of Nottingham Malaysia Campus for allowing us to use its Kuala Lumpur Teaching Centre (KLTC) as our conference venue. Since KLTC is located at the heart of KL city, it will enable our oversea participants to have a chance to explore the beauty of the capital city.

With great gratitude, I would like to thank all the organizing committee members for their hard work and dedications to ensure successful running of this conference. To all the participants, I wish you a fruitful, productive and memorable meeting.

**Chin Chiew Foan**  
**Chair**

**Organizing committee of the International Proteomics Conference 2017**

## *From the President*

On behalf of the organizing committee, it is my great pleasure to welcome you to the International Proteomics Conference in conjunction with the 4th Asia Oceania Agricultural Proteomics Organization Conference to be held at the University of Nottingham Teaching Centre, Kuala Lumpur, Malaysia from 15 to 17 August 2017. This Conference is organized by Malaysia Agricultural Proteomics Society and Asia Oceania Agricultural Proteomics Organization.

The theme for the conference is “Proteomics in Biotechnology and Life Sciences”, which aptly represents the current state of proteomics technology that is applicable in all aspects of biological sciences. This conference covers topics related to proteomics that range from agriculture to biomedical sciences. The organizing committee has put together a program with presentations from distinguished researchers in various fields of proteomics from all over the world. The conference provides a platform for knowledge exchange, networking, information, and technology sharing.

It will enable you to have a fruitful, remarkable, and memorable time in Kuala Lumpur. In Kuala Lumpur, you will experience a plethora of rich diversity in many aspects of life such as nature, people, food, and cultural heritage, which you will find exciting and interesting. I would like to thank all the Committee and sub-Committee members for their hard work and dedication in ensuring the successful delivery of this conference. To all the participants, I wish you a fruitful meeting.

**Setsuko Komatsu**

**President of Asia Oceania Agricultural Proteomics Organization**

## ORGANIZING COMMITTEE

<b>Chairperson</b>	<b>Assoc. Prof. Dr Chin Chiew Foan</b>
<b>International Advisory Committee</b>	
Prof. Dr Setsuko Komatsu Prof. Dr Chen Sixue Prof. Dr Shaojun Dai Prof. Dr Subhra Chakraborty	
<b>Local Organizing Committee</b>	
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<b>Scientific</b>	Dr Jameel R. Al-Obaidi Dr Benjamin Lau Yii Chung Ms Nursyuhaida Mohd Hanafi
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<b>Logistics</b>	Dr Wan Mohd Aizat Dr Dhilia Udie Lamasudin
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## SCIENTIFIC PROGRAM

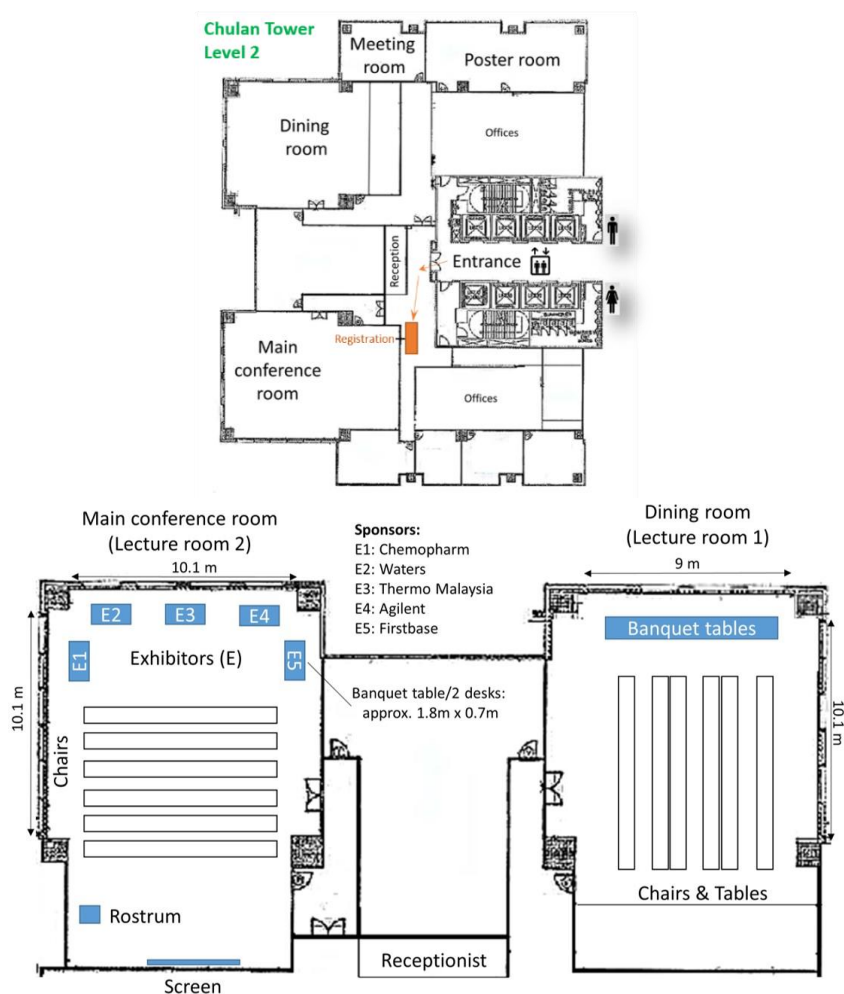
Day 1 – 15 <sup>th</sup> August 2017	
Time	Program
8.00 – 9.00 am	Registration Opens
9.00 – 9.20 am	Opening Ceremony and Welcome Address
9.20 – 10.05 am	<b>Keynote 1:</b> Characterization of Stress Responsive Mechanisms in Soybean under Abiotic Stresses using Quantitative Proteomic Approach <i>Setsuko Komatsu, University of Tsukuba, Japan</i> <b>Chairperson: Chin Chiew Foan</b>
10.05 – 10.30 am	Group Photo, Networking, Refreshment Break and Poster viewing
FOOD PROTEOMICS	
10.30 – 11.00 am	<b>Plenary 1:</b> Comparative Nuclear Proteomic Analyses of Rice Blast Illustrates Novel Regulators of Plant Immunity <i>Subhra Chakraborty, National Institute of Plant Genome Research, India</i> <b>Chairperson: Setsuko Komatsu</b>
11.00 – 11.30 am	<b>Plenary 2:</b> Proteomics Analysis of Drought Stress Response in Two Rice Varieties with Contrasting Phenotypes <i>Paul A Haynes, Macquarie University, Australia</i> <b>Chairperson: Subhra Chakraborty</b>
11.30 – 11.50 am	<b>Talk 1:</b> Protein Pattern of Rice ( <i>Oryza sativa</i> L.) Grain Helps to Analyse Cooking Quality Properties <i>Mohammad-Zaman Nouri, Rice Research Institute of Iran, Iran</i>
11.50 am -12.10 pm	<b>Tech Talk 1:</b> Recent Contributions of Orbitrap Mass Spectrometry-Based Proteomic Techniques in Food Safety and Authenticity Analyses <i>Lin-Tang Goh, Thermo Fisher Scientific</i>
12.10 – 1.30 pm	<b>Lunch Break</b>
1.30 – 2.30 pm	<b>Poster Presentation 1</b>
ANALYTICAL, MICROBIAL AND INDUSTRIAL PROTEOMICS	
2.30 – 3.00 pm	<b>Plenary 3:</b> Reproducible Evaluation of the Accuracy of Proteomics Methods for Protein Identification and Quantification <i>Susan R Wilson, University of New South Wales, Australia</i> <b>Chairperson: Paul A Haynes</b>
3.00 – 3.30 pm	<b>Plenary 4:</b> Development of Redox Proteomics Technologies and Application in Stomatal Guard Cell Immunity Research <i>Sixue Chen, University of Florida, USA</i>
3.30 – 3.50 pm	<b>Talk 2:</b> Utilization of Phage-Host Interaction to Enhance the Role of Phage Therapy as a Tool to Disperse MRSA Biofilm <i>Khulood Hamid Dakheel, Universiti Putra Malaysia, Malaysia</i>
3.50 – 4.10 pm	<b>Talk 3:</b> A proteomic approach to investigate the effect of different cultivation methods on the production of carrageenan in red seaweed, <i>Kappaphycus alvarezii</i>



	<i>Siti Rokhiyah Ahmad Usulidin, Agro-Biotechnology Institute, Malaysia</i>
4.10 – 4.30 pm	Networking and Refreshment Break
4.30 – 4.50 pm	<b>Tech Talk 2: Promarker™: A Comprehensive Mass Spectrometry Based Biomarker Discovery and Validation Platform as Applied to Diabetic Kidney Disease</b> <i>Scot D Bringans, Proteomics International, Australia</i> <b>Chairperson: Sixue Chen</b>
4.50 – 5.10 pm	<b>Talk 4:</b> Comparative Proteomics Profiling of Two Ganoderma Species during <i>In-Vitro</i> Interaction with Oil Palm Root <i>Siti Nahdatul Isnaini Said Hussin, Agro-Biotechnology Institute, Malaysia</i>
5.10 – 5.30 pm	<b>Talk 5:</b> Comparative Proteomic Study of Oil Palm Roots in Response to <i>In Vitro</i> Inoculation with Pathogenic & Non-pathogenic <i>Ganoderma</i> spp. <i>Norasfaliza Rahmad, Universiti Putra Malaysia, Malaysia</i>
<b>END OF DAY 1</b>	
<b>Day 2 – 16<sup>th</sup> August 2017</b>	
<b>AGRICULTURAL PROTEOMICS</b>	
<b>Time</b>	<b>Program</b>
9.00 – 9.45 am	<b>Keynote 2:</b> Protein Degradation and Synthesis Rates in Leaf Growth and Development to Understand Energy Use and the Maintenance of Enzyme Function <i>A Harvey Millar, University of Western Australia, Australia</i> <b>Chairperson: Wan Mohd Aizat</b>
9.45 – 10.15 am	<b>Plenary 5:</b> Quantitative Proteomic Atlas Uncovering the Widespread Ubiquitylation Regulation in the Embryo of Germinating Rice Seed <i>Pingfang Yang, Chinese Academy of Sciences, China</i>
10.15 – 10.45 am	<b>Plenary 6:</b> Na <sub>2</sub> CO <sub>3</sub> -responsive Photosynthetic and ROS Scavenging Mechanisms in Chloroplasts of Alkaligrass Revealed by Phosphoproteomics <i>Shaojun Dai, Shanghai Normal University, China</i>
10.45 – 11.00 am	Networking and Refreshment Break
11.00 – 11.20 am	<b>Talk 6:</b> Identification of Proteins in Pitcher Fluid of <i>Nepenthes</i> Species Through Proteomics Informed by Transcriptomics Approach <i>Hoe-Han Goh, Universiti Kebangsaan Malaysia, Malaysia</i> <b>Chairperson: A Harvey Millar</b>
11.20 – 11.40 am	<b>Talk 7:</b> Proteome-wide Response of Aquilaria Tree to Agarwood Induction Treatments <i>Shiou Yih Lee, Universiti Putra Malaysia, Malaysia</i>
11.40 am – 12.00 pm	<b>Talk 8:</b> Molecular Comparison between Jojoba Male and Female Individual Plants Revealed Differences in Protein Functions <i>Nursyuhaida Mohd Hanafi, Agro-Biotechnology Institute, Malaysia</i>

12.00 – 12.20 pm	<b>Talk 9:</b> Sub-Zero Acclimation Induces Enhancement of Freezing Tolerance Accompanied by Changes of Extracellular Proteome and Cell Wall Characteristics in <i>Arabidopsis</i> <i>Daisuke Takahashi, Max-Planck Institute of Molecular Plant Physiology, Germany</i>
12.20 – 1.30 pm	<b>Lunch Break</b>
1.30 – 2.30 pm	<b>Poster Presentation 2</b>
2.30 – 3.00 pm	<b>Plenary 7:</b> The Omics for Agriculture and Biodiversity <i>Mohd Hasnain Hussain, Universiti Malaysia Sarawak, Malaysia</i> <b>Chairperson: Jameel R. Al-Obaidi</b>
3.00 – 3.30 pm	<b>Plenary 8:</b> Barley and <i>Piriformospora indica</i> Relationship: What Has an Integrative Omics Approach Taught Us? <i>Ghasem Hosseini Salekdeh, Agricultural Biotechnology Research Institute of Iran, Iran</i>
3.30 – 3.50 pm	<b>Talk 10:</b> The Proteomic Landscape of Soybean Responses Against Flooding Stress: Insights from Post-Translational Modifications <i>Akiko Hashiguchi, University of Tsukuba, Japan</i>
3.50 – 4.20 pm	<b>Talk 11:</b> Quantitative Proteomics Analysis of Soybean Mitochondria on Exposure to Varying Sizes of Aluminium Oxide Nanoparticles Under Flooding Stress <i>Ghazala Mustafa, Quaid-i-Azam University, Pakistan</i>
4.20 – 4.40 pm	Networking and Refreshment Break
4.40 – 5.10 pm	<b>Talk 12:</b> Soybean Proteomics: A Leading Tool in Exploring Effects of Flooding Stress Using Gel-based Proteomic Technique <i>Amana Khatoon, Kohat University of Science and Technology, Pakistan</i> <b>Chairperson: Hoe-Han Goh</b>
5.10 – 5.30 pm	<b>Talk 13:</b> Drought Stress Characterization of Tolerant and Sensitive Barley Genotypes Through Proteomics Analysis <i>Rehana Kausar, University of Azad Jammu &amp; Kashmir, Pakistan</i>
5.30 – 8.00 pm	<b>AOAPO Council Meeting</b>
8.30 -10.30 pm	<b>Conference Dinner</b>
END OF DAY 2	
Day 3 – 17 <sup>th</sup> August 2017	
MEDICAL PROTEOMICS	
<b>Time</b>	<b>Program</b>
9.00 – 9.45 am	<b>Keynote 3:</b> From Proteomic Analysis to Functional Analysis <i>Fook Tim Chew, National University of Singapore, Singapore</i> <b>Chairperson: Shaojun Dai</b>
9.45 – 10.15 am	<b>Plenary 9:</b> Cancer Biomarker Discovery: Lectin-Based Proteomic Studies <i>Onn Haji Hashim, University of Malaya, Malaysia</i>
10.15 – 10.45 am	<b>Plenary 10:</b> Chemical Proteomics Investigation of Artemisinin's Antimalarial and Anticancer Mechanisms <i>Qingsong Lin, National University of Singapore, Singapore</i>
10.45 – 11.00 am	Networking and Refreshment Break

