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Abstract Booklet

Understanding the role of acyl-CoA-binding proteins in soybean

Nur Syifaq AZLAN and Mee Len CHYE

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Speaker 1

Acyl-CoA binding proteins (ACBPs) are essential proteins involved in lipid metabolism, present in animals, plants, some eubacteria, and archaebacteria. They possess a highly conserved domain of approximately 80 to 90 residues, that bind to acyl-CoA esters, known as the acyl-CoA binding (ACB) domain. Investigations conducted in *Arabidopsis thaliana* (thale cress), *Oryza sativa* (rice), and other plant species such as *Brassica napus* (oilseed rape) categorized plant ACBPs into four classes based on their sizes and additional adjoining domains: Class I or small ACBP, Class II, ACBPs with ankyrin repeats, Class III also known as large ACBPs, and Class IV, which is defined by the presence of kelch motifs. Their differences contribute to their diverse roles mediating in stress responses and plant development. It is imperative to explore the potential functions of these proteins in *Glycine max* (soybean) given that it is an important global crop, as food sources for humans and livestock, as well as in chemical industries, such as bioplastics and cosmetics.

In this study, the *in silico* method was utilized to gain an insight into the roles of soybean ACBPs. Eleven members of soybean ACBPs (GmACBPs) were identified and grouped into four classes. Their domain architecture and subcellular localization were also predicted. Data mining of RNA-sequencing analyses provides an overview of *GmACBP* expression profiles across different organs, including root nodules, a special nitrogen-fixing organ limited to leguminous plants. The analyses revealed high expression of some Class III *GmACBPs* in these nodules hinting on their function in nodulation. An examination of *GmACBP* putative expression patterns in response to both biotic and abiotic stress conditions was also indicated serving as a stepping stone for further research on GmACBPs for crop improvement.

Chromatin landscapes Associated with Brassinosteroid Signaling in *Arabidopsis*

Jiacheng ZHANG and Junxian HE

State Key Laboratory of Agrobiotechnology (CUHK) and

School of Life Sciences, The Chinese University of Hong Kong

Speaker 2

Brassinosteroids (BRs), defined as the sixth group of plant hormones, are steroid hormones in plants and play vital roles in plant growth, development, and stress responses. Chromatin structure controls transcription factor access to the genome and previous studies have revealed that BR signaling is involved in epigenetic regulation, however the chromatin landscapes in response to BR have not yet been studied.

Here, by employing the assay for transposase accessible chromatin with sequencing (ATAC-seq), we profiled changes in the chromatin landscape of *Arabidopsis* shoots associated with BR treatment and mapped the dosage-dependent changing pattern of the differentially accessible regions (DARs). Particularly, we found that activated BR signaling can increase the accessibility of the auxin-induced/growth-related genes, while inhibiting BR signaling increases the accessibility of the ethylene-induced/immune-related genes, which exhibited the phytohormone crosstalk from an epigenomic perspective. Motif analysis with the DARs further identified a catalog of cis-elements including CACGTG, the E-box motif associated with the BZR1/BES1 family TFs that activates BR signaling. These results also uncovered the importance of the BZR1/BES1 family TFs in BR-regulated chromatin dynamics.

Taken together, this study provides a genome-wide landscape of chromatin accessibility changes associated with BR signaling in *Arabidopsis*, and revealed new mechanisms for epigenetic regulation of BR signaling in plants.

