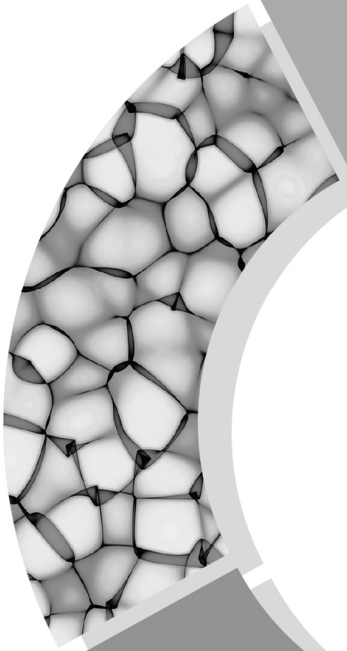
포스터 세션



- GM: Genetics/Molecular Biology
- DP: Development/Physiology
- ST: Signal Transduction
- ETM: Environment/Ecology/Taxonomy/Morphology
- PB: Plant Biochemistry

[GM1]

Characterization of *Rosellinia necatrix* transcriptome and genes related to pathogenesis and cell cycle by single-molecule mRNA sequencing

Hyeongmin Kim[1], Jinsu Lee[1], Hyemin Kim[1], Hojin Ryu*[1]

[1]*Department of Biology, Chungbuk National University, Cheongju 28644, Republic of Korea*

White root rot disease caused by infection of *Rosellinia necatrix* is one of the world's most devastating plant fungal diseases in particular fruit trees. Recent global outbreaks and molecular advances of *R. necatrix* have been increased the interest in this pathogen. Less information for genome structures and transcriptome of the *Rosellinia necatrix* has been a barrier to the progress of functional genomics research and control in this harmful disease. We here newly identified 10,616 full length transcriptomes from filamentous hyphal tissue of *Rosellinia necatrix* KACC 40445 strain using PacBio single-molecule sequencing technology. After annotation from the unigene sets, we selected 14 cell cycle related genes, which were probably either positively or negatively involved in hyphae growth by cell cycle control. The expression level of the selected genes was further compared in two strains which are showed in differential growth rate on the nutritional media. Furthermore, we predicted pathogen related effector genes and cell wall degrading enzymes from the annotated gene sets. These results provide the most comprehensive expressed gene resources for *Rosellinia necatrix,* and could facilitate functional genomics and further analyses in this important phytopathogen

***Corresponding Author email :** hjryu96@chungbuk.ac.kr

[GM2]

A clathrin-independent endocytic pathway is implicated in PIN-FORMED 5 trafficking in Arabidopsis root epidermal cell

Richard Dong Wook Lee[1], Anindya Ganguly[2], Hyung-Taeg Cho*[1]

[1]*Department of Biological Sciences, Seoul National University, Seoul 151-742, South Korea2Department of Biological Sciences, Washington University, Saint Louis, MO 63130, USA*

The phytohormone auxin is crucial to plant development. Therefore many auxin transporters, including the auxin efflux carrier, PIN-FORMED (PIN) proteins, have been a major topic of interest. However, while the trafficking mechanisms of long PINs, or PIN proteins with long hydrophilic loop (HL), have been observed, the short PINs (with a short HL) still remain to be elucidated. One of the reasons for the lack of short PINs research is their internal localization, making a direct comparison impossible with long PINs that normally resides at the plasma membrane. Here, we expressed PIN5, one of short PINs, under the PIN2 promoter, which allowed PIN5 to target to the plasma membrane rather than to internal compartments. Auxin treatment was not able to block PIN5 from internalizing while impeding PIN5 localization to the plasma membrane. TyrA23, a clathrin inhibitor, did not inhibit PIN5 internalization indicating that PIN5 is able to internalize through a clathrin-independent pathway. The clathrin-independent pathway was further supported when PIN5 was able to form BFA bodies when co-treated with BFA and auxin/TyrA23. As the HL of PIN proteins are crucial to their trafficking, a deletion study of PIN5-HL was conducted. The deletion of HL limited PIN5 from targeting to the plasma membrane, indicating that the PIN5-HL contains certain motifs distinct from long PINs that directs PIN5 trafficking.

***Corresponding Author email :** htcho@snu.ac.kr

[GM3]

Application of CRISPR/Cas9 technology to increase hydroxy fatty acid in transgenic Arabidopsis

Mid Eum Park[1], Hyun Uk Kim*[1]

[1]*Department of Bioindustry and Bioresource Engineering, Plant Engineering Research Institute, Sejong University, Seoul, 05006 Republic of Korea*