11.00 – 11.20 am	<b>Talk 14:</b> Proteomic Analysis to Reveal Molecular Mechanism of Phenolic Acids Accumulation in Leaves of <i>Salvia miltiorrhiza</i> by UV-B Radiation <i>Xiaojian Yin, China Pharmaceutical University, China</i> <b>Chairperson: Fook Tim Chew</b>
11.20 – 11.40 am	<b>Talk 15:</b> Proteomics Analysis of Kesum ( <i>Persicaria minor</i> Huds.) Herbal Plant Upon Methyl Jasmonate Treatment <i>Wan Mohd Aizat, Universiti Kebangsaan Malaysia, Malaysia</i>
11.40 am - 12.00 pm	<b>Talk 16:</b> Proteomic Analysis of BmNPV Resistance in the Silkworm Reared on UV-B Induced Mulberry Leaves by SWATH-MS <i>Wei Zhu, Zhejiang University, China</i>
12.00 – 12.20 pm	<b>AWARDS AND CLOSING CEREMONY</b>
12.20 – 2.00 pm	Lunch
<b>END OF CONFERENCE</b>	





## KEYNOTE

**Keynote 1:** Characterization of Stress Responsive Mechanisms in Soybean under Abiotic Stresses using Quantitative Proteomic Approach

*Setsuko Komatsu, University of Tsukuba, Japan*

**Keynote 2:** Protein Degradation and Synthesis Rates in Leaf Growth and Development to Understand Energy Use and the Maintenance of Enzyme Function

*A Harvey Millar, University of Western Australia, Australia*

**Keynote 3:** From Proteomic Analysis to Functional Analysis

*Fook Tim Chew, National University of Singapore, Singapore*

## PLENARY

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*Subhra Chakraborty, National Institute of Plant Genome Research, India*

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*Paul A Haynes, Macquarie University, Australia*

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*Onn Haji Hashim, University of Malaya, Malaysia*

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*Qingsong Lin, National University of Singapore, Singapore*

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**Talk 16:** Proteomic Analysis of BmNPV Resistance in the Silkworm Reared on UV-B Induced Mulberry Leaves by SWATH-MS

*Wei Zhu, Zhejiang University, China*

## TECHNICAL TALKS

**Tech Talk 1:** Recent Contributions of Orbitrap Mass Spectrometry-Based Proteomic Techniques in Food Safety and Authenticity Analyses

*Lin-Tang Goh, Thermo Fisher Scientific*

**Tech Talk 2:** Promarker™: A Comprehensive Mass Spectrometry Based Biomarker Discovery and Validation Platform as Applied to Diabetic Kidney Disease

*Scot D Bringans, Proteomics International, Australia*

## POSTER PRESENTERS

**P01** Comparative Proteomic Analysis of *in vitro* Fungal-Bacterial Interaction

*Seo Hyun Lee, Pusan National University, Republic of Korea*

**P02** Identification and Validation of Putative *Erwinia mallotivora* Effector Proteins through iTRAQ Protein and Real Time PCR Analysis

*Norliza Abu Bakar, MARDI, Malaysia*

**P03** Proteomic Profiling of Human Cervical Cancer Cells Treated with a Selected Diarylpentanoid

*Rakesh Naidu, Monash University Malaysia, Malaysia*

**P04** Liver Heme Oxygenase-1 Expression is Positively Induced by Palm Oil-Derived Tocotrienol Rich Fraction (TRF) Supplementation in Mice

*Azman Abdullah, Universiti Kebangsaan Malaysia, Malaysia*

**P05** Leucine-rich alpha-2-glycoprotein 1 (LRG1) in Colorectal Cancer: A Real Potential Biomarker?

*Siok-Fong Chin, Universiti Kebangsaan Malaysia, Malaysia*

- P06** Protein Profiles Of LPS-Induced and RECA Treated In Rat Hippocampus  
*Nor Datiakma Mat Amin, FRIM, Malaysia*
- P07** Protein Profiling of Human Gut Secretome in Healthy and Colorectal Cancer Patients: A Preliminary Finding  
*Putri Intan Hafizah Megat Mohd Azlan, Universiti Kebangsaan Malaysia, Malaysia*
- P08** Secreted Proteome of Human Bronchial Epithelial Cells (BEAS-2B) After Interaction with *Cladosporium sphaerospermum*  
*Sing Gee Lo, International Medical University, Malaysia*
- P09** Development of a Novel Method for the Enrichment of Anticancer Peptides from *Glycine max* Seeds  
*Ye Eun Cheon, Pusan National University, Republic of Korea*
- P10** Chemometric Elucidation of Oil Palm Mesocarp Proteomes  
*Hasliza Hassan, MPOB, Malaysia*
- P11** Identification of Wound-Response Proteins using 2D-Electrophoresis and LC-MS: A Case Study on Agarwood Tree Species, *Aquilaria malaccensis*  
*Muhammad Syahmi Hishamuddin, Universiti Putra Malaysia, Malaysia*
- P12** Proteomic Analysis on Root Callus and Protocorm-Like Body of *Vanilla planifolia* Andrews  
*Nadiatul Akmar Abd Aziz, University of Nottingham Malaysia Campus, Malaysia*
- P13** A Proteomic Analysis of Mahogany Embryos During Cold Stress Response  
*Noraliza Alias, FRIM, Malaysia*
- P14** Investigation of Differential Proteins in Calli of *Capsicum frutescens* Treated with Different Concentrations of Ferulic Acid  
*Nurshahirah Shuhaimi, University of Nottingham Malaysia Campus, Malaysia*
- P15** Identification of Virulence Related Proteins Associated to Blood Disease Bacterium, a Pathogen Causing Banana Blood Disease.  
*Rafidah Badrun, MARDI, Malaysia*

- P16** Mass Tagging for Oil Palm Root Proteome Discovery  
*Syahanim Shahwan, MPOB, Malaysia*
- P17** Prolonged Culture of *Boesenbergia rotunda* Cells Reveals Decreased Growth and Shoot Regeneration Capacity and Changes in Proteins  
*Aiman Faizudin Aziz, Universiti Teknologi MARA, Malaysia*
- P18** Identifying Lipid Peroxidation End Products (LPEPs) – Modified Proteins in Wheat Plants  
*Yen Yeen Chor, University of Western Australia, Australia*



## Keynote 1

### Characterization of Stress Responsive Mechanisms in Soybean under Abiotic Stresses using Quantitative Proteomic Approach

**Setsuko Komatsu\***, Xiaojian Yin, Xin Wang

*University of Tsukuba, Japan*

\***Correspondance:** komatsu.setsuko.fu@u.tsukuba.ac.jp

Global climate changes influence the magnitude and frequency of hydrological fluctuations and cause unfavorable environment for plant growth and development. Soybean is the important food crop with high abundant of protein, vegetable oil, and several phytochemicals. With the predominate values, soybean is cultivated with a long history, while it is sensitive to flooding and drought, which lead to deleterious effects on plant growth. Root growth was suppressed under flooding and drought, while the parameters of root elongation and root diameter differed between two conditions. To unveil the response mechanisms of soybean under flooding and drought, proteins were analyzed using the gel-free/label-free proteomic technique. The current findings suggest that biotin and biotinylation might participate in energy regulation under flooding and drought in the early-stage soybean. Calnexin together with protein disulfide isomerase -like proteins or heat shock proteins is responsive for the proper folding of the glycoproteins in the endoplasmic reticulum under flooding or drought. Moreover, calcium homeostasis is associated with endoplasmic reticulum-associated folding/degradation and alters the fermentative enzyme of pyruvate decarboxylase in energy regulation of soybean under flooding and drought.

**Keywords:** Abiotic stresses; Endoplasmic reticulum; Proteomics; Soybean

#### Biography



Setsuko Komatsu is the Chief of Field Omics Research at the National Institute of Crop Science and a Professor at the University of Tsukuba, Japan. She serves as an associate editor in Journal of Proteome Research and Frontiers in Plant Science. Furthermore, she is the President of Asia Oceania Agricultural Proteomics Organisation (AOAPO), which is a platform for the betterment in the field of agriculture proteomics.

## Plenary 1

### Comparative Nuclear Proteomic Analyses of Rice Blast Illustrates Novel Regulators of Plant Immunity

Kanika Narula, Pooja Choudhary, Sudip Ghosh, Pooja Aggarwal, Niranjan Chakraborty, and **Subhra Chakraborty\***

National Institute of Plant Genome Research, Aruna Asaf Ali Marg, New Delhi-110067, India

\*Correspondence: subhrac@hotmail.com

Blast disease caused by *Magnaporthe grisea* is a major impediment for global rice productivity. To elucidate molecular mechanism associated with cellular immunity, the temporal changes of nuclear proteome and metabolome was studied in blast resistant and susceptible rice (*Oryza sativa*) cultivars upon *M. grisea* infection. Temporal proteome and metabolome was developed with nuclear enriched fraction using quantitative 2-DE and iTRAQ coupled ESI-MS/MS and Triple-TOF/MS and GC-MS analyses, respectively. The differential display of *Magnaporthe* infected resistant and susceptible rice cultivar proteomes revealed 215 immune-responsive proteins and 150 patho-stress responsive proteins presumably associated with nucleic acid biogenesis and chromatin remodeling. Furthermore, blast-responsive metabolome profile displayed significant alteration in 165 and 198 metabolites associated with global metabolic pathways paralleling the proteomic analysis. Network analysis identified major protein hubs enriched in known and novel disease- and immunity-related prognostic proteins pointing towards the onset and context of disease signaling and metabolic pathway activations. Multivariate and network-based analyses successfully revealed the difference between the covariance structures of the integrated data sets. Combined analyses of multi-omics landscape of rice not only provide useful insights into the underlying mechanism of blast resistance, but also enlist novel biomarkers for targeted genetic manipulation for food and nutrition security.

**Keywords:** Food and nutrition; Immunity; Interactomics; Nucleus; Rice blast

#### Biography



Subhra Chakraborty, received her PhD training at Jawaharlal Nehru University, New Delhi, India and did Visiting Scientist work at Yale University, USA. She is currently Professor and one of the founding faculties at National Institute of Plant Genome Research (NIPGR), New Delhi, India. She has been instrumental in initiating and establishing Plant Proteomics and Translational Genomics research in India and contributed significantly in the basic and applied bio-technology research. The central aim of her research is to elucidate the molecular mechanism, proteomic pathways, and metabolic fingerprints involved in nutrient- and stress-response and to identify key genes for modifying such traits in plants.

## Plenary 2

### Proteomics Analysis of Drought Stress Response in Two Rice Varieties with Contrasting Phenotypes

Paul A. Haynes<sup>1</sup>, Yunqi Wu<sup>1</sup>, Mehdi Mirzaei<sup>1</sup>, Dana Pascovici<sup>2</sup> and Brian Atwell<sup>3</sup>

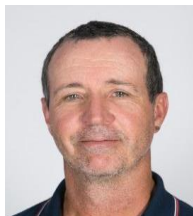
<sup>1</sup>Department of Chemistry and Biomolecular Sciences; <sup>2</sup>Australian Proteome Analysis Facility; <sup>3</sup>Department of Biological Sciences, Macquarie University, North Ryde, Australia

\*Correspondence: paul.haynes@mq.edu.au

We performed an extensive quantitative proteomics study of the two rice varieties Nipponbare and IAC 1131. The IAC 1131 variety is known to be relatively tolerant to drought stress, while Nipponbare is sensitive to drought stress. We analysed protein extracts from both mature leaf tissue, and shoot growing zone tissue prepared by microscopic dissection. Both tissue samples were analysed first by label free quantitation and then by tandem mass tags (MT) quantitation. We found that IAC1131 and Nipponbare respond very differently to drought stress – and different tissues from the same plant employ different strategies to deal with drought stress. IAC1131 appears to be better able to cope with stressful conditions by up-regulating a suite of proteins potentially involved in stress response in the mature leaf tissue, as well as maintaining the active stage of the growing zone by up-regulating a suite of proteins related to cell division. Secondary metabolism was repressed in IAC1131, apparently to save energy for stress response. Nipponbare, in contrast, lacks the range of stress responses shown by the drought tolerant variety and cannot maintain cell growth under drought stress.

**Keywords:** Drought stress; Normalized spectral abundance factors; *Oryza sativa*; Quantitative proteomics; TMT labelling

#### Biography



Paul A. Haynes graduated from Macquarie University with a Ph.D. in chemistry in 1994. After postdoctoral Fellowship positions at the Rockefeller University in New York and the University of Washington in Seattle, he was a principal scientist at the Tory Mesa Research Institute in San Diego, before taking up a position as an associate Professor at the University of Arizona in Tucson. He returned to Australia in 2006 to take up a position at Macquarie, and has been a Professor in the Department of Chemistry and Biomolecular Sciences since 2011. He is a senior editor of

Proteomics, Director of the ARC Training Centre for Molecular Technologies in the Food Industry, and vice president of the Asia Oceania Agricultural Proteomics Organization. His research interests lie in quantitative proteomics analysis across a range of biological systems, mainly focusing on plant and environmental proteomics.