The role of m^{6}A modification of mRNAs in plant adaptation to abiotic stresses

Shengjie CHEN and Hon-Ming LAM

State Key Laboratory of Agrobiotechnology (CUHK) and

School of Life Sciences, The Chinese University of Hong Kong

Speaker

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RNA modification is a common post-transcriptional modification in Eukaryotes and plays a critical role in regulating gene expression via modulating mRNA stability, pre-mRNA processing and translation. Among the numerous identified RNA modifications, m^{6}A modification in mRNA has garnered significant attention due to its relatively high abundance and validated role in gene expression regulation. The methylome of m^{6}A is dynamically regulated by methyltransferase (writers) and demethylases (erasers). While the involvement of m^{6}A modification in mammalian stress responses has been extensively studied, its function and mechanism in plant responses to abiotic stresses remain largely unexplored. To investigate the relationship between m^{6}A modification and plant stress responses, we designed a project to unravel the underlying mechanisms. Our findings support the involvement of m^{6}A modifiers in *Arabidopsis* adaptation to abiotic stresses. We observed that abiotic stresses downregulate the expression of m^{6}A writers and upregulate the expression of m^{6}A eraser, leading to a decrease in m^{6}A levels. Mutants lacking m^{6}A writers display salt-related phenotypes, highlighting the pivotal role of m^{6}A writers in plant salt stress responses. Furthermore, we discovered that loss of m^{6}A leads to the absence of stress granules during salt stress, and two reader proteins, ECT2 and ECT3, are involved in the assembly of stress granules under salt stress conditions. To gain insight into the transcriptome-wide distribution of m^{6}A under salt stress, we performed polysome profiling and employed Oxford Nanopore Technology Direct RNA Sequencing (ONT DRS) on wild-type plants subjected to mock and stress conditions. These analyses provided a comprehensive map of m^{6}A modification and revealed a novel layer of m^{6}A function in selectively translating mRNAs during salt stress. In summary, our study provides a comprehensive understanding of the vital role of m^{6}A in regulating plant stress responses. These findings contribute to the growing body of knowledge on RNA modifications and their significance in plant biology.

Transcriptional regulation of the Casparian strip formation in maize root exodermis

Weilun LIU and Silin ZHONG

State Key Laboratory of Agrobiotechnology (CUHK) and

School of Life Sciences, The Chinese University of Hong Kong

Speaker

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Transcription Root architecture is optimized to selectively absorb nutrients and water, while protecting against toxic compounds and pathogens. This selectivity t is largely depend on the formation of an apoplastic diffusion barrier, called 'Casparian strips' (CSs). For most plants, the CSs are found in the root endodermis, an inner layer of cells that surrounds the vascular bundles. Past studies in the model plant Arabidopsis have identified several important genes of CSs formation in endodermis. The CASPARIAN STRIP MEMBRANE DOMAIN PROTEINS (CASPs) are acting to bring together NADPH oxidase and peroxidase, allowing localized ROS production toward peroxidases, thus ensuring localized and efficient lignification. These genes are under transcriptional controls of a network of TFs including SHR, SCR and MYBs in accordance with endodermis development and in response to environmental stresses.

Unlike Arabidopsis, many plant species such as rice, tomato and maize have a more complex root architecture with an extra specialized cell layer, called exodermis. Exodermis is a cell layer outside the cortex and underneath the epidermis and it can also form CSs similar as endodermis. In maize, the exodermis demonstrated great development plasticity and has been shown to be related with stress tolerance. However, the development pathways for exodermis and its CSs formation regulation remain uncharacterized. Our analysis of previously published scRNA-seq of maize root identified endodermis and exodermis cell clusters and the genes specifically expressed in them. We also conducted single cell ATAC-seq to identify differential accessible chromatin regions (ACRs) in endoermis and exodermis cell clusters, along with enriched TF binding motifs in ACRs. All these showed that different sets of CASPs were expressed in endodermis and exodermis suggesting that these two types of cells utilize different sets of genes to construct CSs. Using protoplsat transformation, a scalable and fast ChIP-seq method for mapping genome-wide binding sites was developed and applied to 10 exodermis-TFs. By inferring their target genes, these TFs are shown to play important and diverse roles in the lignin biosynthesis and Casparian strip formation. Next, ChIP-seq will be applied to more exodermis-TFs to identify more TFs regulating Casparian strip formation and construct a transcriptional network for it.

Is photosynthesis of guard cell chloroplast important for stomatal opening?