Triacylglycerol (TAG) is accumulated in plant seeds that is composed of a glycerol with three fatty acids. There are various kinds of fatty acids in plants. Among them, hydroxy fatty acid (HFA) is an useful for industrial feedstocks. Castor bean plants have a large amount of HFA in seeds, but they also have a toxin ricin and allergenic 2S albumin in seeds. Because of this drawback as a crop plants, many researchers have tried to produce HFA in Arabidopsis as a model oil crops using transformation with genes related to HFA biosynthesis from castor bean plants. Expression of castor bean HFA synthesis gene, *RcFAH12* in Arabidopsis could produce HFAs up to 17% in seed oil. When we coexpressed *RcFAH12* and *RcPDAT1-2* encoding a phospholipid: diacylglycerol acyltransferase, HFA was accumulated up to 22~23% in seed oil. Additional expression of *RcDGAT2* encoding a diacylglycerol acyltransferase identified from castor bean to *RcFAH12+RcPDAT1-2* transgenic Arabidopsis increased HFA up to 27% in seed oil. Currently, we are trying to knock-out genes encoding Arabidopsis PDAT and DGAT using CRISPR/Cas9 technology. Elimination the function of competitive endogenous genes involved TAG biosynthesis in transgenic Arabidopsis will increase HFA more than current maximum 27% level, so these result will contribute to produce better and more HFA in oil crops.

***Corresponding Author email :** hukim64@sejong.ac.kr

[GM4]

Identification of Mildew Locus O (MLO) genes required for susceptibility to powdery mildew in melon

Hyun Seong Lee[1], Haeun Jung[1], Hyun Uk Kim*[1]

[1]*Department of Bioindustry and Bioresource Engineering, Plant Engineering Research Institute, Sejong University, Seoul, 05006 Republic of Korea*

Melon (*Cucumis melo* L.) is an important fruit crop and is produced more than 31 million tons in worldwide. However, powdery mildew (PM) diseases, the main factor of reducing crop yields, affect melon farming these days. For this reason, we are developing the molecular markers to detect PM resistance in melon breeding lines. We choose *Mildew Locus O (MLO)* gene as the molecular marker. MLO encodes 7 transmembrane protein. Although, their exact functions are not revealed, some MLO proteins cause susceptible to PM pathogen. If the MLO proteins, which are susceptible to powdery mildew pathogen, lose their function by integration of a transposable element, plants have resistance to PM diseases. We identified 13 MLO genes from melon genome database. They are categorized into six clades, and clade V MLO proteins cause susceptible to powdery mildew disease in dicot plants. On the basis of previous researches, we can pick out the melon clade V MLO proteins using phylogenetic tree. *CmMLO3, 5*, and *12* are grouped in clade V. To find the differences between PM susceptible and resistance melon breeding lines, we use PCR to identify genome structure of *CmMLO3, 5*, and *12*. We find differences in some regions in *CmMLO5*, these regions would be occurred insertions. To seek accurate differences between susceptible and resistance lines, we will identify cDNA sequence of two lines, and compare to reference genes. Finding sequence differences between susceptible and resistance lines will guide us to identify where the loss of function mutations is usually occurred. It can help to develop the PM molecular marker.

***Corresponding Author email :** hukim64@sejong.ac.kr

[GM5]

Study on Fibrillin 1a,b,2 roles in abscisic acid- and jasmonate-mediated photoprotection in Arabidopsis

Inyoung Kim[1], Hyun Uk Kim*[1]

[1]*Department of Bioindustry and Bioresource Engineering, Plant Engineering Research Institute, Sejong University, Seoul, 05006 Republic of Korea*

Fibrillin 1a, b and 2 are lipid-associated proteins of plastids and accumulated under abiotic Fibrillin 1a, b and 2 are lipid-associated proteins of plastids and accumulated under abiotic stress. In high light stress, FBN1a,b-2 accumulation are regulated by abscisic acid (ABA) response regulators ABA-insensitive 1 (ABI1) and ABI2. It enhances resistance of photosystem II photoinhibition. The *FBN1-2* transcriptional and posttranscriptional levels increased in the presence of ABA. The phenotype of *FBN1a,b-2* suppressed transgenic plant show a lower shoot growth development and reduced anthocyanin accumulation under high light/cold stress. These phenotype of transgenic plants are restored by jasmonate (JA) treatment. These results that FBN1a,b-2 are expected to mediate by jasmonate biosynthesis. However the precise functions of FBN1a,b-2 enhancing photoprotection under the high light/cold stress through ABA and JA pathway are still mystery. To investigate major roles of FBN1a,b-2 in photoprotection mediated with ABA and JA signalling pathway, we are generating each of single, double, and triple mutants of *fbn1a,b-2* using selection of T-DNA insertional mutants and combined with CRISPR/Cas9 gene editing system.