## Talk 1

### Protein Pattern of Rice (*Oryza sativa* L.) Grain Helps to Analyze Cooking Quality Properties

**Mohammad-Zaman Nouri**<sup>1\*</sup>, Asefeh Latifi<sup>1</sup>, Fatemeh Habibi<sup>2</sup>, Seyyed Hamidreza Hashemi-petroudi<sup>3</sup>, Fatemeh Tavassoli<sup>1</sup>, Nahid Fat'hi<sup>1</sup>, Morteza Nasiri<sup>1</sup> & Mitra Yekta<sup>2</sup>

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Protein pattern of grain in three Iranian rice (*Oryza sativa* L.) genotypes, including Tarom-mahali (high quality landrace), Fajr and Shafagh (high-yielding varieties) were compared under different climate conditions to identify quality related proteins. Cooking quality related traits were measured and viscosity properties were analyzed before and after removal of proteins from samples. Protein content of rice grains ranged from 7.38 to 12.44 %. Amylogram analysis showed that removal of protein reduced the viscosity and the presence of protein makes the gel harder after cooling. According to the SDS-PAGE protein pattern, 10 protein bands were identified in which the expression of globulin-like protein, glutelin type-B1 and  $\beta$ -glutelin were higher in the landrace in cold climate condition. Expression of prolamin, a low-molecular weight protein, was higher in high-yielding varieties in both climate conditions. This protein can be considered as an index for quality testing of Iranian rice varieties. These results suggest that protein plays a significant role in quality properties of rice grains.

**Keywords:** Cooking quality; *Oryza sativa*; Prolamin

#### Biography



Dr. Mohammad-Zaman Nouri has completed his PhD in 2011 from University of Tsukuba, Japan. He worked on quantitative proteome analysis of plant plasma membrane under abiotic stress. He is currently the head of agronomy and plant breeding department at the rice research institute of Iran in Mazandaran province. He has published more than 30 papers and book chapters.

## Technical Talk 1

### Recent Contributions of Orbitrap Mass Spectrometry-Based Proteomic Techniques in Food Safety and Authenticity Analyses

**Lin-Tang Goh**

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Our food supply and variety are very expansive due to decades of advancement in food technology, production and distribution. To ensure consumption safety, regulatory monitoring needs to match this massive growth in scale and molecular physico-chemical diversity. The protein-based foods are an important category which must be monitored across its entire process (i.e. farm to fork) since they are more vulnerable to degradation/contamination. However, this class of food materials has been analytically challenging to measure due to its inherent molecular complexity, especially in the presence of their associated matrices. A decade ago, a newly developed high-resolution mass spectrometric technique (orbitrap mass analyzer) has found its way to offer both superior qualitative and quantitative measurement capabilities for protein-based samples, which many have exploited widely in the food research arena and food production industry. This talk highlights the key contributions of using such a novel HRMS technique to support the required assessments of food safety and authentication.

**Keywords:** Authenticity; Food safety, HRMS; Orbitrap mass analyzer

#### Biography



Dr Lin-Tang Goh is a senior Product Manager, Thermo Fisher Scientific. He has more than 15 years of mass spectrometry experience due to his previous employment as a senior scientist at the Bioprocessing Technology Institute (A\*STAR, Singapore) and the market development and applications manager at Waters (Asia Pacific). His current interests are in the emerging field of mass spectrometry-based Omics, where these technologies are being adapted and applied to other system-wide studies in biomedical, environmental and food sciences. He has also published widely in peer-reviewed scientific journals and given more than 120 technical presentations at various conferences, workshops and invited meetings. He obtained his Bachelor of Applied Science (Hons I) and PhD degree from the University of Queensland.

## Plenary 3

### Reproducible Evaluation of the Accuracy of Proteomics Methods for Protein Identification and Quantification

**Susan R Wilson**

*Stats Central, University of New South Wales, Australia*

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The generation of high-throughput data has become ubiquitous in all facets of modern ‘omics. Correspondingly, many approaches to dealing with such data have been developed, and associated software made widely available. What generally has been lacking is the reproducible and comprehensive evaluation of the accuracy of the various methods. To assess the different approaches to iTRAQ data in particular, we performed an experiment with a replicated Latin square design, and the data are publicly available. These data can be used for assessing the accuracy of methods for both protein identification and quantification. In this talk, particular emphasis will be on comparing preprocessing methods, and some interesting results from several case studies will be summarized. One of our primary findings highlights the need for experimenters to be using the principles of randomization and blocking to avoid results from analyses of the measurements being confounded by technical factors.

**Keywords:** Benchmarking; Reproducible research; iTRAQ; Latin square design

#### Biography



Susan R Wilson is currently Professor in Stats Central, University of New South Wales (fractional appointment). She obtained her B.Sc. from the University of Sydney, and Ph.D. from the Australian National University (ANU), was a Lecturer in the Department of Probability and Statistics, Sheffield University, UK, and then held various research positions at ANU where she is now an Emeritus Professor in the Mathematical Sciences Institute. Sue has over two hundred refereed scholarly publications in biostatistics and bioinformatics, with a particular emphasis in statistical genetics/genomics and statistical analyses of data produced by other omics technologies, including proteomics. These papers have been motivated primarily by her extensive consulting with researchers in

the biological and medical sciences, leading to statistical modelling developments to answer substantive research questions. Sue is an elected member of the International Statistical Institute, elected Fellow of the American Statistical Association and elected Fellow of the Institute of Mathematical Statistics. She has held the prestigious position of President of the International Biometric Society (IBS). In 2011 Sue was awarded the inaugural E. A. (Alf) Cornish Award for her contributions to Biometrics, and in 2012 awarded Honorary Life Membership of IBS ‘for outstanding contributions to the development and promotion of the discipline of Biometry’.



## Plenary 4

### Development of Redox Proteomics Technologies and Application in Stomatal Guard Cell Immunity Research

**Sixue Chen**

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Plant pathogens have caused serious crop losses, and they gain entry into leaves through natural stomatal pores on leaf surface. Stomatal innate immunity responses are fast processes that take place within the first few minutes of pathogen exposure. One objective of my lab research is to identify molecular switches that regulate the fast processes of stomatal movement in response to pathogen invasion. A novel redox proteomics method was developed to identify protein redox switches. The method was called cystMTTRAQ that combines two types of isobaric tags, cysteine tandem mass tags and isobaric tag for relative and absolute quantification, in one experiment. The method not only enables simultaneous analysis of cysteine redox changes and total protein level changes, but also allows the determination of bona fide redox modified cysteines in proteins through the correction of protein turnover. Using this powerful method, we were able to create an inventory of previously unknown potential redox regulated proteins, and highlight some potential regulatory mechanisms in stomatal guard cell innate immunity. Among these proteins, we identified a lipid transfer protein-II (LTP-II) undergoing oxidation in response to flg22 during stomatal closure. LTPs are small, basic proteins present in higher plants. They are known to be involved in key cellular processes such as stabilization of membranes, cell wall organization, and signal transduction. LTPs are also known to play important role in response to biotic and abiotic stresses, and in plant growth and development. Using reverse genetics, we have conducted functional studies of the LTP-II in stomatal guard cells. Here a potential mechanism by which LTP-II functions in stomatal guard cell defense response will be discussed.

#### Biography



Professor Sixue Chen completed his PhD in China and postdoctoral studies in Germany, Denmark, and University of Pennsylvania, USA. He is a Professor in Department of Biology, and Director of Proteomics and Mass Spectrometry at Interdisciplinary Center for Biotechnology Research of University of Florida, USA. His areas of expertise fall in Biochemistry, Plant Metabolism, Functional Genomics, Proteomics, Metabolomics, and Mass Spectrometry. He learned mass spectrometry when he was collaborating with a senior chemist at the Danish Royal Veterinary and Agricultural University 18 years ago. Dr. Chen carried out a lot of small molecule work at that time. Since joining University of Pennsylvania in 2001, he has worked on many different projects using proteomics and mass spectrometry. He serves as Assoc. Editor of Metabolomics & Front. Plant Proteomics, Editorial Advisory Board Member of J. Proteome Research & J. Proteomics, and Review Editors of Front. Plant Metabolism and Chemodiversity.

## Talk 2

### Utilization of Phage-Host Interaction to Enhance the Role of Phage Therapy as a Tool to Disperse MRSA Biofilm

**Khulood Hamid Dakheel<sup>1</sup>, Jameel R. Al-Obaidi<sup>2</sup>, Raha Abdul Rahim<sup>3,4</sup>, Vasantha Kumari Neela<sup>5</sup>, Tan Geok Hun<sup>6</sup> and Khatijah Yusoff<sup>1,4\*</sup>**

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Methicillin resistant *Staphylococcus aureus* (MRSA) are biofilm producers. A major problem of the biofilms is their inherent tolerance to host defenses and antibiotic therapies. This work explores the ability of phages to degrade MRSA biofilm. Enrichment culture was used to isolate bacteriophage against two isolates MRSA t127/4 and MRSA t223/20 that have ability to produce biofilm. Isolation two phages (UPMK\_1 and UPMK\_2) target MRSA t127/4 and MRSA t223/20 respectively, was performed. Both phages showed the ability to produce a halo around the clear zone of lysis on agars with their specific host. These phages demonstrated antagonistic infectivity on their planktonic host culture. This was further validated in a static biofilm (MTP) assay. In addition, the architecture of the biofilms labeled with SYTO9 and propidium iodide was studied *in situ* on confocal laser scanning microscopy (CLSM) before and after infection with the bacteriophages. The MTP assay and CLSM confirmed that these phages were able to degrade the biofilm. Phage-host interactions based on the detection of extracellular 2DE protein profile followed by MALDI-TOF-TOF mass spectrometer were used to clarify the role of these phages in biofilm degradation. We identified metabolic enzymes, proteins involved in the translation and transcription machineries and protein A as adaptive stress response. It was concluded that these proteins were important in the remodeling and dispersal of the biofilms during phage infection.

**Keywords:** Bacteriophage; MALDI-TOF/TOF; Microtiter plate; MRSA biofilm; Two-dimensional electrophoresis (2DE)

#### Biography



Khulood Hamid Dakheel completed her Bachelor and Master in Al-Mustansiriya University in Baghdad, Iraq in the field of Microbiology. In 2014, she has enrolled as PhD student in the field of genetic engineering and molecular biology, Department of Microbiology, Faculty of Biotechnology and Biomolecular Sciences at University Putra Malaysia (UPM). She is interested in studying the interaction between phages and their host.

### Talk 3

#### **A proteomic approach to investigate the effect of different cultivation methods on the production of carrageenan in red seaweed, *Kappaphycus alvarezii***

**Siti Rokhiyah Ahmad Usuldin<sup>1\*</sup>**, Jameel R. Al-Obaidi<sup>1</sup>, Nurhanani Razali<sup>2</sup>, Sarni Mat Junit<sup>2</sup>; Muhamad Johnny Ajang<sup>1</sup> & Norihan Mohd Saleh

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Red seaweed, *Kappaphycus alvarezii* is the main sources to produce κ-carrageenan in Malaysia. Carrageenan is a polysaccharide extracted from the red seaweed for use in pharmaceuticals and cosmetics industries. Due to many challenges to meet the increasing industrial demand for carrageenan, tissue culture technique has been used to improve the production of *K. alvarezii* seedlings and providing a sustainable source of better quality carrageenan. However, the understanding of the molecular mechanisms behind the technical aspects of macroalgal culture is still very limited compared to higher plants, necessitating further research. Thus, a proteomic study was conducted to compare the changes in protein expression in tissue-cultured and liquid-cultured *K. alvarezii* after 60 days of cultivation. A total of 45 protein spots were identified to be significantly different. Furthermore, changes in the proteins expression level were noticed in proteins related to energy production, metabolism and cellular maintenance. The protein changes in tissue-cultured seaweed possibly play an essential role in the production of carrageenan. This study is considered the first report of the *K. alvarezii* proteome, focusing on the carrageenan biosynthesis in different cultivation methods.

**Keywords:** Carrageenan; *Kappaphycus alvarezii*; Rhodophyta; Seaweed proteomics; Tissue culture

#### **Biography**



Presenter is a researcher from Agro-Biotechnology Institute, Malaysia (ABI) under the National Institutes of Biotechnology Malaysia (NIBM). She has received her degree from The University of Manchester, UK in Chemical Engineering with Biotechnology. She has involved in the seaweed project under ABI R&D Initiative grant and developed her interest in proteomics research.

## Technical Talk 2

### **Promarker™: A Comprehensive Mass Spectrometry Based Biomarker Discovery and Validation Platform as Applied to Diabetic Kidney Disease**

**Scott D. Bringans<sup>1\*</sup>**, Kirsten Peters<sup>1,2</sup>, Wendy A. Davis<sup>2</sup>, Timothy M.E. Davis<sup>2</sup> & Richard J. Lipscombe<sup>1</sup>

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A protein biomarker discovery platform, termed Promarker™, was applied to plasma samples from patients at different stages of diabetic kidney disease. The proteomics platform produced a panel of significant plasma biomarkers that were statistically scrutinised against the current gold standard tests. Biomarkers were significantly associated with diabetic kidney disease defined by albuminuria (ACR) and renal impairment (eGFR). From following a large clinical cohort over 4 years a predictive test called PromarkerD was developed from the biomarker panel and simple clinical parameters that could accurately predict 95% of otherwise healthy diabetics who would go on to develop chronic kidney disease. The results prove the suitability and efficacy of the Promarker™ discovery platform, and introduce a novel predictive test for diabetic kidney disease.

**Keywords:** Biomarkers; Diagnostic; Proteomics

#### **Biography**



Dr Bringans completed his PhD in 2001 from the University of Canterbury with a postdoctoral Fellowship at the National Cancer Institute, USA, studying bioactive proteins. He is currently Research Manager at Proteomics International involving oversight of Biomarker discovery and development projects encompassing PromarkerD, PI's predictive test for diabetic nephropathy.