Shey-Li LIM and Boon Leong LIM

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Speaker

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When the sun rises, the plant leaves require large amounts of energy (ATP) to open stomata so as to facilitate gaseous exchange and photosynthesis. However, some earlier studies showed that guard cell chloroplasts (GCCs) do not undergo CO_2 fixation, resulting in no or only limited photosynthesis in GCCs, which contradicts with the findings of some later studies. Whether GCC photosynthesis directly contributes to stomatal opening has been debated for decades and remains a topic of interest to the scientific community. Recently our group has successfully addressed this fundamental question using genetically encoded fluorescent proteins and GCs starch staining technique. We show that the photosynthetic ability of GCCs is much weaker than that of mesophyll chloroplasts. Due to the limitation of GCC photosynthesis, the importation of cytosolic ATP into GCC via the nucleotide transporter 1 (NTT1) is important for stomata opening. The ATP required for starch biosynthesis in GCCs mainly comes from GC's mitochondria, which consumes sugars imported into GC from adjacent mesophyll cells.

Impacts of Biochar Amendments on Reactive Nitrogen Emissions from U.S. Agricultural Soils

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Speaker

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Fertilizer-intensive agriculture is a leading source of reactive nitrogen (Nr) emissions in the United States, damaging air quality, human health, and climate. Nr emitted from agricultural soils includes air pollutants nitric oxide (NO) and ammonia (NH_3), which contribute to the formation of health-damaging tropospheric ozone and particulate matter air pollution, and the potent greenhouse gas nitrous oxide (N_2O). Biochar, a carbon-rich material produced from biomass pyrolysis under oxygen-limited conditions, has gained attention as a soil amendment for its potential to reduce Nr emissions, boost crop yields, and provide long-term carbon storage in soils. However, the impacts of biochar on Nr emissions vary widely across different agricultural regions, ranging from positive to negative and even neutral. Previous studies have typically relied on field measurements or field-scale agroecosystem models, which fail to adequately characterize such complex variations across U.S. agricultural lands.

We incorporated biochar algorithms into a regional-scale agroecosystem model to simulate Nr emission changes in the year following application of biochar. Scenario analyses were then performed with application rates of 5 and 20 ton ha^{-1} for biochar in U.S. fertilized soils.

Our simulations predicted that the impacts of biochar amendments on Nr emissions would vary widely (-17% to + 27% under 5 ton ha^{-1} applications, -38% to +18% under 20 ton ha^{-1} applications) and depend mostly on how nitrification is affected. Low-dose biochar application (5 ton ha^{-1}) stimulated emissions of all three nitrogen species in 75% of simulated agricultural areas, while high-dose applications (20 ton ha^{-1}) mitigated emissions in 76% of simulated areas. Biochar amendments are most likely to mitigate emissions if applied at high rates in acidic soils (pH < 5.84) with low organic carbon (< 55.9 kgC ha^{-1}) and inorganic nitrogen (< 101.5 kgN ha^{-1}) content. Our simulations could inform where the application of biochar amendments would be most likely to mitigate Nr emissions and their associated adverse impacts.

Molecular Mechanism of Autophagy in the Stress Response and Sequestration of Environmental Pollutants in Plants

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Speaker 7

During evolution, eukaryotic cells and organisms develop diverse strategies to effectively handle stressors in a manner often specifically selected to address certain threats, such as pathogen presence, nutrient scarcity, adverse environmental conditions, or accumulation of misfolded proteins within the cells. While several biotic and abiotic stressors are well reported, there is increasing interest in environmental pollutants due to their nexus with health, environmental sustainability, and food security, of which plants are the primary subject. In plants, the survival mechanisms employed during stress responses vary in an attempt to promote survival, and in view of the sequestration pathway, while previous studies have elucidated the complex transcriptional regulatory networks involved in plants' heavy metal stress response. However, the translational regulation and mechanisms of organellar remodeling and homeostasis remain elusive. Herein, it is hypothesized that autophagy, an evolutionarily conserved process that mediates the spatiotemporal degradation of superfluous cellular constituents through autophagosome formation, may play a crucial role in maintaining plant cellular homeostasis under heavy metal stress in a highly regulated manner.