***Corresponding Author email :** hukim64@sejong.ac.kr

[GM6]

Functional analysis of *cryptochrome-interacting basic-helix-loop-helix1 (OsCIB1)* in controlling leaf angle and grain size in rice

Hyoseob Seo[1], Nam-chon Paek*[1]

[1]*Department of Plant Science, Seoul National University, Seoul 08826, Korea*

Cryptochrome-Interacting basic-helix-loop-helix (CIB), a critical transcription factor in plant, plays important roles associated several development processes, including hypocotyl elongation, flowering, and plastid development. It has been reported that a soybean CIB orthologue gene *Cryptochrome-interacting bHLH1* (*GmCIB1*) promotes leaf senescence by activating transcription of senescence-associated genes such as *WRKY DNA BINDING PROTEIN53b* (*WRKY53b*). However, any functions of *CIB1* have not been studied or reported yet in rice. In this study, we screened two T-DNA mutants to identify the function of *OsCIB1*. Especially, a rice gain of function mutant, *oscib1-D,* displayed wide leaf angles and slender grains, similar to plants with increased brassinosteroid (BR) levels or enhanced BR signaling. qRT-PCR analysis showed that genes in brassinosteroid signaling pathway are upregulated in *oscib1-D*, but there was no significant difference of the expression level of BR biosynthesis-related genes between WT and *oscib1-D*. In addition, *osicb1-D* showed more sensitive phenotype than WT to BR. Histological analysis revealed that increased cell length in adaxial surface of lamina joint is responsible for larger angles. Moreover, expression level of genes involved in cell elongation such as expansins and xyloglucan endotransglycosylase/hydrolase(XTH), two major cell wall-loosening enzymes, was significantly increased in *oscib1-D*. Thus, these results strongly suggest that *OsCIB1* is involved in the BR signaling pathway and determines not only leaf inclination but also grain shape by regulating cell-elongation-related genes.

***Corresponding Author email :** ncpaek@snu.ac.kr

[GM7]

The regulation of plant immunity by NAC4 transcription factor and *miR164* in *Arabidopsis thaliana*

Myoung-Hoon Lee[1], Hwi Seong Jeon[1], Hye Gi Kim[1], Ohkmae K. Park*[1]

[1]*Department of Life Sciences, Korea University, Seoul, Republic of Korea*

Hypersensitive response (HR) is one of programmed cell death (PCD) and the primary immune response. It has been known that several genes and processes are involved in regulation of HR-PCD. Here, we show that a *miR164* and its target gene NAC4 transcription factor (At5g07680) play essential roles in the regulation of HR-PCD in *Arabidopsis thaliana*.

In T-DNA insertion mutant for GDSL lipase 1 (GLIP 1), HR-PCD symptoms were not restricted and spread to uninfected leaf tissue in response to avirulent bacterial pathogens. The *miR164c*, one of miR164 family was down- or up-regulated in *glip1-1* mutant or GLIP1-overexpressing (*35S:GLIP1*) plants, respectively. Especially, HR-PCD symptoms were enhanced in NAC4-overexpressing (*35S:NAC4*) and *mir164* mutants in response to avirulent bacterial pathogens, but no difference in leaf senescence. NAC4 expression was induced by pathogen infection and negatively regulated by *miR164*. Moreover, the plants expressed the *miR164*-resistant mutant NAC4 gene were shown the enhanced mRNA level and PCD symptoms. These results suggest that NAC4 regulates PCD by infection with avirulent bacterial pathogen through positive, and this regulation is controlled by function of *miR164*, negatively.

In addition, microarray analysis data showed that the most genes shown different expression were down-regulated or up-regulated in *35S:NAC4* or nac4-1 plants, respectively, and cell death-related genes were up-regulated in nac4-1 plants. Through various experiments, I found that the NAC4-binding DNA sequence is 'ACAAGCAAC' and several cell death-related genes shown up-regulated in *nac4-1* plant possess the NAC4-binding sequence in their promoter region. These data mean that NAC4 transcription factor may have a negative role in the gene expression.

In particular, the increased cell death ratio of protoplast isolated in *35S:NAC4* is decreased when the *LURP1*, *WRKY40* or *WRKY54* genes were expressed in the protoplast. Among these genes, promoter activities of *LURP1* and *WRKY40* were decreased or increased in protoplasts of *35S:NAC4* or *35S:miR164*, respectively. The spreading PCD symptoms were enhanced in *wrky54* mutants, *nac4-1/wrky54-2* double mutant plants. These results suggest that NAC4 promotes hypersensitive cell death by suppressing its target genes acts as negative regulator of cell death, and this immune process is fine-tuned by the negative action of *miR164*.

***Corresponding Author email :** omkim@korea.ac.kr

[GM8]

Generation of healthy low-phytate plants by overexpressing Gle1 variants

Ho-Seok Lee[1], Du-Hwa Lee[1], Il Yeong Choi[1], Hyun-Sook Pai*

[1]*Department of Systems Biology, Yonsei University, Seoul 120-749, Korea*

Myo-inositol-1,2,3,4,5,6-hexakisphosphate (InsP6), also known as phytic acid, accumulates in large quantities in plant seeds, serving as a phosphorus reservoir, but is an animal antinutrient and an important source of water pollution. Here, we report that Gle1 (GLFG lethal 1) in conjunction with InsP6 functions as an activator of the ATPase/RNA helicase LOS4 (low expression of osmotically responsive genes 4), which is involved in mRNA export in plants, supporting the Gle1-InsP6-Dbp5 (LOS4 homolog) paradigm proposed in yeast. Interestingly, plant Gle1 proteins have modifications in several key residues of the InsP6 binding pocket, which reduce the basicity of the surface charge. Arabidopsis thaliana Gle1 variants containing mutations that increase the basic charge of the InsP6 binding surface show increased sensitivity to InsP6 concentrations for the stimulation of LOS4 ATPase activity in vitro. Expression of the Gle1 variants with enhanced InsP6 sensitivity rescues the mRNA export defect of the ipk1 (inositol 1,3,4,5,6-pentakisphosphate 2-kinase) InsP6-deficient mutant and, furthermore, significantly improves vegetative growth, seed yield, and seed performance of the mutant. These results suggest that Gle1 is an important factor responsible for mediating InsP6 functions in plant growth and reproduction and that Gle1 variants with increased InsP6 sensitivity may be useful for engineering high-yielding low-phytate crops.