## Talk 4

### Comparative Proteomic Analysis of *Ganoderma* Species During *In Vitro* Interaction with Oil Palm Root

Siti Nahdatul Isnaini Said Hussin<sup>1,2\*</sup>, Jameel R. Al-Obaidi<sup>1</sup>, Noor Baity Saidi<sup>2</sup>, Idris Abu Seman<sup>3</sup>, & Noor Azmi Shaharuddin<sup>2</sup>

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*Ganoderma* spp. is the biggest threat in most oil palm plantations around the world. In this study, response of the fungus at the molecular level during interaction with oil palm was investigated. Changes in protein expression of two *Ganoderma* spp., namely *G. boninense* and *G. tornatum* during in-vitro interaction with oil palm root was investigated. Phenol/ammonium acetate in methanol was shown to be the most effective protein extraction method for 2-DE proteomic studies of *Ganoderma* spp. mycelia. Attachment and colonization of both species on the oil palm root surface after 72 h of inoculation was confirmed by Scanning Electron Microscope (SEM) images. Comparative proteomic analysis showed that mycelial proteins from oil palm root exhibited different expression profiles when compared to the mycelia grown on Potato Dextrose Agar (PDA). Proteins were also expressed differentially in both species that may have either direct or indirect link to pathogenicity. The identified proteins may have possible roles in virulence and pathogenicity, metabolism, growth and maintenance. Identification of these proteins during the interaction with the oil palm root may provide a fundamental for further investigation on specific roles of the identified proteins towards *Ganoderma* infection mechanism as well as potential markers for early detection of *Ganoderma* disease.

**Keywords:** Comparative proteomic; *Ganoderma boninense*; *Ganoderma tornatum*; MALDI-TOF/TOF

#### Biography



Siti Nahdatul Isnaini Binti Said Hussin completed her first degree in B. Eng. (Biochemical-Biotechnology) from International Islamic University Malaysia (IIUM). She is currently a scientist in Plant Biotechnology Division of Agro-Biotechnology Institute (ABI) with 8 years' experience in algae tissue culture and 4 years' experience and publications in proteomics area.

## Talk 5

### Comparative Proteomic Analysis of Oil Palm Roots in Response to *In Vitro* Inoculation with *Ganoderma boninense* & *Ganoderma tornatum*

Norasfaliza Rahmad<sup>1,2\*</sup>, Jameel R. Al-Obaidi<sup>1</sup>, Noor Azmi Shaharuddin<sup>3</sup>, Idris Abu Seman<sup>4</sup>, Noor Baity Saidi<sup>2</sup>

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Basal stem rot (BSR) is an aggressive disease in oil palm caused by basidiomycete fungus known as *Ganoderma*. The disease is considered the most serious disease affecting the commercial oil palm plantations of South East Asia by reducing the productivity of the palm oil plant. BSR causes interruption in the allocation of water and nutrient from the root to the other parts of plant. To understand the mechanism involved in the early stage of interaction between *Ganoderma* spp. and its host, proteomic analysis of oil palm roots was conducted on protein samples collected at 120 hours post-inoculation with pathogenic *Ganoderma boninense* and non-pathogenic *Ganoderma tornatum*. Thirty-six differentially expressed proteins were successfully identified by mass spectrophotometry (MALDI TOF/TOF) in response to *Ganoderma* spp. inoculations. These proteins are mainly involved in signaling, stress/defense response and energy and lignin biosynthesis. Selected proteins with important role defense mechanism such as ascorbate peroxidase, malate dehydrogenase and PR10 proteins were verified using real-time PCR. The results obtained from this study provide candidate proteins that might be linked to the molecular pathways involved during interaction between oil palm and *Ganoderma* spp and might be essential for the future disease management program in oil palm plantation.

**Keywords:** Basal stem rot; *G. boninense*; *G. tornatum*; Proteomic; Root

#### Biography



Norasfaliza Rahmad has completed her Bachelor Degree in Biochemistry in 2008 from Universiti Putra Malaysia. Currently, she pursued her education for Master Degree in Plant Biotechnology in UPM. She has published several papers in various journals. Her interest is in plant and pathogen interaction study.



## Keynote 2

### Protein Degradation and Synthesis Rates in Leaf Growth and Development to Understand Energy Use and the Maintenance of Enzyme Function

**A. Harvey Millar\***

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Proteome studies focus almost exclusively on measuring abundance of proteins and documenting the fact that abundance changes in specific circumstances. This requires detection of statistically significant changes in the protein pool sizes to show that 'something has occurred'. Protein abundance data are then sandwiched in systems biology models as a layer between transcript responses and metabolite levels. Analyzing protein synthesis and degradation rates with progressive stable isotope labelling provides a new window into the control of protein abundance and the energy expended in maintaining the steady-state proteome across genotypes, development and environments. It provides the first and second derivative of protein abundance with respect to time: how fast are proteins turning over to achieve steady-state or gaining or lowering abundances and do these speeds differ in response to development or the environment? This approach can also enable the relative age distribution of a protein population to be assessed. This has implications for the energetic effort employed by the cell to build or maintain a particular activity and gives clues to the impact of age on the function in different protein types. We are using progressive  $^{15}\text{N}$  labelling of Arabidopsis to provide a birds-eye view of the activity of the proteolysis network as it maintains and sculpts the plant proteome. Using peptide mass spectrometry, the progressive labelling of new peptides and the decrease in the abundance of peptides with natural isotope profiles enabled the degradation rate of 1,228 leaf proteins to be determined by combining over 60,000 peptide relative isotope abundance (RIA) measurements. The exponential constant of the decay rate ( $K_D$ ) for each protein during growth showed a wide distribution, ranging from 0 to 2 per day, which was equivalent to protein half-lives of several hours to several months. We are also using this approach to dissect the in vivo action of proteases through analysis of knockout mutants. We have found new rapidly degrading subunits in a variety of protein complexes, identified the set of plant proteins whose degradation rate correlated positively or negatively with leaf growth rate, calculated the protein turnover energy costs for different leaves and their key determinants within the proteome, and are beginning to interpret transcriptome analyses from the point-of-view of maintenance of the proteome.

#### Biography



Prof. A. Harvey Millar completed his PhD in 1997 from The Australian National University and has held research fellowships at the University of Oxford and the University of Western Australia. He is currently the Director of the ARC Centre of Excellence in Plant Energy Biology, a premier plant proteomics research organization in Australia. He has published more than 220 papers in reputable journals and serves on the editorial boards of The Plant Cell, Plant Methods and The Biochemical Journal.

## Plenary 5

### Quantitative Proteomic Atlas Uncovering the Widespread Ubiquitylation Regulation in the Embryo of Germinating Rice Seed

Dongli He, Ming Li, Rebecca Njeri Damaris, and **Pingfang Yang\***

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Seed germination is a complex physiological process which is mainly regulated at post-transcriptional level, particular in proteome status modulation. Protein ubiquitylation plays an important role in almost all aspects of plant growth and development. In this study, we found that interference the ubiquitylation cycle with deubiquitylating enzyme inhibitor resulted in the delayed seed germination, showing the potential role of protein ubiquitination in seed germination. Using high affinity K-ε-GG antibody integrating highly sensitive MS, we analyzed the quantitative atlas of ubiquitylome as well as the proteome in the embryos of germinated rice seed. A list of 2576 lysine ubiquitinated sites in 1171 proteins were identified in the first 0, 12 and 24 hours after seed imbibition (HAI). The ubiquitin were more likely to attach to the lysine sites that locate in the polar acidic other than basophilic regions. Of these modified proteins, 1435 sites in 781 proteins were significantly changed in abundance (folds change > 2,  $P < 0.05$ ), most ubiquitinated sites were increased rapidly in the first 12 HAI, in general, the ubiquitylome dynamic were not significantly correlated to the global proteome, indicating the ubiquitylation played diversified regulatory roles in seed germination initiation. In addition, protein ubiquitylation was found overlapped with other PTMs in germinating rice seed, such as phosphorylation, carbonylation and acetylation, suggesting the potential crosstalk in co-regulatory events in this process. Taken together, our results exhibit that ubiquitylation widely involving in various intracellular processes and provides a rich quantitative dataset to further explore individual target roles in regulation of initial seed germination.

**Keywords:** Rice; Seed germination; Ubiquitylome

#### Biography



Prof. Pingfang Yang obtained his PhD in Plant Proteomics from Institute of Botany, Chinese Academy of Sciences in 2006. After that, he did his Postdoc training in Hong Kong University of Science and technology and Michigan State University. He is now a Professor in Wuhan Botanical Garden, Chinese Academy of Sciences since 2009. His research interests include proteomics on seed germination, lotus genomics, genetics and breeding. He is a Council member for Asia Oceania Agricultural Proteomics Organization (AOAPO), Chinese Society of Seed Biology, as well as editor for PloS one and Journal of Plant Science. He has published over 40 papers in different SCI journals, with a 17 H-index.

## Plenary 6

### Na<sub>2</sub>CO<sub>3</sub>-responsive Photosynthetic and ROS Scavenging Mechanisms In Chloroplasts of Alkaligrass Revealed by Phosphoproteomics

Shaojun Dai<sup>1,2,\*</sup>, Jinwei Suo<sup>2</sup>, Heng Zhang<sup>2</sup>, Qi Zhao<sup>1</sup>, Quanxi Wang<sup>1</sup>, Sixue Chen<sup>3</sup>

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Alkali-salinity exerts severe osmotic, ionic, and high-pH stresses to plants. Plant photosynthetic machinery is especially sensitive to saline-alkali stress. In this study, the photosynthetic characteristics, chloroplast ultrastructure, and reactive oxygen species (ROS) scavenging mechanisms were analyzed in halophyte alkaligrass (*Puccinellia tenuiflora*) under Na<sub>2</sub>CO<sub>3</sub> stress. Importantly, we found 102 proteins and 84 phosphoproteins in chloroplasts in response to Na<sub>2</sub>CO<sub>3</sub> treatment using isobaric tags for relative and absolute quantification (iTRAQ) and stable isotope dimethyl labeling proteomic approaches. Moreover, the expression level of 28 homologous genes of the chloroplast phosphoproteins were evaluated by quantitative real-time PCR (qRT-PCR) analysis. The abundance and post-translational modification (PTM) patterns of these proteins/genes highlight the Na<sub>2</sub>CO<sub>3</sub>-responsive chloroplast structure modulation and a series of photosynthetic regulatory mechanisms, such as thermal dissipation, state transition, cyclic electron transport, photorespiration, repair of photodamaged PSII, alteration of PSI activity, and ROS homeostasis. Especially, more than 56 amino acid sites were found as newly phosphosites in 56 Na<sub>2</sub>CO<sub>3</sub>-responsive phosphoproteins in chloroplasts, which are crucial for the regulation of photosynthesis, ion transport, signaling transduction, and energy homeostasis. All these data at the level of gene expression, protein accumulation, and PTM have improved our understanding of the Na<sub>2</sub>CO<sub>3</sub>-responsive molecular mechanisms of photosynthesis, ROS homeostasis, nuclear/ chloroplastic gene expression regulation, as well as protein PTM and transport in halophytes.

**Keywords:** Alkaligrass; Chloroplasts; Na<sub>2</sub>CO<sub>3</sub> stress; Phosphoproteomics

#### Biography



Professor Shaojun Dai obtained his PhD degree in Northeast Forestry University, China in 2002. He received his postdoctoral training in the Institute of Botany, Chinese Academy of Sciences in 2006 and in the University of Florida in 2009. Currently, he is a professor at the Shanghai Normal University. Professor Dai is the secretary general of Asia Ocean Agricultural Proteomics Organization (AOAPO). He is also a proteomics committee member of China Human Proteomics Organization (CNHUPO). Professor Dai's laboratory mainly focuses on the functional proteomics of plant in response to stress.

## Talk 6

### Identification of Proteins in Pitcher Fluid of *Nepenthes* Species through Proteomics Informed by Transcriptomics Approach

**Hoe-Han Goh\***, Muhammad Mu'izzuddin Zulkapli & Wan Mohd Aizat

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Tropical pitcher plants, *Nepenthes* spp., are among the most species-rich carnivorous plants which acquire nutrient from insects via passive trapping organ known as pitcher. The unique pitcher secretes acidic fluid containing various enzymes for the digestion of trapped insects. It also functions in the uptake of nutrient. Past studies have reported enzymes such as aspartic proteases (nepenthesin I and II), and pathogenesis-related proteins ( $\beta$ -1, 3-glucanase, class IV chitinase, and thaumatin-like protein). In this study, we investigate the protein composition in the pitcher fluid of three *Nepenthes* species with different feeding habits. *N. ampullaria* is a detritivore which can utilize leaf litter, whereas *N. rafflesiana* is a true carnivore which feed on insects, and their natural hybrid, *N. x hookeriana* which is also a carnivore. We adopted PacBio isoform sequencing (Iso-Seq) to generate full-length transcriptome reference sequences from pitcher tissues to enable protein identification from pitcher fluid via proteomics informed by transcriptomics (PIT) approach. The list of new proteins identified in the pitcher fluid and their relevance to the physiology of pitcher function will be discussed during the presentation.

**Keywords:** Carnivorous plants; Enzyme; *Nepenthes* sp.; Nepenthesin; Pitcher

#### Biography



Dr Hoe-Han Goh, a plant molecular biologist, graduated from the University of Sheffield, UK in 2011. He is currently a Head of Centre for Bioinformatics Research at the Institute of Systems Biology, National University of Malaysia. He has established a Plant Functional Genomics Research Group (gohlab.weebly.com) focusing on crop improvement and molecular exploration of tropical plant species using NGS and functional genomics approaches.

## Talk 7

### Proteome-wide Response of *Aquilaria* Tree to Agarwood Induction Treatments

Shiou Yih Lee<sup>1</sup>, Muhd Syahmi Hishamuddin<sup>1</sup>, Nurulfiza Mat Isa<sup>2</sup> Dhilia Udie Lamasudin<sup>2</sup>, Rozi Mohamed<sup>1,\*</sup>

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*Aquilaria malaccensis* Lam. (Thymelaeaceae) is an endangered tree species endemic to Malaysia. Upon wounding and pathogenic infection, the tree produces the fragrant resin known as agarwood or *gaharu*. Agarwood acts as a physical barrier to pathogens, but is also rich in secondary metabolites. For this reason, agarwood became a highly valuable raw material in manufacturing incense, perfumes and medicines. Currently, more than 1.2 million *Aquilaria* trees have been planted in Peninsular Malaysia. Mechanical wounding and use of modern inducers are among the common treatments applied to artificially induce agarwood. Gene regulation controlling agarwood formation has been revealed recently, however, the same at proteome-wide level is not known. Here, we report the first proteome from *Aquilaria malaccensis* profiled using liquid chromatography-mass spectrometry (LC-MS). Our preliminary analysis identified a total of 400 proteins across two treatments and control plant. Among them, 68 were specific to wounding, 85 to a formulated inducer, while 9 were shared. This study demonstrates the potential of proteomics approach in identifying important proteins controlling agarwood formation especially those involving with specific induction method.