Exploring an autophagy-deficient mutant in a mixed heavy metal coupled system, we investigated the germination, growth, and survival rates of *Arabidopsis thaliana*. The chlorophyll profile revealed a concentration-dependent relationship with the survival rate. The *Arabidopsis* autophagy-related (ATG) system transcriptional study reveals that all nine ATG8 family genes are upregulated by heavy metal stress. Our recent study showed that heavy metal stress increased the formation of YFP-ATG8e-labeled autophagic structures and the autophagic flux in root cells of *Arabidopsis* seedlings. Preliminary results from experiments of the proteolytic cleavage of YFP-ATG8e, the vacuolar degradation of endogenous ATG8 proteins, and the lipidation of ATG8, indicate that heavy metal stresses do drastically alter the autophagic flux in plants. A selective autophagy receptor NBR1 will be examined using a double transgenic system in the future.

In summary, our recent findings reveal that heavy metals are stress inducers of autophagy. However, further biochemical studies are needed to characterize the stress type while exploring microscopic investigations to reveal the subcellular autophagy cargo regulated by heavy metal stress. Further understanding of these mechanisms will elicit the remodeling of the autophagic activity in plants upon environmental pollutant stresses. Moreover, this study will likely bridge environmental sustainability and sustainable agriculture, contributing to specific UN sustainable development goals (SDGs).

Understanding Cerebellar Ataxia: Unraveling the Role of Wnt10b and Yy1 in Purkinje Neuron Development

Ying Lam LUI, and Kin Ming KWAN

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Speaker

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Yin Yang 1 (YY1) haploinsufficiency syndrome was first reported in 2017, and patients often have cerebellar ataxia, cognitive impairment, behavioral alterations, and numerous congenital malformations. Although YY1 is a well-known zinc-finger-like transcription factor due to its dual role in gene regulation, acting as both an activator and a repressor, its role in postnatal cerebellar development is still elusive. Therefore, to better understand pathological mechanisms regarding motor coordination in YY1 syndrome, we sought to reveal the developmental mechanism regulated by YY1 in Purkinje cells (PCs), an integration center in cerebellar networking for motor coordination in the cerebellum.

In this study, we show that the genetic abolishment of Yy1 PCs postnatally resulted in cerebellar ataxia phenotype following progressive cerebellar atrophy. Interestingly, the significantly defective PC dendritogenesis found in mutant mice caused the weakening of synaptic plasticity and Protein kinase C gamma (PKCγ) dysregulation. By the transcriptomic study of ataxic-like mutants, Wnt10b, a specific activator of the Wnt/β-catenin pathway and expresses particularly in postnatal PCs, showed reduced transcriptional and translational expression. A functional study of Wnt10b performed ex vivo by siRNA and recombinant Wnt10b delivery indicated that Wnt10b positively regulated PKCγ expression in postnatal PCs. More surprisingly, knockdown of Wnt10b ex vivo, PCs had comparable phenotypes to Yy1 ataxic mouse PCs.

Our study suggested that Yy1 transcriptionally regulates Wnt10b to control postnatal PC development highlighting a critical role in dendritogenesis mediated with PKCγ. In addition, the novel pathway we suggested could provide insights into the potential roles of Yy1 in PC development and additional molecular clues on YY1 haploinsufficiency.

Coordination between chlorophyll biosynthesis and plastid translation in plants

Jiashuo ZHANG, Dali FU, Shaoti SUI, and Peng WANG

State Key Laboratory of Agrobiotechnology (CUHK) and

School of Biological Sciences, The University of Hong Kong

Speaker

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Photosynthesis is an indispensable process that converts light energy into chemical energy stored in carbohydrates. During this process, light absorption relies on a group of photosystem core complexes and peripheral light-harvesting complexes. In nature, these complexes are pigment-protein complexes, in which chlorophylls (Chls) are associated with nucleus-encoded light-harvesting chlorophyll-binding proteins (LHCPs) and plastid-encoded chlorophyll-binding proteins such as D1 and PsaA. More importantly, Chl plays an essential role in the folding and membrane integration of Chl-binding proteins, whereas free chlorophyll and most of its precursors are phototoxic. In this context, optimal photosynthesis and plant fitness cannot be achieved without the tight regulation of Chl metabolism and the precise coordination between Chl biosynthesis and the assembly of various Chl-binding proteins. However， molecular mechanisms underlying this coordination is largely unknown.