***Corresponding Author email :** hspai@yonsei.ac.kr

[GM9]

Mutation of *SPOTTED LEAF4 (SPL4)* encodinga microtubule severing protein produces reactive oxygen species (ROS) and delays leaf senescence in rice

Giha Song[1], Choon-Tak Kwon[1], Suk-Hwan Kim[1], Hee-Jong Koh[1], Gynheung An[2], Nam-Chon Paek*[1]

[1]*Department of Plant Science, Plant Genomics and Breeding Institute, and Research Institute of Agriculture and Life Science, Seoul National University, Seoul 08826, Republic of Korea2Department of Plant Molecular Systems Biotechnology, Crop Biotech Institute, Kyung Hee University, Yongin 446-701, Republic of Korea*

The mutants show autonomous lesion formation, spontaneous cell death without any pathogen attack and resistance to a pathogen are classified to lesion mimic mutants (LMMs). Through these phenotypes, LMMs were used to study the mechanisms of programmed cell death pathway and response to a pathogen. In this study, the *spotted leaf4 (spl4)* mutant which is derived from γ-ray irradiation were used to study the spontaneous cell death mechanism. It has been reported that many LMMs its encoding genes were identified but its molecular mechanism of lesion formation and pathogen resistance is still unclear. The reactive oxygen species (ROS) is the product of senescence and ROS can be found near the spots in LMMs during autonomous lesion formation in LMMs even dough it is a developmental stage. Stay-green is the phenomena of delayed senescence and this is the one character that breeders want to achieve. Also, scientists study this delayed senescence phenotype to elucidate the leaf senescence mechanism. In this study, we analyzed the rice *spl4* mutant, which shows autonomous lesion formation on leaf blades, ROS accumulation and shows the stay-green phenotype. The *spl4* locus was identified by map-based cloning. This locus encodes a putative microtubule severing protein, spastin. Our data may suggest that the malfunctioning of microtubule severing protein results in pleiotropic phenotypes of autonomous lesion formation, ROS accumulation and delayed senescence in *spl4* mutant.

***Corresponding Author email :** ncpaek@snu.ac.kr

[GM10]

Rice ZEBRA3 mutation disrupts citrate distribution in leaves and produces leaf variegation

Suk-Hwan Kim[1], Nam-Chon Paek*[1]

[1]*Department of Plant Science, Seoul National University, Seoul 08826, Korea*

Leaf variegation mutants have been reported in many species of higher plants. The leaves of these mutants have green and yellow (or white) sectors, and in most cases, this abnormal phenotype is caused by impaired chloroplast biogenesis in yellow or white sectors. Here, we showed the new type of leaf variegation mutant, *zebra3* (*z3*), which has transverse dark-green/green stripes in leaves with no yellow or white sectors. Map-based cloning revealed that Z3 encodes a protein predicted to be a CitMHS family citrate transporter. CitMHS family members in bacteria have been extensively studied as the secondary transporters which transport metal-citrate complexes, but their functional existences in eukaryotes remain controversial. To investigate whether Z3 has a function in transport of citric acids like prokaryotes, we measured citrate concentrations in dark-green and green sectors of *z3* leaves, and found that dark-green parts had highly accumulated citric acids. These results suggest that leaf variegation in *z3* leaves is involved in the differential accumulation of citric acids in leaves, possibly. Taken together, we propose that Z3 has roles in citrate transport and distribution in leaves, and is a possible candidate for CitMHS family members in eukaryotes.

***Corresponding Author email :** ncpaek@snu.ac.kr

[GM11]

A dominant negative OsKAT2 mutant delays light-induced stomatal opening and improves drought tolerance without yield penalty in rice

Yong Sang Lee[1], Seok-Jun Moon[1], Myung Ki Min[1], Jin-Ae Kim[1], Insun Yoon[1], Taekryoun Kwon[1], Beom-Gi Kim*[1]

[1]*Genetic engineering division, National Institute of Agricultural Sciences, Jeonju, South Korea*

Stomatal pores are the main gateways for water and air transport between leaves and the environment. Inward-rectifying potassium channels regulate photo-induced stomatal opening. Rice contains three inward rectifying shaker-like potassium channel proteins, OsKAT1, OsKAT2 and OsKAT3. Among these, only *OsKAT2* is specifically expressed in guard cells. Here, we investigated the functions of OsKAT2 in stomatal regulation using three dominant negative mutant proteins, OsKAT2(T235R), OsKAT2(T285A) and OsKAT2(T285D), which are altered in amino acids in the channel pore and at a phosphorylation site. Yeast complementation and patch clamp assays showed that all three mutant proteins lost channel activity. However, among plants overexpressing these mutant proteins, only plants overexpressing OsKAT2(T235R) showed significantly less water loss than the control. Moreover, overexpression of this mutant protein led to delayed photo-induced stomatal opening and increased drought tolerance. Our results indicate that OsKAT2 is an inward- rectifying shaker-like potassium channel that mainly functions in stomatal opening. Interestingly, overexpression of OsKAT2(T235R) did not cause serious defects in growth or yield in rice, suggesting that OsKAT2 is a potential target for engineering plants with improved drought tolerance without yield penalty.