**Keywords:** Gaharu; LC-MS; Inducer; Wounding

#### Biography



Dr. Shiou Yih Lee received his PhD in 2016 from Universiti Putra Malaysia. He specializes in plant molecular and systematics, forest management, wood forensic and plant proteomics. His main research focuses on agarwood and medicinal plants. Currently, he is a postdoctoral researcher with the Faculty of Forestry, UPM.

## Talk 8

### Comparing Protein Abundance and mRNA Expression Level on Jojoba (*Simmondsia chinensis*) Male and Female Individual Plant

Jameel R. Al-Obaidi<sup>1,\*</sup>, Nursyuhaida Mohd Hanafi<sup>1</sup>, Norasfaliza Rahmad<sup>1</sup>, Mohammed Farouq Halabi<sup>2</sup>, Abdul Rahman A. Al-Soqeer<sup>3</sup>

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Jojoba (*Simmondsia chinensis*) is a small waxy-leaved shrub that been cultivated over the world. Female plants are more important than the male plants as seeds produced by female plants contains the liquid wax. Since population of jojoba is male biased, identification of female plants are vital to the industry. Development of protein molecular markers for early gender differentiation revealed 18 known proteins. Those identified proteins were involved in photosynthesis, energy metabolism and response towards biotic and abiotic stress. Rubisco and ATP synthase were up-regulated in male compared to female, hence, proceed with mRNA expression validation. Result showed, Rubisco was up-regulated in female compared to male while ATP synthase was up-regulated in male compared to female. Correlations between the level of mRNA and level of protein expression is different due to several factors such as effect of post-transcriptional mechanisms that not yet sufficiently well-defined and amount of error/noise produced has limit the ability to understand the whole process. However, both proteins have the potential to serve as protein biomarkers for early differentiation between male and female individuals. The results presented in this study provided valuable resources in understanding the molecular mechanism of gender differentiation in jojoba.

**Keywords:** Biomarkers; Gene Expression; Rubisco

#### Biography



Nursyuhaida has completed her BSc in 2007 from Universiti Kebangsaan Malaysia and now pursuing Master in System Biology from the same university. She is currently working as a research officer at Agro-Biotechnology Institute, Malaysia since 2008. She has published several presentation and papers over the time and involved in few research projects related to molecular biology of plant. Among of the research involved are fingerprinting on *Jatropha curcas*, transcriptomic of red seaweed,

*Kappaphycus alvarezii*.



## Talk 9

### Sub-Zero Acclimation Induces Enhancement of Freezing Tolerance Accompanied by Changes of Extracellular Proteome and Cell Wall Characteristics in *Arabidopsis*

Daisuke Takahashi<sup>1\*</sup>, Michal Gorka<sup>1</sup>, Alexander Erban<sup>1</sup>, Alexander Graf<sup>1</sup>, Joachim Kopka<sup>1</sup>, Ellen Zuther<sup>1</sup> & Dirk K. Hincha<sup>1</sup>

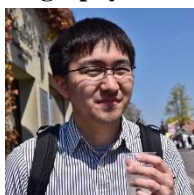
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Although non-freezing low temperature results in enhancement of plant freezing tolerance (cold acclimation, CA), further enhancement of freezing tolerance is induced by sub-zero temperature after CA (sub-zero acclimation, SZA). The response to extracellular freezing is thought to determine plant survival. However, changes in the extracellular matrix (ECM) induced by CA and SZA are not yet fully characterized. We therefore investigated responses of *Arabidopsis* ECM to CA and SZA from the aspects of cell wall composition analysis and proteomics. The amounts of cell wall material in *Arabidopsis* leaves increased in response to CA and was maintained during SZA treatment. FTIR and GC-MS analysis clearly showed that cell wall structure and monosaccharide compositions responded to SZA. From ECM proteomics, 11.9% and 11.6% of ECM proteins were specifically up- and down-regulated during SZA, respectively (e.g. pectin methylesterases, xyloglucanases, O-glycosyl hydrolases). Furthermore, knock-down of O-glycosyl hydrolase gene *At3g04010* resulted in impaired freezing tolerance during SZA accompanied by excess accumulation of callose. Taken together, the relationship between molecular changes in the ECM and enhancement of freezing tolerance during SZA will be discussed. This study was partly supported by a JSPS Fellowship for Research Abroad (#27-328), the Alexander von Humboldt Foundation and the Max-Planck Society.

**Keywords:** *Arabidopsis*; Cold acclimation; Extracellular matrix; Freezing tolerance; Sub-zero acclimation

#### Biography



Dr Daisuke Takahashi has completed his PhD in 2015 from Iwate University. He got a postdoctoral position at Max-Planck-Institut für Molekulare Pflanzenphysiologie (MPI-MP) as JSPS (Japan Society for the Promotion of Science) fellow. He is currently supported by the Alexander von Humboldt Research Fellowship. The Botanical Society of Japan is going to award Young Botanist Prize to him on September 2017.

## Plenary 7

### The Omics for Agriculture and Biodiversity

#### Mohd Hasnain Hussain\*

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A combined approach of discovery through genomics, proteomics, and other associated -omic branches of research can be effective for knowledge discovery and used as biotechnological tools in agriculture related field. Here, we narrate our experience using omics approach in UNIMAS, related agriculture and biodiversity. *Piper nigrum* known as pepper plant are commercially exported and one of main contributor towards Sarawak economy. However, some diseases had affected this plant causing loss to farmers. Differential proteomic analysis is used to compare protein profiles of these plants using 2-D PAGE which helps to study the function of proteins in diseased plants. Diseased plants had shown some unique protein spots in both stems and roots samples, thus has potential to identify defence mechanism and other biological processes. In another important commodity that contributes to the economy of Malaysia, sago palm (*Metroxylon sagu*), we are investigating the reasons for non-trunking of this plant. In some areas of deep peat soil of sago plantation in Sarawak, the palm remains at rosette stage even after 17 years of plantation. This set back of sago palm growth reduced the starch storage capacity of the palm, thus eliminating the economic value of the plant. Analysis of proteins from sago palm indicated protein of interests, upon analysis showed different function in the biological processes. In rice, we compare the proteomes of the amyloplasts of *sugary* and normal rice, with the aim of discovering differentially expressed proteins which may be involved in starch biosynthesis. Using proteomics approach, we attempt to understand how the actions of these biosynthetic enzymes are coordinated to produce amylopectin with specific chain length distribution that can crystallize and produce starch granules. Most current studies are limited to enzymes known to be involved in starch biosynthesis and do not involve a proteome-scale approach. Related to biodiversity, we are attempting to identify peptide-based compounds from natural resources for use as potential antitumor and antimicrobial agents. Results from our current work show that peptides extracted from two plants have a potential antitumor and antimicrobial activities.

**Keywords:** *Metroxylon sagu*; *Piper nigrum*; Proteome; Rice amyloplasts; Sago

#### Biography



Assoc. Prof. Dr Mohd Hasnain Hussain is the Director, Centre for Sago Research in Universiti Malaysia Sarawak (UNIMAS). He was a former Dean of Faculty of Resource Science and Technology, UNIMAS and prior to that he has held other administrative positions at the faculty such as Deputy Dean and Program Coordinator at the Dept. of Molecular Biology. He obtained MSc from University of Newcastle upon Tyne, UK in Agricultural Biotechnology followed by PhD in Plant Molecular Biology from University of East Anglia UK. His main research interests are in elucidation of enzymes and their interactions during starch biosynthesis in plants. He is currently pursuing metabolomics and proteomics of sago palm and other plants such as rice, pepper and local indigenous plants in Borneo used as therapeutic agents.



## Plenary 8

### Barley and *Piriformospora indica* Relationship: What Has an Integrative Omics Approach Taught Us?

Ghasem Hosseini Salekdeh and Reza Ghaffari

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Plant beneficial microbes have a clear positive impact on the productivity and quality of crops. A better understanding of their impact will address a number of both food sustainability and security issues. The root endophyte *Piriformospora indica* is known to promote plant production under abiotic stresses. We have applied a combination of proteomic, transcriptomics, epigenomics, metabolomic and ionomic analyses to study the drought- and salt-stressed responses of barley plants colonized by *P. indica*. An integrative analysis of data highlighted molecular mechanisms underlying enhanced stress tolerance induced by *P. indica* colonization. Furthermore, a correlation-based network analysis revealed a set of proteins, miRNA, RNAs and metabolites strongly correlated to improved ability of plants to withstand unfavorable environmental conditions. We will discuss how *P. indica* may contribute to barley adaptation to abiotic stresses.

**Keywords:** Endophyte; Genomics; Proteomics; Plant-endophyte interactions; Symbiosis

#### Biography



Dr. Salekdeh Joint Agricultural Biotechnology Research Institute of Iran after received his PhD from International Rice Research Institute in 2002. His researches focus on discovering novel pathways and genes involved in crop response to biotic and abiotic stresses. He is council member of AOHUPO and HUPO. On a national level, Dr. Salekdeh is Cofounder of Iranian Proteomics Society and President elect of society since 2004. He is on a number of editorial boards including Proteomics Journal, Journal of Proteome Research, Frontiers in Plant Science and Nature Scientific Reports. He received several awards and honors including National Biotechnology Award (2007), National Razi Medical Science award for advance technologies (2009), the Khwarizmi International Award for fundamental research (2010), Hadavi award from Iranian Academy of Medical Sciences (2010 and 2014), Distinguished Scientist in Biotechnology (2013), and Best National Researcher (2015), and Distinguished Scientist in Genetics (2016). He has published over 150 peer-reviewed international journals and has edited two international books.

## Talk 10

### The Proteomic Landscape of Soybean Responses against Flooding Stress: Insights from Post-Translational Modifications

**Akiko Hashiguchi\***, Setsuko Komatsu

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Flooding is the major environmental stress for soybean which brings about severe growth inhibition. Previous studies carried out analyses on post-translational modifications (PTMs) such as phosphorylation, ubiquitination, and glycosylation, substantiating understanding of underlying mechanisms of soybean responses to flooding stress. However, global overview of the response and the relationship between the PTMs have not been fully elucidated. The shift from aerobic to anaerobic respiration is crucial for responses to flooding stress although its regulatory mechanism at initial stage is not clear. Here, another analysis was performed focusing on protein S-nitrosylation-mediated regulation of energy production. Among diverse metabolic pathways that were influenced at initial stage of flooding by protein S-nitrosylation, development and fermentation categories showed remarkable increase in protein S-nitrosylation status. Increase in the protein S-nitrosylated status of alcohol dehydrogenase was confirmed, suggesting a distinctive role of protein S-nitrosylation in modulation of energy production. Combined with previous reports on other PTMs, current results add a touch on comprehensive picture of mechanism of soybean's adaptation to flooding stress.

**Keywords:** Energy production; Flooding stress; *Glycine max*; Post-translational modification; Protein homeostasis

#### Biography



Dr Hashiguchi has obtained her PhD in 2007 from The University of Tokyo and experienced postdoctoral training at National Institute of Crop Science, Japan. Her current research interests at Tsukuba University, Japan are proteomic elucidation of soybean responses against flooding stress and application of proteomics for evaluating health-promoting property of medicinal plants. She has published 13 papers in reputable journals.

## Talk 11

### Quantitative Proteomic Analysis of Soybean Mitochondrion on Exposure to Varying Sizes of Aluminum Oxide Nanoparticles Under Flooding Stress

**Ghazala Mustafa**<sup>1,2\*</sup>, Setsuko Komatsu<sup>1</sup>

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Rapid developments in nanotechnology have led to the increasing use of nanoparticles (NPs) in agricultural sector. To investigate the possible interactions between NPs and crops under flooding stress, the molecular mechanisms in soybean affected by various sizes of Al<sub>2</sub>O<sub>3</sub>-NPs were analyzed using a proteomic technique. In plants exposed to 30-60 nm Al<sub>2</sub>O<sub>3</sub>-NPs, length of root including the hypocotyl was increased and proteins related to glycolysis were suppressed. Exposure to 30-60 nm Al<sub>2</sub>O<sub>3</sub>-NPs mediated the scavenging activity of cells by regulating the ascorbate/glutathione pathway. Ribosomal proteins were also increased on exposure to flooding-stressed plants with 30-60 nm Al<sub>2</sub>O<sub>3</sub>-NPs. Mitochondrion was the target organelle of Al<sub>2</sub>O<sub>3</sub>-NPs under flooding stress. Mitochondrial proteomic analysis revealed that the abundance of voltage-dependent-anion-channel protein was increased on exposure to flooding-stress with 135 nm Al<sub>2</sub>O<sub>3</sub>-NPs, indicating the permeability of mitochondrial membrane was increased. Furthermore, isocitrate dehydrogenase was increased on exposure to 5 nm Al<sub>2</sub>O<sub>3</sub>-NPs under flooding stress. These results suggest that Al<sub>2</sub>O<sub>3</sub>-NPs of various sizes affect mitochondrial proteins under flooding by regulating membrane permeability and tricarboxylic acid cycle activity.

**Keywords:** Aluminum oxide nanoparticles; Flooding stress; Mitochondrion; Proteomics; Soybean

#### Biography



Dr Ghazala Mustafa has completed her PhD in 2016 from University of Tsukuba, Japan. She is currently working as an Assistant Professor in the Department of Plant Sciences, Quaid-i-Azam University, Islamabad, Pakistan. She has published more than 15 publications in reputable journals and serving as an editorial board member of Journal of Open Proteomics.

## Talk 12

### Soybean Proteomics: A Leading Tool in Exploring Effects of Flooding Stress Using Gel-based Proteomic Technique

Amana Khatoon<sup>1</sup> \*, Shafiq Rehman<sup>1</sup>, Muhammad Jamil <sup>2</sup> & Setsuko Komatsu<sup>3</sup>

<sup>1</sup>Department of Botany, Kohat University of Science and Technology, Kohat 26000, Pakistan; <sup>2</sup>Department of Biotechnology and Genetic Engineering, Kohat University of Science and Technology, Kohat 26000, Pakistan; <sup>3</sup>Faculty of Life and Environmental Sciences, University of Tsukuba, Tsukuba 305-8572, Japan.