The chloroplast signal recognition particle (cpSRP) pathway is responsible for sorting the LHCPs and plastid-encoded Chl-binding proteins to the thylakoid membranes. We have shown that ATP-independent molecular chaperone cpSRP43 contributes to the post-translational transportation of LHCPs and the thermoprotection of multiple Chl biosynthetic proteins in chloroplasts. This regulation enables plants to couple Chl biosynthesis with the biogenesis of LHCPs during chloroplast development and heat shock response. However, how Chl biosynthesis is orchestrated with the biogenesis of plastid-encoded Chl-binding proteins remains open. It is hypothesized that the co-translational cpSRP pathway and plastid translational machinery might directly interact with the key enzymes of Chl biosynthesis to facilitate the timely integration of Chl into the plastid-encoded Chl-binding proteins. To examine this hypothesis, we characterized Chl biosynthesis in the mutants with defects in the co-translational cpSRP pathway and plastid ribosomal subunits and tested the protein interaction of Chl biosynthetic enzymes with the candidate cpSRP components and plastid ribosomal subunits. Our results outline the factors coupling Chl biosynthesis with plastid translational machinery. Taken together, our study will elucidate a novel insight into the complex control of Chl biosynthesis and plastid protein translation and will provide a theoretical basis for improving the photosynthetic efficiency of crops, increasing crop yields, and expanding high-quality germplasm resources.

Structural insight into how VSR1-PA interact with cargo c-terminal peptide sequence

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Centre for Protein Science and Crystallography, State Key Laboratory of Agrobiotechnology, School of Life Sciences, The Chinese University of Hong Kong

Speaker

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Vacuolar trafficking of the storage protein travels from the endoplasmic reticulum (ER) to the vacuoles for storage or degradation, or else being secreted out. Storage/ cargo proteins possess mainly two types of motifs for transport to correct destination, denoted as the vacuolar sorting determinant VSD, classified as the sequence-specific VSD (ssVSD) and C-terminal VSD (ctVSD), which interact with trafficking receptor. The ssVSD has the conserved NPIR-like consensus sequence motif, whereas ctVSD is positioned at the c terminus. Type I transmembrane protein - vacuolar sorting receptor (VSR) composes of a protease associated domain (PA), a Central domain and 3 epidermal-growth-factor like (EGF) domains at the N terminal, a transmembrane domain, and a cytosolic c-terminal domain. VSR aids cargo trafficking, that mis-sorting of the cargo protein was observed under VSR mutated background. The protease associated domain of VSR1 (VSR1-PA) interacts with the sequence preceding the conserved NPIR motif of aleurain through structural determination – arginine 95 of VSR1-PA is essential for cargo recognition and sorting.

The co-crystal structure of VSR1-PA with the c-terminal sequence pentapeptide of 12S globulin Cruciferin 1 (Cru1) (PDB:7F2I) proved the cargo binding site is of important for ctVSD recognition - the fourth last and antepenultimate residues of the pentapeptide fit the cargo binding site with backbone hydrogen bond formation, and the carboxyl group of the ctVSD pentapeptide form salt bridges with R95 of VSR1-PA.

Up to date the VSR1-PA structure in complexed with VL22 is newly solved: the penultimate phenylalanine of the pentapeptide is located next to Y99 in the cargo binding site giving a more promising hydrophobic interaction in comparison to interaction with CRU1 alanine rich tail. The side chain of the antepenultimate arginine of the pentapeptide despite facing away from the cargo binding loop, the structure integrity should be harmonized by the electrostatic interaction with conserved residue D119 and E123, reserving the receptor cargo interaction. Supplemented with the thermal shift assay to study VSR1-PA/ ctVSD interaction, results suggest that the motif specificity of ctVSD is much more complex than the general assumption that ctVSD is abundantly hydrophobic.