***Corresponding Author email :** bgkimpeace@gmail.com

[GM12]

Misexpression of *AtTX12* Encoding a TIR Domain Induces Temperature-insensitive Growth Defects Partially Independently of EDS1

Bang Heon Lee[1], Woo Chang Han[1], Hyeon-Yong Park[1], Sang-Kee Song*[1]

[1]*Department of Biology, Chosun University, Gwangju 61452, Korea*

To identify genes regulating development of *Arabidopsis*, a tissue-specific GAL4/UAS activation tagging system was introduced into the Q2610 enhancer trap line, and a dominant mutant exhibiting stunted growth was isolated and named *defective root development 1-D* (*drd1-D*). The T-DNA insertion site that induced the *drd1-D* phenotype was located within the promoter region of *AtTX12*, which is predicted to encode a truncated nucleotide-binding leucine-rich repeat (NLR) protein, containing a Toll/interleukin-1 receptor (TIR) domain. The expression of *AtTX12* and defense-related genes was increased in the *drd1-D* background, and transgenic misexpression of *AtTX12* recapitulated the *drd1-D* phenotype. Tissue-specific misexpression of *AtTX12* led to temperature-insensitive local growth inhibition. A Petal/stamen-specific expression of *AtTX12* suppressed development of corresponding organs. Several point mutants of *AtTX11/12* indicates that TIR domain is essential for the activity. AtTX12 activity is partially dependent on EDS1, a key regulator of immunity triggered by TIR-type NLRs (TNLs) and independent of PAD4, an EDS1-interacting partner and NDR1, required for the immune responses triggered by coiled-coil-type NLRs. High-level misexpression of *AtTX12* induced EDS1-independent defense-related gene expression differently from canonical TNLs.

***Corresponding Author email :** sangkeesong@chosun.ac.kr

[GM13]

Identification of novel components involved in abscission zone development in Arabidopsis

Huikyong Cho[1], June K. Kwak*[1,2], Yuree Lee*[1]

[1]*Center for Plant Aging Research, Institute for Basic Science, Daegu 42988, Republic of KOREA,* [2]*Department of New Biology, DGIST, Daegu 42988, Republic of KOREA*

Abscission is a natural, advantageous process that plays important roles in plant development. Plant organs such as leaves, flowers, and fruits in response to developmental and environmental cues at the abscission zones (AZ) that are composed of four to six layers of cells responsible for the shedding events by releasing cell wall hydrolyzing enzymes. Signaling pathways initiated by the leucine-rich repeat receptor kinase HAESA and the peptide hormone IDA have been identified, but little is known about how the AZ is defined. To identify novel components involved in the spatio-temporal regulation of AZ, an *Arabidopsis* transgenic line harboring the β-glucuronidase (GUS) reporter driven by an AZ specific promoter was mutagenized with EMS. Approximately 10,000 M1 plants were individually harvested, and M2 screen is currently in progress by analyzing the expression pattern of GUS. This genetic screen based on alteration in GUS expression will provide an opportunity to identify novel molecular players of AZ development.

***Corresponding Author email :** yuree@ibs.re.kr

[GM14]

Studies on Arabidopsis thaliana AUTOPHAGY-RELATED PROTEIN 13 Modulation of Programmed Cell Death in Plant Immunity

Hwi Seong Jeon[1], Ohkmae K. Park*[1]

[1]*Division of Life Sciences, Korea University, Seoul 02841, Korea*

Autophagy is an evolutionarily conserved pathway for the bulk degradation of cytoplasmic components from yeast to mammalian. Pre-autophagosomal structure(PAS) is regulated by several AUTOPHAGY-RELATED PROTEIN(ATG)s for phagophore formation. PAS formation is mediated by the ATG1-ATG13 complexes. In yeast, the TOR kinase signaling pathway that regulates autophagy through the control of the phosphorylation status of ATG13 and ATG1. In plants, the ATG1-ATG13 complex functions mediate and adjust nutrient-dependent autophagic signaling. The plant immune response includes the hypersensitive response(HR), a form of programmed cell death(PCD). PCD It is known that the autophagy has an important role in the interplay between pro-death and pro-survival signaling pathways. Especially, the autophagic pathway regulates programmed cell death (PCD), and then, it functions in the suppression of spreading chlorotic cell death associated with pathogen response. It has been known that ATG13 is an essential regulatory component of autophagic initiation complex in other eukaryotes. However, the role of AtATG13 defense response has been unidentified. In this study, we investigate the function of AtATG13 in plant immunity. In *atg13* mutants, the chlorotic cell death by infection with *Pseudomonas syringae* pv. DC3000 (*avrRpm1*) was shown the different phenotypes, comparing with wild type plant. These data suggest that *ATG13* is involved in the regulation of the chlorotic cell death by infection with the avirulent bacterial pathogen in *Arabidopsis*.