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Among legumes, soybean ranks high in susceptibility to flooding stress. With respect to high importance of soybean production worldwide and an increasing threat of flooding stress towards its yield, its production must be evaluated against flooding stress. To understand soybean protein networks regulating responses towards flooding stress, 2-DE based proteomic technique was used. Among various mechanisms adapted by soybean against flooding stress, regulation of carbon metabolism to meet cells energy requirements to cope with the stress is common in all cases. Soybean was affected more severely by flooding stress compared to low oxygen stress, indicating that protein destination/storage and disease/defense related proteins were significantly changed in flooded seedlings leading to more growth suppression. Decreased isoflavone reductase in root, shoot and leaf, and upregulation of its gene at transcript level, clearly indicated that flooding stress interfered at translation level. Inoculation of soybean with *Bradyrhizobium japonicum* resulted in regulation of disease/defense and metabolism-related proteins causing increased number of root hair during early symbiotic differentiation. Proteomic results also clarified that recovery after flooding may involve alteration of cell structure through changes in cell wall metabolism and cytoskeletal organization in soybean seedlings. These results suggest that proteomics provide a reliable method to unravel the regulation of complex protein network involved in soybean response towards flooding stress.

**Keywords:** 2-DE based proteomics; Flooding; Soybean

#### Biography



Dr Khatoon has completed her PhD in 2014. She did her PhD research work under supervision of Prof. Komatsu in National Institute of Crop Sciences, Japan. She is currently teaching in Kohat University and has established first proteomics research laboratory in her university. She has published more than 12 papers in reputable journals.

## Talk 13

### Drought Stress Characterization of Tolerant and Sensitive Barley Genotypes Through Proteomics Analysis

**Rehana Kausar**<sup>1,\*</sup>, Muhammad Arshad<sup>2</sup>, Armghan Shahzad<sup>3</sup>, Setsuko Komatsu<sup>4</sup>

<sup>1</sup> Department of Botany, University of Azad Jammu & Kashmir, Muzaffarabad, 13100, Azad Kashmir; <sup>2</sup> Department of Botany, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi 46300, Pakistan; <sup>3</sup> Plant Biotechnology Program, National Agriculture Research Center, Islamabad 45500, Pakistan; <sup>4</sup> Graduate School of Life and Environmental Science, University of Tsukuba, Tsukuba 305-8577, Japan

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Plant productivity is severely impaired by drought stress and it is an important factor limiting barley yield as well. Proteomics technique was used to investigate the initial response of barley under drought stress. Three-day-old barley seedlings of tolerant genotype 004223 and sensitive genotype 004186, were treated with 20% polyethylene glycol and by withholding water to impose drought. Proteins were extracted after three days of treatments, separated by two-dimensional polyacrylamide gel electrophoresis and stained with Coomassie brilliant blue. In response to drought among the common proteins between tolerant and sensitive genotype, Photosystem I reaction centre II was decreased and Vacuolar proton ATPase subunit E was increased. In the tolerant group, many of the Metabolism related proteins were increased under drought stress, however such type of proteins were decreased in sensitive genotypes. These results indicate that chloroplastic metabolism and energy related proteins might play a significant role in the adaptation process of barley seedlings under drought stress.

**Keywords:** Barley; Drought; Proteomics; Sensitive genotype; Tolerant genotype

#### Biography



Dr Rehana Kausar has completed her PhD in 2015 from PMAS-Arid Agriculture University, Rawalpindi, Pakistan. She is currently working as Assistant Professor in Department of Botany, UAJK, Pakistan. She has published 10 research papers in journals of International repute and author of two books as well. She is the life time member and Vice President (AJK) chapter of Pakistan Botanical Society.

## Keynote 3

### From Proteomic Analysis to Functional Analysis

**Fook Tim Chew\***

*Department of Biological Sciences, National University of Singapore (NUS), Lee Hiok Kwee Functional Genomics Laboratory Cluster, Block S2, Level 5, Science Drive 4, Singapore*

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Allergic conditions such as Asthma, Allergic Rhinitis, and Atopic Dermatitis (AD) are heterogeneous diseases influenced by multiple underlying pathogenesis as well as diverse environmental factors. The development of the diseases involves a complex immunologic cascade inter-playing with the disruption of the epidermal skin barrier, IgE dysregulation, defects in the cutaneous cell-mediated and innate immune functions, neurological and possibly many other genetic factors not yet well defined. The exact pathogenesis or etiology is complex, and it is likely that these allergic diseases are a continuum of syndromes where different patients (although presenting with similar clinical manifestations - e.g., flexural rash, persistent itch, erythema, scaling, etc. for AD) may have different underlying dysregulation or pathogenesis (or combinations of them). I will highlight our large-scale epidemiological, proteomic and genomic studies encompassing more than 10,000 Chinese Singaporean individuals. In the case of AD, we have since discovered that the underlying disease continuum may be related to a certain extent to: (a) auto-allergic responses, (b) novel anti-microbial, and (c) anti-viral components, (d) epidermal wound-healing components in sweat and skin, (e) differential protease (and protease inhibitor) levels / activities, (f) levels of natural moisturizing factors and other metabolites, as well as (g) novel filaggrin null mutations, in tandem or combinations. Proteomic analysis of the skin stratum corneum, as well as the human sweat repertoire, provided much insights to the components that are present in the disease vs control states. Genetic variants controlling these functions play a major role in influencing the skin barrier function(s), and thereafter coupled with atopic disposition, lead to the manifestation of the disease. I would like to highlight the translation of large scale proteomic work coupled with meaningful functional characterization of the components can provide insights into the pathogenesis of the disease as well as become key biomarkers for disease progression and prognosis.

**Keywords:** Atopic Dermatitis, Skin Protein, Stratum corneum

#### Biography



Dr Fook Tim Chew is an Assoc. Prof. at the Dept. of Biological Sciences and the Vice Dean of Undergraduate Education and Student Life, at the Faculty of Science, NUS. Originally from Malaysia, and a graduate of UPM and NUS, he is now a Governing Board Member of SEARCA, Chairman of the Academic Advisory Board at MDIS, Scientific Board Member of several Technology Start-Ups, and Lead Scientific Consultant to several major Companies. He has published more than 150 Publications, several patents, and presented at over 300 international and regional conferences, with more than 150 invited talks. He won NUS Teaching Excellence and Outstanding University Researcher Awards. He was awarded the Singapore Youth Award in 2004.



## Plenary 9

### Cancer Biomarker Discovery: Lectin-based Proteomic Studies

**Onn Haji Hashim\***

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We have applied champedak galactose binding lectin in proteomic studies to identify low abundant glycoproteins that can be used as cancer biomarkers. Our lectin-based proteomic analysis of urine samples detected the aberrant expression of a truncated fragment of inter-alpha-trypsin inhibitor heavy chain 4, saposin B and bikunin in patients with prostate cancer compared to those with benign prostatic hyperplasia. Currently, diagnosis of prostate cancer is very much reliant on the levels of serum prostate specific antigen and trans-rectal ultrasound-guided biopsy of the prostate gland. In more than eighty percent of patients subjected to prostate biopsy, the procedure appears unnecessary as malignancy was ruled out since patients had benign prostatic hyperplasia. Hence, we have proposed the use of the urinary peptide biomarkers to discriminate patients with benign prostatic hyperplasia from those with prostate cancer so that they do not have to be subjected to the invasive and costly prostate biopsy procedure. In our breast cancer study, higher levels of proteoglycan 4 and lower levels of plasma protease C1 inhibitor were detected in sera of stage 0 and stage I patients compared to healthy control women. In view of the reciprocal trend of altered levels of proteoglycan 4 and plasma protease C1 inhibitor between breast cancer patients and controls, the two serum *O*-glycosylated proteins provide strong complementary biomarker candidates for screening of early breast cancer.

**Keywords:** biomarker, breast cancer, lectin, prostate cancer, proteomics

#### Biography



Onn Hashim received his PhD from the University of Glasgow in December 1987. He joined the University of Malaya in January 1988 and is currently a Professor at the Dept. of Molecular Medicine since 2003. Onn had spent short sabbaticals at the University of Alabama at Birmingham USA and University of Osaka Medical School Japan under sponsorships of the American Fulbright Scholarship and JSPS Fellowship, respectively. He was appointed as Visiting Professor at Prince of Songkla University, Thailand, in 2016. Onn is recipient of the National Academic Award 2007 and currently chairs the Selection Committee of the award. He is former Editor-in- Chief of the Malaysian Journal of Biochemistry and Molecular Biology, and presently member of the editorial board of Biomarker Research. He is Head of the University of Malaya Centre for Proteomics Research.

## Plenary 10

### Chemical Proteomics Investigation of Artemisinin's Antimalarial and Anticancer Mechanisms

Lin Qingsong\*

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Artemisinin and its derivatives are the most potent antimalarial drugs at present. To understand its mechanism of action in the malaria parasites, we generated an alkyne-tagged artemisinin analogue and used it to treat the malaria parasite *Plasmodium falciparum*. Upon coupling of biotin tags by click chemistry, followed by affinity enrichment and mass spectrometry, we identified over 100 artemisinin covalent-binding protein targets. Many of these targets are involved in essential biological processes of the parasite, thus artemisinin treatment disrupts the biochemical landscape of the parasite and causes its death. By coupling the artemisinin targets with a fluorescent tag, we have also shown that heme, rather than free ferrous iron, is predominantly responsible for artemisinin activation. The source of heme derives primarily from parasite's heme biosynthesis pathway at the early ring stage, and from hemoglobin digestion at the latter stages. Recent studies have shown that artemisinin also possesses anticancer activities. To understand its anticancer mechanism, we made use of alkyne- or biotin-tagged artemisinin probes to identify over 300 specific artemisinin targets, revealing its mechanism of action in killing cancer cells via promiscuous targeting of multiple critical biological pathways, similar to its action in malaria parasites. We also demonstrated that artemisinin specifically kills colorectal cancer (CRC) cells rather than normal colon epithelial cells, and the specificity may stem from the elevated capacity of heme synthesis in cancer cells, which results in higher level of artemisinin activation. Guide by this mechanism, we further increased the heme level in cancer cells by treating the cells with a clinically used heme synthesis precursor,  $\alpha$ -Aminolevulinic Acid (ALA), which dramatically enhanced the anticancer effects of artemisinin. This novel artemisinin/ALA combination therapy was proven effective with a mouse xenograft CRC model.

**Keywords:** Artemisinin; Cancer; Chemical Proteomics; Drug mechanism of action  
Malaria

#### Biography



Dr Lin Qingsong obtained his PhD in 2002 from University of Toronto, Canada (major in Clinical Biochemistry), currently he is a senior Research Fellow, in Department of Biological Sciences, and Co-director for Protein and Proteomics Centre NUS. His Research Interests in Proteomics and mass spectrometry, Cancer biomarker discovery and cancer biology, Chemical proteomics and drug target identification. He is a Member of editorial boards in Scientific Reports and Frontiers in Plant Science Journal. He is a member in many proteomics related organizations and societies. He has published over 80 peer-reviewed papers.



## Talk 14

### Proteomic Analysis to Reveal Molecular Mechanism of Phenolic Acids Accumulation in Leaves of *Salvia miltiorrhiza* by UV-B Radiation

Xiaojian Yin<sup>1\*</sup>, Shi-Lei Wang<sup>1</sup>, Pingfang Yang<sup>2</sup>, Setsuko Komatsu<sup>3</sup>, & Lian-Wen Qi<sup>1\*</sup>

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*Salvia miltiorrhiza* Bunge known as danshen, which mainly contains lipid-soluble tanshinones and water-soluble phenolic acids, is used as traditional Chinese medicinal material. To improve content of phenolic acids, danshen was treated with UV-B radiation in this study. As results, phenolic acids including salvianolic acid B, rosmarinic acid, and caftaric acid were improved more than 50 times in leaves of danshen under UV-B radiation for 4 h compared to untreated danshen. To reveal the molecular mechanism on accumulation of phenolic acids in danshen leaves by UV-B radiation, isobaric tags for relative and absolute quantitation (iTRAQ)-based proteomics was performed. Compared to untreated danshen, 96 proteins in leaves were significantly changed by 4 h of UV-B treatment. Among them, phenolic acids synthesis related key enzymes including cinnamoyl-CoA reductase, rosmarinic acid synthase, phenylalanine ammonia-lyase, CYP749 A39, CYP98 A78, and CYP716 C12 were increased in UV-B treated danshen. In addition, transcription factors such as basic helix-loop-helix transcription factor and NAC transcription factor were also significantly changed under UV-B radiation. These results suggest that UV-B radiation with short time can improve content of phenolic acids in danshen leaves through affecting phenolic acids synthesis related enzymes and transcriptional regulation might play important role in this process.

**Keywords:** Phenolic acids; Proteomics; *Salvia miltiorrhiza* Bunge; UV-B radiation

#### Biography



Dr Yin has completed his PhD in 2016 from the University of Tsukuba in the lab of Prof Komatsu. He is currently the associate researcher of China Pharmaceutical University, a premier Chinese medicinal plant proteomics research organization. He has published more than 11 papers in reputable proteomics related journals such as Journal of Proteome Research, Journal of Proteomics, and so on.

## Talk 15

### Proteomics Analysis of Kesum (*Persicaria minor* Huds.) Herbal Plant Upon Methyl Jasmonate Treatment

Wan Mohd Aizat\*, Sarah Ibrahim, Reyhaneh Rahnamaie-Tajadod, Kok-Keong Loke, Hoe-Han Goh, Normah Mohd Noor  
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Kesum (*Persicaria minor* Huds.) is commonly used in various traditional cuisine and medicine as it exhibits pungent smell as well as possessing antimicrobial and antioxidant activities. Previous studies have shown that this herbal species produced significant physiological changes during stresses including increasing its secondary metabolite production. One of the stress-response hormone that may regulate this is methyl jasmonate (MeJA) hormone. However, how it specifically regulates a non-model herbal species such as kesum have not been adequately reported. Hence, the aim of the study is to profile the proteome of kesum elicited with MeJA to elucidate the molecular regulation of this hormonal cue. We believe that this is the first proteomics study on such non-model species particularly using the label-free quantification of SWATH-MS approach. Both 1D and 2D-information dependant acquisition (IDA) followed by SWATH-MS were performed which successfully profiled a comprehensive proteome coverage of 751 proteins. Forty proteins were found to be significantly different between control and MeJA-treated samples. The modulated levels of these proteins suggest that the hormone invoked proteins related to defense and recovery response but suppressed proteins involved in growth and development.