A Novel reciprocal regulation mechanism for SH3P2 in crosstalk between endocytosis and autophagy

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Centre for Cell & Developmental Biology and State Key Laboratory of Agrobiotechnology, School of Life Sciences, The Chinese University of Hong Kong

Speaker

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The serine/threonine kinase, Sucrose non-fermenting 1 (SNF1)-related protein kinase 1 (SnRK1), functions as a master energy sensor in plants by phosphorylating the substrate proteins to activate cellular processes, including autophagy. In our preliminary screening, we identified the plant-specific autophagic regulator SH3 domain-containing protein 2 (SH3P2) as a substrate candidate of SnRK1. Previous studies have shown that under normal condition, SH3P2 mainly participates in the clathrin-coated vesicle (CCV)-mediated endocytosis and cell plate formation. Upon autophagic induction, SH3P2 interacts with the autophagosomal marker autophagy-related (ATG) protein 8 and is recruited to the autophagosomal membrane. Our recent study demonstrated that SH3P2 carries an atypical ATG8-interacting motif (AIM) for binding with ATG8. Intriguingly, by mass spectrometry analysis, we have identified several phosphorylation sites upstream/downstream of the SH3P2 AIM. Here we will present our recent data on how SnRK1 regulates the phosphorylation of SH3P2 AIM to modulate its activity in plant autophagy, highlighting a novel SnRK1-dependent postmodification mechanism for fine-tuning autophagic and endocytic activities to balance cellular homeostasis.

Using baleen to identify combinatorial RNA modification events at transcript isoform level through Nanopore direct RNA sequencing

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Speaker

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Chemical modifications of RNA play crucial roles in post-transcriptional regulation, dynamically regulating RNA turnover, including export, translation, and degradation. Despite this, the significance of RNA modification is often overlooked, making an in-depth investigation essential to understand how aberrant RNA modifications and their regulation contribute to various phenotype changes. Although existing screening technologies allow for the examination of RNA modification on the transcriptome, the limitations are obvious: 1. Covering a few types of modification (m1A, m5C and pseudouridine etc.); 2. detecting RNA modifications in low-resolution (on gene-level or region detection); 3. Identifying one type of modifications in one-shot (no mutual interaction between different modifications provided). Working on native RNA provides an alternative option. Although combining nanopore direct RNA-seq (dRNA-seq) and in vitro transcription has shown theoretical feasibility for the detection of RNA modifications, it is still limited by the throughput and challenging detection algorithm. To overcome these limitations, I proposed an approach called BALEEN that integrating a whole transcriptome in vitro transcription protocol and a high-resolution modification detection algorithm. By generating whole transcriptome in vitro transcription products, playing as pure control with identical sequence, and leveraging the dynamic time warping algorithm, identifying differences between signals of modified and unmodified nucleotides, we could identify single modified nucleotide on single RNA molecule-level. The newly developed approach could help identify almost all the modified bases on RNA, allowing us to study the combinatorial RNA modification events on different RNA molecules of the same transcript.

Establishing multiple metabolite profiling platforms and rapid gene function characterization system

Xia ZHANG and Pan LIAO

Department of Biology, School of Science, Hong Kong Baptist University

Speaker

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Plants synthesize a wide range of volatile organic compounds that are chemically diverse and primarily consist of terpenoids, fatty acid derivatives, benzenoids, and phenylpropanoids. These compounds play essential roles in plant survival and interaction with the environment while also serving as natural products for humans. Our laboratory primarily focuses on studying the biosynthetic pathways and transport mechanisms of volatile and non-volatile compounds such as monoterpenes, phytosterols, and lipids. Since last July, we have successfully established the following technical platforms: 1) Extraction and analysis of terpenoids using GC-MS analysis; 2) Extraction and analysis of tropane alkaloids using LC-MS/MS analysis; 3) Extraction and analysis of fatty acids by GC-MS analysis; 4) Gene transient expression system in *Nicotiana benthamiana* for rapid gene function characterization followed by GC-MS, LC-MS/MS or HPLC analysis. In summary, the establishment of these platforms will facilitate our ongoing projects and can also be utilized by any members of SKLA if deemed necessary.