***Corresponding Author email :** omkim@korea.ac.kr

[GM15]

Genome-wide identification of rice collar preferred genes using meta-expression analysis (RNA-seq) and construction of the regulatory network

Hyo-Yong Kim[1], Ki-Hong Jung*[1]

[1]*Graduate School of Biotechnology & Crop Biotech Institute, Kyung Hee University, Yongin 446-701, Republic of Korea*

Little and tiny organ, ligule is able to have an effect on whole plant. The ligule is a thin and tongue-like white membrane in some plants including Rice. In rice, the ligule works many things as acolumn, umbrella and humidifier. However, we actually don¡?t know the ligule specific gene and sequence. To effectively address this limitation, selection of useful candidate genes and identification of major regulatory factors through global approaches are necessary. So This paper used meta-expression analysis data and network analysis data from NCBI Gene Expression Omnibus about collar preferred gene to find out ligule specific gene. And identified 654 rice genes commonly differentially expressed under collar specific conditions. Gene ontology enrichment analysis show the quality and correlation about selected genes for ligule specific genes. Additionally , Regulation, Metabolism, Transcription and biotic stress overview were predicted with MapMan analysis. Using these methods, we will analyze the suitability of candidate genes. uncovering veiled information of the ligule specific genes should contribute to increase major crops production and aid additional experiment.

***Corresponding Author email :** khjung2010@khu.ac.kr

[GM16]

Crosstalk between diurnal rhythm and water stress reveals an altered primary carbon flux into soluble sugars in drought-treated rice leaves

Seo-Woo Kim[1], Sang-Kyu Lee[1], Hee-Jeong Jeong[1], Gynheung An[1], Jong-Seong Jeon*[1], Ki-Hong Jung*[1]

[1]*Graduate School of Biotechnology & Crop Biotech Institute, Kyung Hee University, Yongin, 446-701, Korea*

Plants retain rhythmic physiological responses when adapting to environmental challenges. However, possible integrations between drought conditions and those responses have not received much focus, especially regarding crop plants, and the relationship between abiotic stress and the diurnal cycle is generally not considered. Therefore, we conducted a genome-wide analysis to identify genes showing both diurnal regulation and water-deficiency response in rice (Oryza sativa). Among the 712 drought-responsive genes primary identified, 56.6% are diurnally expressed while 47.6% of the 761 that are down-regulated by drought are also diurnal. Using the -glucuronidase reporter system and qRT-PCR analyses, we validated expression patterns of two candidate genes, thereby supporting the reliability of our transcriptome data. MapMan analysis indicated that diurnal genes up-regulated by drought are closely associated with the starch-sucrose pathway while those that are down-regulated are involved in photosynthesis. We then confirmed that starch-sucrose contents and chlorophyll fluorescence are altered in a diurnal manner under drought stress, suggesting these metabolic diurnal alterations as a novel indicator to evaluate the drought response in rice leaves. We constructed a functional gene network associated with the starch-sucrose KEGG metabolic pathway for further functional studies, and also developed a regulatory pathway model that includes OsbZIP23 transcription factor.

***Corresponding Author email :** khjung2010@khu.ac.kr, jjeon@khu.ac.kr

[GM17]

Tracheophytes contain conserved orthologs of a Basic Helix-loop-helix transcription factor that modulate ROOT HAIR SPECIFIC genes

Youra Hwang[1], Hee-Seung Choi[1], Hyun-Min Cho[1], Hyung-Taeg Cho*[1]

[1]*Department of Biological Sciences, Seoul National University, Seoul 151-742, Korea*

ROOT HAIR SPECIFIC (*RHS*) genes, which contain the root hair-specific *cis*-element (RHE) in their regulatory regions, function in root hair morphogenesis. Here, we demostrate that an *Arabidopsis thaliana* basic helix-loop-helix transcription factor, ROOT HAIR DEFECTVE SIX-LIKE 4 (RSL4), directly binds to the RHE *in vitro* and *in vivo*, up-regulates *RHS* genes, and stimulates root hair formation in Arabidopsis. Orthologs of RSL4 from a eudicot (poplar, *Populus trichocarpa*), a monocot (rice, *Oryza sativa*), and a lycophyte (*Selaginella moellendorffii*) each restored root hair growth in the Arabidopsis *rsl4* mutant. In addition, the rice and *Selaginella* RSL4 orthologs bound to the RHE in *in vitro* and *in vivo* assays. The *RSL4* orthologous genes contain RHEs in their promoter regions, and RSL4 was able to bind to its own RHEs *in vivo* and amplify its own expression. This process likely provides a positive feedback loop for sustainable root hair growth. When *RSL4* and its orthologs were expressed in cells in non-root hair positions, they induced ectopic root hair growth, indicating that these genes are sufficient to specify root hair formation. Our results suggest that RSL4 mediates root hair formation by regulating *RHS* genes and that this mechanism is conserved throughout the tracheophyte (vascular plant) lineage.