**Keywords:** Label-free proteomics; LC-MS/MS; *Polygonum minus* herb; SWATH-MS analysis

#### Biography



Dr. Wan Mohd Aizat completed his PhD at the University of Adelaide, Australia. He is specialized in Plant Science field, utilizing various postharvest analyses and analytical techniques such as proteomics and metabolomics to study various plants. Currently, Dr. Aizat mainly focuses on mangosteen fruit to investigate its ripening process using both postharvest and molecular approaches.

## Talk 16

### Proteomic Analysis of BmNPV Resistance in the Silkworm Reared on UV-B Induced Mulberry Leaves by SWATH-MS

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Mulberry leaves are the main food for silkworm. And the secondary metabolites in mulberry leaves were improved after UV-B induction, particularly for moracin N. In the present study, we found that feeding on UV-B induced mulberry leaves and moracin N could enhance the BmNPV resistance in silkworm. To gain an insight into the mechanism of anti-BmNPV in silkworm at protein level, proteomic analyses of silkworm midguts from UV-B induced mulberry leaves and moracin N groups were performed by SWATH-MS. The results showed that the abundance of ribosomal proteins in UV-B induced mulberry leaves and moracin N groups was significantly changed to maintain the synthesis of total protein levels and cell survival. While, the expression of proteins involved in apoptotic process such as cytochrome c oxidase, calcium ATPase, and programmed cell death was up-regulated in silkworm feeding on moracin N groups. Taken together, these results suggest that moracin N might be the main active component in UV-B induced mulberry leaves which could improve the immunity of silkworm infected by BmNPV and inhibit the viral replication. It also presents an innovative process to reduce the mortality rate of silkworms infected with BmNPV.

**Keywords:** BmNPV resistance; Moracin N; Silkworm; SWATH proteomics

#### Biography



Dr Zhu has completed his PhD in 2016 from Zhejiang University and now is a postdoctoral researcher in Zhejiang University. He is majored in medicinal plant proteomics.

## P01

### Comparative Proteomic Analysis of *in vitro* Fungal-bacterial Interaction

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*Fusarium fujikuroi*, the causal agent of the bakanae disease of rice, is one of the major threats to rice productivity. Results from the last two decades have shown that various bacteria can inhibit the growth of this particular fungus and thus can be used as biocontrol agents. However, the underlying mechanism of this fungal-bacterial interaction is currently unknown. Therefore, this study was performed with an aim to analyze the proteins involved in fungal-bacterial interactions. Further, we also wanted to compare the antifungal activity of bacteria *in vitro* and *in planta*. In order to find promising biocontrol agents, we isolated *Bacillus* species bacteria from weed leaves and assayed their antifungal activities against *F. fujikuroi*. *B. amyloliquefaciens* showed remarkable antagonist effect against *F. fujikuroi* on solid media. Following this observation, we extracted the proteins of two microbes for proteome analysis in four combinations: only inoculation bacteria (Con\_B), only inoculation fungus (Con\_F), co-culturing bacteria (BF\_B), co-culturing fungus (BF\_F). After preparation of samples set, proteins were extracted using MgNP-40 extraction buffer followed by TCA/Acetone precipitation. SDS-PAGE analysis of extracted proteins showed clear differences in the protein pattern, and identification of differential proteins by mass spectrometry (MS) is underway.

**Keywords:** *Bacillus amyloliquefaciens*; Bakanae disease; Fungal-bacterial interactions; *Fusarium fujikuroi*; Proteome analysis

## P02

### Identification and Validation of Putative *Erwinia mallotivora* Effector Proteins through iTRAQ Protein and Real Time PCR Analysis

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*Erwinia mallotivora* which was phenotypically, biochemically and genotypically shown as the close relative of *E. papayae*, is the pathogen responsible for the devastating papaya dieback disease in Malaysia. Similar to other gram-negative plant pathogens, *E. mallotivora* depends on hypersensitive response and pathogenicity (hrp) genes or effectors to cause disease in susceptible papaya variety. To further understand the molecular mechanisms leading to the bacteria pathogenesis, *in vitro* protein profiling of *Erwinia mallotivora* for the identification of virulence proteins from papaya dieback pathogen were conducted using iTRAQ mass spectrometry following its growth in selected nutrient rich media and minimal media that stimulate the expression of targeted virulent proteins. Putative virulence related proteins with increased fold change expression which includes outer membrane proteins, flagellin proteins, Hrp family proteins, chorimate mutase, proteases and hydrolases were revealed. Validations of selected proteins were further carried out and confirmed by Real Time PCR analysis. The identified proteins may represent important proteins that contribute to the bacteria pathogenicity and virulency.

**Keywords:** *Erwinia mallotivora*; iTRAQ; Virulent factors

## P03

### Proteomic Profiling of Human Cervical Cancer Cells Treated with a Selected Diarylpentanoid

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Curcumin, an active component from the root of *Curcuma longa* has shown to exhibit significant growth-inhibitory effect on various cancers through modulation of biological pathways. Our preliminary data showed that the diarylpentanoid MS13 demonstrate significant growth-suppressive activity in HeLa cervical cancer cells at EC<sub>50</sub> value of 10µM following 48 hours of treatment. The aim of the present study was to determine differentially expressed proteins (DEPs) in HeLa cells treated with 10µM of MS13 for 48 hours. Protein expression profile was performed using two-dimensional gel electrophoresis and DEPs were identified by LC-MS/MS. Functional classification and pathway analysis were performed on the DEPs. We have identified highly significant (p<0.05) 24 up- and 8 down-regulated DEPs in response to MS13 treatment. Data analysis revealed that the up-regulated proteins were associated with structural molecule activity, protein folding, metabolic process, proteolysis, transcriptional regulation, cell communication, cell cycle and translational regulation, whereas the down-regulated proteins were associated with structural molecular activity, translational regulation, metabolic process and proteolysis. Pathways significantly modulated by MS13 were protein ubiquitination pathway, aldosterone signaling in epithelial cells and PRPP biosynthesis I. These findings suggest that MS13 may exhibit its growth-suppressive effect on cervical cancer cells through modulation of these proteins and pathways.

**Keywords:** Cervical cancer; Curcumin analogue; Proteomic profiling; Mass spectrometry

## P04

### **Liver Heme Oxygenase-1 Expression is Positively Induced by Palm Oil-Derived Tocotrienol Rich Fraction (TRF) Supplementation in Mice**

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The antioxidant activities of tocotrienols are more robust than tocopherols. Palm oil is a rich natural source of tocotrienols. Heme oxygenase-1 (HO-1) is an enzyme that possess antioxidant, anti-inflammatory and cytoprotective functions. The objective of this study is to determine the effects of different doses of TRF oral supplementation on HO-1 protein expression in mice livers. Thirty male ICR white mice (25–30 g) were divided into five groups; three groups were administered TRF orally for 14 days at doses of 200, 500 and 1000 mg/kg respectively (n=6 per group), a positive control group administered 100 mg/kg butylated hydroxyanisole (BHA) orally for 14 days (n=6), and a control group (n=6) where mice were only administered vehicle (corn oil). At day 15, mice were sacrificed and their livers isolated. Livers were then homogenized and HO-1 protein expression was determined by Western blotting. It was observed that TRF oral supplementation at concentrations of 200, 500 and 1000 mg/kg for 14 days resulted in significant concentration-dependent increase in HO-1 protein expression in mice livers, compared to controls. In conclusion, TRF supplementation induced HO-1 protein expression in mice liver dose dependently, with the highest expression seen in mice receiving 1000 mg/kg TRF.

**Keywords:** Heme oxygenase-1; Liver; Mice; Protein expression; TRF



## P05

### **Leucine-rich alpha-2-glycoprotein 1 (LRG1) in Colorectal Cancer: A Real Potential Biomarker?**

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Colorectal cancer (CRC) contributes to significant cancer-related deaths worldwide with an immense urgency for reliable biomarkers either for screening, monitoring or as prognostic factors. Finding the specific proteins overexpressed in CRC cells and concurrently available in the patients' blood is thought to be the best approach in CRC biomarker discovery. We aim to identify potential protein markers present in both human serum and CRC cells, and its role in CRC. Quantitative proteomics was performed using SWATH-MS analysis on 15 human serum samples representing 4 stages of CRC and normal control. Western Blot was conducted to confirm the expression of target proteins in cell line model followed by functional analysis. We have identified significant upregulation of LRG1 in the sera of CRC patients, particularly in Dukes' D. The endogenous level of LRG1 in human CRC cells increased with the advancing stage whereas present at very low level in the normal. Overexpression of LRG1 in HT29 caused the cells to grow in clusters with slight increment in migration but reduced in cell proliferation ( $p < 0.01$ ) as compared to the GFP-control cells. In conclusion, LRG1 is a promising marker for CRC and its overexpression may pose higher risk of metastasis via collective migration.

**Keywords:** Biomarker; Colorectal cancer; LRG1; Metastasis; Overexpression

## P06

### Protein Profiles Of LPS-Induced and RECA Treated in Rat Hippocampus

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Proteins are called the workhorses of life, taking part in essentially every structure and activity of life with a vast array of functions. Proteomics, via 2-Dimensional Electrophoresis (2DE) is a valuable tool for protein profiling since it has opportunity to diagnose the quantitative and qualitative difference in the protein samples. It is a good pathology investigative tool to study the neuroprotective activity of Bioactive Extract of *Centella asiatica* (BECA). Neuroprotection is the mechanism to protect against neuronal injury or degeneration in the Central Nervous System (CNS) including neurodegenerative diseases such as Alzheimer's or Multiple Sclerosis, stroke, traumatic brain injury, and spinal cord injury. In this study, the neuroprotective effect of BECA was investigated in LPS-induced rats. On day 20, rats were sacrificed and hippocampus samples from control (LPS-induced) and RECA treated were subjected to protein extraction. 2-DE profiling of the rat brain were performed to observe the changes in protein expressions towards understanding the mechanism of action. The protein profiles obtained were analysed for differential protein expression by using Image Master® Platinum 7.0 Software. Results showed that 41 proteins were differentially expressed in 2DE profile of the brain protein samples from rat supplemented with BECA as compared to the control group where; where; 25 proteins were down-regulated and 16 were up-regulated. This analysis will be a basis for the protein identification in understanding the mode of action of neuroprotective effect of BECA.

**Keywords:** Bioactive Extract; Proteomics; Rat; Two-Dimensional Electrophoresis (2-DE); Neuroprotection

## P07

### **Protein Profiling of Human Gut Secretome in Healthy and Colorectal Cancer Patients: A Preliminary Finding**

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Colorectal cancer (CRC) is the most common cancer in Malaysia and dysbiosis of gut microbiome is one of the risk that is frequently associated with CRC. Human gut and microbes secreted many proteins into the extracellular environment of the colon. Study on the secreted proteome may provide information on host-microbe relationship in CRC. The objective of this study is to identify the main protein components in gut secretome of healthy and CRC patients. Fecal materials from eleven healthy individuals and seven diseased samples; one basaloid squamous cell carcinoma (BSCC), one tubulovillous adenoma (high grade dysplasia) and five CRC patients (Stage I, Stage II and Stage III) were collected, homogenized, filtered and profiled using SWATH-MS. We have identified a total of 179 proteins; 132 proteins were secreted by human and 47 were from the microbes. Seven proteins were significantly higher ( $p < 0.05$ ) in CRC with fold change  $> 2.0$ . Interestingly, Ig gamma-4 chain C region (IgG4), Ig gamma-1 chain C region (IgG1), Ig gamma-2 chain C region (IgG2) and protein S100-A9 increased as the disease progresses. These preliminary results suggest the involvement of S100-A9 alongside the immunoglobulins during the progression of CRC in which warrants further investigation.

**Keywords:** Colorectal cancer; Gut microbiome; Secretome; Stool

## P08

### **Secreted Proteome of Human Bronchial Epithelial Cells (BEAS-2B) After Interaction with *Cladosporium sphaerospermum***

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*Cladosporium* spore is found abundantly in indoors and outdoors, and may potentially trigger allergic responses upon inhalation. Other than allergy, some species of *Cladosporium* are known to be pathogenic to human beings. No study to date has investigated how lung epithelial cells would react after the spores are inhaled into the lower respiratory tract. Hence, this study was conducted to investigate the secreted proteome of *Cladosporium sphaerospermum*-infected Human Bronchial Epithelial Cells (BEAS-2B). *C. sphaerospermum* conidia were harvested and co-cultured with BEAS-2B cells for 48 hours. After co-incubation, the spent culture supernatants were collected as treated samples and the controls. Two-dimensional gel electrophoresis was carried out and the gels were stained with silver stain. A total of 24 protein spots together with their relative molecular weights and pIs was identified. The Spot Quantity of each spot is listed. The molecular weight and pI of each protein spot enable speculation of the identity of the protein. Further verification is needed for protein identity.