Investigating the Dispersion and Factors Impacting the Distribution of *Bradyrhizobium* in Soil

Jinjin TAO and Haiwei LUO

State Key Laboratory of Agrobiotechnology (CUHK) and

School of Life Sciences, The Chinese University of Hong Kong

Speaker

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By developing an efficient selective medium, we obtained 432 novel *Bradyrhizobium* isolates from the soil. These new strains contribute to basal branches within different supergroups of *Bradyrhizobium*, providing valuable insights into the evolutionary history of the *Bradyrhizobium* genus. Utilizing an updated database, we have demonstrated that all *Bradyrhizobium* supergroups, except the Photosynthetic supergroup, originate from non-nitrogen-fixing ancestors. Additionally, we discovered two novel nitrogen fixation gene clusters, namely FL3 and Kakadu clusters. The nitrogen-fixing potential of these newly identified clusters may have implications in fields such as soil health, ecological restoration, and sustainable agriculture.

Through 16s *rRNA* and *nifH* amplicon surveys, we found that *Bradyrhizobium* is the predominant species within both the soil bacterial and diazotrophic bacterial community. Furthermore, by designing specific primers for the *Bradyrhizobium rpoB* and *nifH* genes, we conducted a comprehensive investigation into the diversity and absolute abundance of the *Bradyrhizobium* community. The results showed that the average population size of *Bradyrhizobium* is 6.61×10^7 per gram of soil, while the average abundance of *nif*-carrying *Bradyrhizobium* is 1.37×10^7 per gram of soil. On average, 20.3% of *Bradyrhizobium* in the soil possesses *nif* genes, but the *nif*-carrying ratio in *Bradyrhizobium* varies significantly across different locations. Mean annual temperature, mean annual precipitation, and total organic carbon are the main factors that affect the dispersion of *Bradyrhizobium*. *Bradyrhizobium* carrying the *nif* genes prefers warm, moist, and organic-rich soils, whereas *Bradyrhizobium* lacking the *nif* genes exhibits stronger adaptability to low temperatures and drought, and a preference for neutral pH soils.

Modulating Metabolic Plasticity for Obesity Prevention: The Role of Asperuloside and its Target NRF2/PD-L1 Axis

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Speaker

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Adipose tissue (AT) is a central metabolic organ controlling whole-body energy homeostasis. Hypertrophic obesity resulting from impaired white ATs (WATs) functionality and lipid turnover promotes insulin resistance and dyslipidemia, which greatly contributes to obesity-associated premature death and the development of cardiometabolic disorders and cancers. Though several compounds have been identified with anti-obesity effect such as asperuloside (ASP), their precise mechanisms remain unidentified.

Programmed cell death ligand-1 (PD-L1), an immune coinhibitory checkpoint, is a transmembrane protein expressed on adipocytes. Deletion of PD-L1, either globally or specifically in adipocytes, exacerbates AT dysfunction by polarizing pro-inflammatory macrophages and dendritic cells. Similarly, pharmacological blockades also worsen AT pathology following overnutrition. However, these studies primarily focus on immunological events, and the intrinsic regulation of adipocyte functionality, such as metabolic reprogramming, by PD-L1 signaling, remains largely unknown.

Unlike the role of WAT, brown AT (BAT) is responsible for the dissipation of energy under cold temperature through uncoupled respiration via the uncoupling protein 1 (UCP1). Improvement in insulin sensitivity and lipids metabolism are observed in activated BAT, suggesting the metabolic benefits of browning, the transdifferentiation of white adipocyte towards brown adipocyte. Recent studies have proposed PD-L1 as a marker in brown adipocytes and downregulated UCP1 expression in WATs from PD-L1-deficient mice. These findings suggest a potential link between PD-L1 and adipose browning. However, the involvement of PD-L1 in the white-to-beige transition remains largely unexplored. To further investigate the role of adipocyte PD-L1, white adipocytes differentiated from mice subcutaneous WATs-isolated preadipocytes were used. Activation of PD-L1 by PD-1 in the mature adipocytes results in the downregulation of the de novo lipogenesis pathway and increase in lipolysis as well as fatty acid oxidation, implying the potential role of PD-L1 in lipid metabolism restoration under obesity. Moreover, PD-L1 activation stimulates the induction of brown phenotypes and the upregulation of glycolysis in white adipocytes, is abolished in drug-induced browning mice model with PD-L1 blockade. Additionally, we found that ASP restores PD-L1 expression in obese WATs by activating NRF2, which has never been reported.