***Corresponding Author email :** htcho@snu.ac.kr

[GM18]

IAA7 interacts with TPL/TPRs via its two EAR motifs to suppress auxin responses

Hee-Seung Choi[1], Min-Soo Lee[1], Ji-Hyun An[1], Hyung-Taeg Cho*[1]

[1]*Department of Biological Sciences, Seoul National University, Seoul 151-742, Korea*

In the nuclear auxin signaling pathway, AUXIN/INDOLE ACETIC ACIDs (Aux/IAAs) play as major transcriptional repressors by interacting with TOPLESS (TPL)/TPL-Related (TPR) corepressors via their ETHYLENE RESPONSE FACTORassociated Amphiphilic Repression (EAR) motif. Some *Arabidopsis thaliana* Aux/IAAs were predicted to have two EAR motifs, but the function of the 2nd EAR motif and any distinctive functions of two EAR motifs have remained to be verified. Here, we analyzed the biological and biochemical function of two EAR motifs of IAA7 by using the substitution mutant forms of the EAR motifs. In the auxin-sensitive root hair assay system, the 2nd EAR motif showed a minor, compared with the 1st one, but significant function in repression of root hair growth. In its own expression domain as well, the 2nd EAR motif played a minor repressive role in root hair growth, primary root growth, cotyledon curling, and plant dwarfism. The yeast two-hybrid assay indicates that the 2nd EAR motif also interacts with TPL/TPRs, though a stronger interaction given by the 1st one, and that the two EAR motifs interact with TPL/TPRs with different affinities. A protein pull-down analysis further supported the engagement of the 2nd EAR motif in interaction with TPL and TPR1. This study demonstrates that both EAR motifs of IAA7 play repressive roles in diverse auxin responses by interacting with TPL/TPRs with a specificity. Two functional EAR motifs of Aux/IAAs would provide an additional tool for auxin to diversify its biological influence in plants.

***Corresponding Author email :** htcho@snu.ac.kr

[GM19]

Genome-wide identification and analysis of U-box E3 ubiquitin ligase in rice

Yo-Han Yoo[1], Ki-Hong Jung*[1]

[1]*Graduate School of Biotechnology and Crop Biotech Institute, Kyung Hee University, Yongin, South Korea*

The plant U-box (PUB) protein is the E3 ligase, which serves as a member of protein complex for the protein degradation or post-translational modification. According to the recent report, 77 estimated U-box proteins were identified in rice and divided into 8 classes according to the domain configuration. We have constructed a phylogenetic tree using protein sequences for each class and integrated diverse meta-expression data to the phylogenetic tree context. Of them, abiotic stress meta-expression data and quantitative RT expression analyses implied that three, nine and six PUB genes in rice are associated with responses to drought, salinity and cold stress, respectively. Especially, OsPUB2 in class II is upregulated in both salinity and cold stress conditions, suggesting its potential roles to modulate crosstalk between both stress responses. And we confirmed that diurnal rhythms of three PUB genes were observed through qRT-PCR analyses. Interestingly, OsPUB4 disappeared rhythm in rice *gigantia (osgi)* mutants having defects in the diurnal rhythm. These results indicate that the *PUB4* gene is involved to the *OsGI*-mediating diurnal rhythm regulating mechanism. Our analysis provides basic information to improve future research on the PUB family in rice.

***Corresponding Author email :** khjung2010@khu.ac.kr

[GM20]

Identification of microRNAs and their targets in Hibicus syriacus plants by high-throughput sequencing

Taewook Kim[1], June Hyun Park[1], Sang-gil Lee[2], Soyoung Kim[1], Jihyun Kim[1], Chanseok Shin*[1,3,4]

[1]Department of Agricultural Biotechnology, Seoul National University, Seoul, 08826, Republic of Korea, [2]Program in Applied Life Chemistry, Seoul National University, Seoul, 08826, Republic of Korea, [3]Research Institute of Agriculture and Life Sciences, Seoul National University, Seoul, 08826, Republic of Korea, [4]Plant Genomics and Breeding Institute, Seoul National University, Seoul, 08826, Republic of Korea

MicroRNAs (miRNA) are essential small RNA molecules that regulate the expression of target mRNAs in plants and animals. Here, we aimed to identify miRNAs and their putative targets in *Hibiscus syriacus*, the national flower of South Korea. For this purpose, we employed high-throughput sequencing of small RNAs from four different tissues (*i.e.*, leaf, root, flower and young fruit) and identified 34 conserved and 14 novel miRNA families, many of which showed differential tissue-specific expression. In addition, we computationally predicted miRNA novel targets and validated some of them using 5' rapid amplification of cDNA ends analysis. One of the validated novel targets of hsy-miR477 was a terpene synthases, the primary genes involved in the formation of disease-resistant terpene metabolites, such as sterols and carotene. Collectively, this study provides the first reliable draft of the *Hibiscus syriacus* miRNA transcriptome that should constitute a basis for understanding the biological roles of miRNAs in *Hibiscus syriacus*.