**Keywords:** Bronchial epithelial cells; *Cladosporium sphaerospermum*; Two-dimensional gel electrophoresis

## P09

### **Development of a Novel Method for the Enrichment of Anticancer Peptides from *Glycine max* Seeds**

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Many studies suggest that soybean seed consumption can reduce the risk of a variety of diseases, including high cholesterol, diabetes, cancer and osteoporosis. Out of these various functions of soybean seeds, we focused on anti-cancer and anti-inflammatory functions in this study. So, this study puts emphasis on three bioactive proteins: Bowman-Birk Protease Inhibitor (BBI, 8 KDa), Lunasin (5.5 KDa) and Leginsulin (4 KDa), which are well known for these functions. Current methods for purification of these proteins are complex, time-consuming, and expensive, and there has been no report that these proteins can be isolated and purified at the same time in seeds. Here, we report a Warm Water Extraction (WWE) method for isolation of these proteins from soybean seeds in a short time at the same time. We checked these proteins accumulation in the WWE fraction and in 28 different varieties by SDS-PAGE and Western blot analysis. As a result, we confirmed that these proteins are not detected in the total soybean seeds extract and enriched in the WWE fraction with highest accumulation in Socheong 2. Therefore, our WWE method can dramatically reduce the time required for the separation and analysis of these proteins in soybean seeds, and this method is highly valuable for large scale enrichment of these proteins, suitable for their commercialization.

**Keywords:** Bowman-Birk Lunasin; Leginsulin; Protease Inhibitor; Warm Water Extraction (WWE) method

## P10

### Chemometric Elucidation of Oil Palm Mesocarp Proteomes

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Oil palm (*Elaeis guineensis* Jacq.) is the most productive oilseed crop which roughly account for 38% of the world's vegetable oil supply and capable to fulfil the large and growing world demand for vegetable oils that is estimated to reach 240 million tons by 2050. The fruit mesocarp and nut that produce palm and kernel oil made oil palm a high yielding oil-producing crop. As to date, advances in technological development have opened up new possibilities focusing on utilizing the omics tools not only for understanding of physiological process but also valuable information towards biomarker discoveries. Proteomics has been explored towards unravelling the biological processes involved in palm oil production by analyzing the fruits harvested at different time-points during maturation and ripening. The proteome data were visualized using a multivariate statistical analysis tools of principal component analysis (PCA) i.e. MetaboAnalyst, SIMCA and COVAIN toolbox to envisage an overview of the proteome content changes during the development of oil palm fruits. PCA revealed clear metabolic shifts from early through late development stages for the oil palm showing various physiological and biochemical composition and structural differences contribute to the operation of unique pathways, genes and proteins.

**Keywords:** *Elaeis guineensis*; Mesocarp; Oil palm; Proteomics

## P11

### Identification of Wound-Response Proteins using 2D-Electrophoresis and LC-MS: A Case Study on Agarwood Tree Species, *Aquilaria malaccensis*

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Wounding is the main trigger to agarwood (*gaharu*) production in *Aquilaria malaccensis* (*karas*), an endangered tree species native to Malaysia. Agarwood acts as a physical barrier and provides a variety of chemical antagonists to fungal attack. To understand the tree's response to wound elicitor, we applied two proteomic approaches. The aim was to compare the efficiency between gel-based 2D-Electrophoresis with direct sequencing via LCMS/MS. Two 3-year-old *A. malaccensis* trees were drilled along the tree stem. The first tree was harvested after 6 h and the second tree after 24 h. A non-wounded tree served as control (0 h). Proteins were extracted from the wounded tissues and processed for the respective analysis. For the 2DE analysis, a total of four spots displayed regulated expression at 6 hours but only two were successfully identified; while for the LCMS/MS analysis, a total of 294 proteins across treatment and control plant were identified. Among them, 128 were specific to wounding, 41 were expressed at 6 h, 87 at 24 h, and another 14 were continuously expressed from 6 h, while 47 were shared. This study demonstrates the efficiency between two proteomic approaches in identifying wound-response protein related to agarwood formation.

**Keywords:** *Gaharu*; 2D Electrophoresis; LCMS/MS; Mechanical wounding



## P12

### **Proteomic Analysis on Root Callus and Protocorm-Like Body of *Vanilla planifolia* Andrews**

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*Vanilla planifolia* Andrews (*V. planifolia*) has been propagated using *in vitro* regeneration method for their benefits as flavouring pods. Callus induction through indirect propagation can be an effective means for mass propagation of vanilla plantlets. Using root tips as explant can provide many advantages due to its easy availability and relatively high regeneration rate. This study was conducted using two-dimensional gel electrophoresis proteomic analysis and Progenesis SameSpots software to investigate the protein changes occur between the callus derived from the root tips of *V. planifolia* and subsequently its conversion into protocorm-like body (PLBs) tissues. TCA/acetone method was used to extract proteins from the callus and PLBs. Comparisons between callus and PLB protein profiles for 2-DE analysis showed that 18 significant differential protein spots ( $P < 0.05$ ) were detected. A total of 11 spots showed significantly higher spot intensity in callus samples compared to PLBs indicating more proteins accumulation present in embryogenic callus tissue compared to PLBs. Identification of these proteins using MS analytical method will help to elucidate biological mechanisms involve in the conversion of callus to PLB in vanilla plants.

**Keywords:** Callus; Protein; Protocorm; Root; *Vanilla planifolia* Andrews

## P13

### A Proteomic Analysis of Mahogany Embryos During Cold Stress Response

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*Swietenia macrophylla* King (Meliaceae) (mahogany) is an endangered and economically important plant indigenous to tropical and subtropical regions of the world. This species was listed in Appendix II (species that may face extinction if trade is not controlled) by the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) in 2002. In this study, the effects of cold acclimation on the storage of mahogany seed was investigated to better understand its preservation. The seeds were stored at 4 and 10°C up to 12 months and 2D gel proteomics was performed to identify differentially expressed proteins. Twelve protein spots were selected (at least two folding values) and further identified using MALDI-TOF MS. One of the identified protein was chaperone protein ClpB3 which is involved in defense mechanism. However, most of the other identified proteins were mainly hypothetical proteins. Further investigation is needed especially in characterizing these hypothetical proteins to better understand the basis of cold stress tolerance for seeds of this important species.

**Keywords:** Cold stress; ClpB3; MALDI-TOF; *Swietenia macrophylla*

## P14

### Investigation of Differential Proteins in Calli of *Capsicum frutescens* Treated with Different Concentrations of Ferulic Acid

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Vanillin, an endogenous chemical made by *Vanilla* spp. was also found as an intermediate product in capsaicin biosynthesis of chilli (*Capsicum* spp). Vanillin is converted from ferulic acid through phenylalanine pathway of capsaicin production. The objective of this study is to find the possible proteins associated with the conversion of ferulic acid to vanillin in the pathway. *Capsicum frutescens* (*C. frutescens*) was used as it has high capsaicin content. Callus of *C. frutescens* were treated with 0.00, 0.20 and 0.60 mM ferulic acid for 28 days. Proteins were extracted from the calli using TCA/acetone method. The proteins were separated by two-dimensional electrophoresis and analysed by Progenesis SameSpots software. The results showed that 0.20 and 0.60 mM treatments have higher protein content than the control treatment. There were 8 significant ( $p < 0.05$ ) differential protein spots found between the 0.20 and 0.60 mM treated callus which has a fold change higher than 2. The results demonstrated that ferulic acid has an influence on *C. frutescens* protein profiles. Future work on the identification of the significant proteins will open a new opportunity for a potential high production of natural vanillin.

**Keywords:** *Capsicum frutescens*; Capsaicin biosynthesis; Vanillin; 2DE

## P15

### Identification of Virulence Related Proteins Associated to Blood Disease Bacterium, a Pathogen Causing Banana Blood Disease

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Banana production in Malaysia has decreased due to infection by a bacterium known as Blood Disease Bacterium. Extensive proteomics data will help in understanding the virulence mechanism of this pathogen. Therefore, this research is aimed to identify the virulence proteins which are involved in the pathogenicity. BDB were grown in two media namely Nutrient Broth (NB) served as control and Minimal Medium M63 served as virulence inducing medium. Technology ITRAQ was used to identified the intracellular and extracellular proteins from both media. As a result, a total of 1332 proteins were identified with high confidence peptides. A total of 88 proteins were extracted and classified as a upregulated proteins from the nutrient broth. Only 18 proteins were identified as upregulated proteins from virulence inducing media. Bioinformatics analysis was carried out to predict the virulence characteristic of the selected proteins by using tools which were SignalP, TargetP, GPI-SOM, SSPRED, and TMHMM. Motif and domain analysis were also performed. From the 18 proteins identified highly expressed in induced media, only 5 proteins are considered as significant virulence proteins based on the *in silico* prediction. The proteins are Signal peptidase, Lipoprotein, Uncharacterized protein, Membrane protein and Serine protease.

**Keywords:** Bioinformatics; Blood Disease Bacterium; ITRAQ; Virulence proteins

## P16

### Mass Tagging for Oil Palm Root Proteome Discovery

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Mass spectrometry (MS) is a powerful platform to assess relative abundance of proteins in biological samples. There are various methodologies used for relative quantification measurement which proportionally related to protein expression levels and abundances in the biological samples. One of them is stable isotope labeling of peptide samples prior to MS analysis. Isobaric tandem mass tag (TMT) and isobaric tags for relative and absolute quantification (iTRAQ) are available up to 8-plex tags can be used subsequently for labeling on any peptide or protein sample. Dayon *et al.*, 2008 reported that TMT 6-plex was able to determine differential expressed proteins in biological samples. Oil palm root proteins were tested for labelling efficiency using the TMT 0 Kit (Thermo Scientific). A total of 100 µg protein samples were dissolved in 100 mM TEAB. Reduction and alkylation were conducted prior to digestion overnight with trypsin (1 µg/µl). Peptide samples were then labelled with the TMT reagent prior to fractionation using a high-pH reverse phase column (Thermo Scientific). A total of 8 fractions were collected and sent for LC-MS analysis via ORBITRAP Fusion Mass Spectrometer (Thermo Scientific). The mass spectrum was analysed by Proteome Discoverer™ Software Version 2.1 (Thermo Scientific) using Oil Palm Uniprot Database. All peptides were validated using the percolator® algorithm, based on q-value less than 1% false discover rate (FDR).

**Keywords:** Oil palm; Tandem mass tagging

## P17

### **Prolonged Culture of *Boesenbergia rotunda* Cells Reveals Decreased Growth and Shoot Regeneration Capacity and Changes in Proteins**

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*Boesenbergia rotunda* is an important ginger that is commonly used as a food ingredient and in ethnomedicinal preparations. Its ethnomedicinal usage has drawn the attention of scientists to further investigate its medicinal properties. *B. rotunda* is traditionally propagated by vegetative techniques using a rhizome segment. However, low proliferation rate, soil-borne disease infection, and degeneration of rhizomes have affected ginger propagation. In this study, proteins associated with shoot regeneration capacity over 9 months of culture for *B. rotunda* cell suspension culture were investigated. Prolonged culture of *B. rotunda* cells generally revealed decreased growth and shoot regeneration capacity. About 42.2 and 53.8 % of cells decreased in growth after 6 and 9 months of culture, respectively, compared to 0-month-old cells. About 67, 57 and 47 % of cell-derived shoot-like-structures for 0, 6 and 9-month-old cells, respectively, were able to regenerate into shoots. Proteins were extracted from each sample using our optimized TCA-acetone precipitation method and analyzed using two-dimensional gel electrophoresis and mass spectrometry. A total of 13 protein spots showed significant differential expression and 8 protein spots were successfully identified. Three regeneration-related proteins were also presented.

**Keywords:** Cell suspension culture; Ginger; Proteomics; Regeneration

## P18

### Identifying Lipid Peroxidation End Products (LPEPs) – Modified Proteins in Wheat Plants

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Lipid peroxidation is the chain reaction which causes degradation of lipid membrane. The decomposition of lipid hydroperoxides in this reaction forms the reactive lipid peroxidation end product (LPEPs) which can damage the proteins, DNA and lipids in the cell. This reaction is generally initiated by reactive oxygen species (ROS) which is largely unstable while LPEP is more stable to be easily measured and study on its impact on plant cells in relation to oxidative stress. In this study, 4-hydroxynonenal (4-HNE), 4-hydroxyhexenal (4-HHE), acrolein and MDA were the focus to reveal their modified proteins in isolated mitochondria and whole tissue in wheat plants. Heat and herbicide treatments were performed to induce oxidative stress in wheat plants *in vivo*. Among the four LPEPs, 4-HNE and acrolein were present *in vivo* under normal and stress conditions in both mitochondria and whole tissues. In contrast to 4-HHE and MDA, there were no modifications *in vivo*. The *in vitro* standards were applied and various putative proteins were targeted by the four LPEPs. The proteins identified both *in vivo* and *in vitro* were the ones involved in TCA cycle, electron transport chain and photorespiration. 4-HNE and acrolein were the ones found active modifying proteins *in vivo* in wheat plants.

**Keywords:** Lipid peroxidation end products; Modified proteins; Oxidative stress; Wheat mitochondria; Wheat whole tissues



**NOTES**

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## Selected Papers from “International Proteomics Conference 2017—Proteomics in Biology and Life Science”

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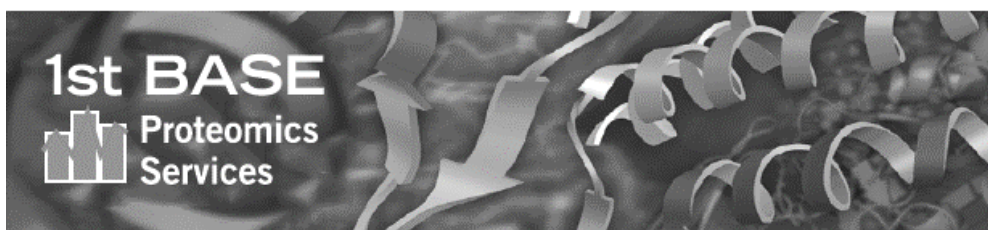
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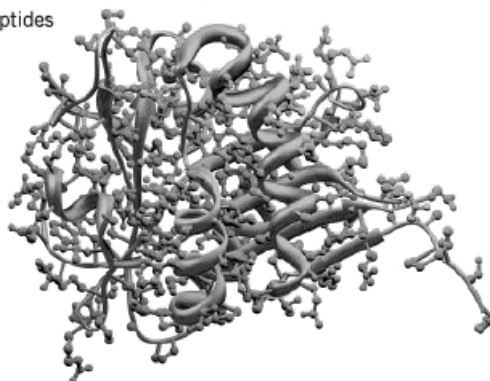
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