Bearskin 2 mediates xylogalacturonan secretion protecting Arabidopsis root from salt stress

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Speaker 16

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Border-like cells (BLCs) constitute a sheet at the surface of the Arabidopsis root cap that is continually shed and replenished. Xylogalacturonan (XGA) is a pectic polysaccharide associated with cell wall degradation and is actively secreted from BLCs. ROOT CAP POLYGALACTURONASE (RCPG) encodes a putative pectinase crucial for BLC shedding. BEARSKIN1 (BRN1) and BRN2 are Arabidopsis NAC family transcription factors, regulating RCPG expression. To investigate the interplay between XGA and cell wall digestion, we investigated XGA secretion in BLCs in which RCPG expression was manipulated. Our findings unveiled that RCPG is a protein cargo of XGA-carrying vesicles budding from the trans-Golgi. XGA secretion remained unaltered in *rcpg or brn1*. By contrast, XGA was not detected in in *brn2* indicating that *BRN2* is required for XGA synthesis. Overexpression of RCPG-GFP (*oeRCPG-GFP*) resulted in accelerated BLC turnover and altered BLC cell wall, leading to disruptions in the root cap cuticle (RCC). Intriguingly, XGA accumulated in the cell wall sites underlying the cracks in RCC of *oeRCPG-GFP*. When subjected to sodium ion stress, *brn2* plants exhibited heightened vulnerability, suggesting a role of XGA in fortifying the compromised cell wall of BLCs.

The function of sesquiterpenoid in chelicerates

Wai Lok SO and Jerome Ho Lam HUI

State Key Laboratory of Agrobiotechnology (CUHK) and
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Speaker

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Sesquiterpenoid juvenile hormone (JH) is a of 15-carbon hormone that has long thought to be confined to the insects for its renowned biological functions, including the control of molting, development and reproduction. While it remains true that JH is specifically synthesized in insects, other types or primitive forms of sesquiterpenoids have also been discovered in other arthropod lineages, for instance methyl farnesoate (MF) and farnesoic acid (FA), which are the precursors of JH, were found to be the functional counterparts of JH in crustaceans and myriapods. However, it remains unclear how the sesquiterpenoid system functions in chelicerates, which is an arthropod group comprising scorpions and spiders. Here, we sequenced and assembled a chromosomal-level genome of the dwarf wood scorpion, *Liocheles australasiae* and the biosynthetic gene cassette, as well as the function of sesquiterpenoids on different life stages have been examined.

Sesquiterpenoids are synthesized by an animal-conserved mevalonate (MVA) pathway, with an arthropod-specific downstream reactions that ultimately yield either FA, MF or JH. In *L. australasiae*, a complete biosynthetic pathway was identified and annotated. *In vitro* injection of FA and MF in early juveniles and adults demonstrate a differential responses between stages, which juveniles are more responsive to FA while adults are more responsive to MF. In particular, the hormone-treated juveniles showed enriched upregulated KEGG pathways including *the insect hormone biosynthesis* and *Homeobox genes*, that are closely related to the known functions of sesquiterpenoid as demonstrated in insects and crustaceans. While juveniles and adults display differential responses to different form of sesquiterpenoids, three pathways relating to development have been upregulated and enriched in both juveniles and adults. This demonstrates the need of different form of sesquiterpenoids to activate the pathways throughout the scorpion life cycle.

The current study expands our knowledge on the function of sesquiterpenoids in an understudied arthropod lineage and provide insights on how endocrine system evolves in animals. This study is going to expand to the other chelicerate members including spiders, to investigate the regulatory effect of sesquiterpenoids from an evolutionary perspective.