This work was supported by the Next-Generation BioGreen 21 Program (No. PJ01101803), Rural Development Administration, Republic of Korea.

***Corresponding Author email :** cshin@snu.ac.kr

[GM21]

CRISPR/Cpf1-mediated DNA-free plant genome editing

Hyeran Kim[1], Sang-Tae Kim[1], Jahee Ryu[1], Beun-Chang Kang[1], Jin-Soo Kim[1,2], Sang-Gyu Kim[1]

[1]Center for Genome Engineering, Institute for Basic Science, 70, Yuseong-daero 1689-gil, Yuseong-gu, Daejeon 34047, South Korea, [2]Department of Chemistry, Seoul National University, Seoul 08826, South Korea

Cpf1, a type V CRISPR effector, recognizes a thymidine-rich protospacer-adjacent motif and induces cohesive double-stranded breaks at the target site guided by a single CRISPR RNA (crRNA). Here we show that Cpf1 can be used as a tool for DNA-free editing of plant genomes. We describe the delivery of recombinant Cpf1 proteins with in vitro transcribed or chemically synthesized target-specific crRNAs into protoplasts isolated from soybean and wild tobacco. Designed crRNAs are unique and do not have similar sequences (less than or equal to 3 mismatches) in the entire soybean reference genome. Targeted deep sequencing analyses show that mutations are successfully induced in FAD2 paralogues in soybean and AOC in wild tobacco. Unlike SpCas9, Cpf1 mainly induces various nucleotide deletions at target sites. No significant mutations are detected at potential off-target sites in the soybean genome. These results demonstrate that Cpf1crRNA complex is an effective DNA-free genome-editing tool for plant genome editing.

[DP1]

Functional roles of a chloroplast-targeted pentatricopeptide repeat protein PPR4 in chloroplast function and the growth and development of Arabidopsis and rice

Kwanuk Lee[1], Hunseung Kang*[1]

[1]*Department of Plant Biotechnology, College of Agriculture and Life Sciences, Chonnam National University, Gwangju 61186, Korea*

Despite increasing understanding of the role of pentatricopeptide repeat (PPR) proteins in organelle RNA metabolism, the functions of a majority of PPR proteins are still unknown. In this study, the roles of chloroplast-targeted *Arabidopsis thaliana* and rice (*Oryza sativa*) ortholog of maize PPR4, designated AtPPR4 and OsPPR4, respectively, were determined. The homozygous Arabidopsis *atppr4* and rice *osppr4* mutants were embryo-lethal and seedling-lethal 3 weeks after germination, respectively, suggesting that PPR4 is essential in the development of both dicot and monocot plants. The artificial microRNA-mediated knockdown mutants of AtPPR4 displayed pale-green, yellowish, and albino phenotypes. Importantly, the degree of abnormal phenotypes of the *atppr4* mutants was closely correlated with the downregulation level of *AtPPR4* expression. Chlorophyll content and photosynthetic activity of the *atppr4* mutants were significantly lower than in wild-type plants, and the chloroplast structure was abnormal in both Arabidopsis and rice *ppr4* mutants. PPR4 was involved in *rps12* intron splicing by directly binding to a specific sequence in *rps12* intron1a and intron1b. Notably, both AtPPR4 and OsPPR4 possessed RNA chaperone activity. Importantly, loss-of-function of AtPPR4 affected the alternative splicing of diverse nuclear genes. Taken together, these results provide compelling evidence that PPR4 plays an essential role in *rps12* intron splicing, and consequently chloroplast biogenesis and function, which is crucial for the development of both dicot and monocot plants.

***Corresponding Author email :** hskang@jnu.ac.kr

[DP2]

Small GTPase RabG3bcontrols xylem development in transgenic poplars

Na Young Kim[1], HwiSeong Jeon[1], Myoung-Hoon Lee[1], Dongsook Lee[1], Ah Ra Cha[1], Hye-Jung Lee Lee[1], Young-Im Choi[2], Seo-Kyung Yoon[2], Hyosine Lee[2], Ohkmae K. Park*[1]

[1]*School of Life Sciences and Biotechnology, Korea University, Seoul 136-701, Korea,* [2]*Department of Forest Genetic Resource, National Institute of Forest Science, 39 Onjeong-ro, Suwon 441-841, Republic of Korea*

Populus tremula var. glandulosa) overexpressing a constitutively active form of RabG3b (RabG3bCA). We observed wild type (WT) and RabG3bCA transgenic poplars in a field (Korea national institute of forest genetic resource, Suwon, 3716'N, 12701'E). We demonstrated that growth phenotype, including stem height and diameter, was grew faster RabG3bCA transgenic poplars than WT poplars. Internode number and Leaf number was also increase in RabG3bCA transgenic poplars. In particular, RabG3bCA transgenic poplars enhanced xylem development. The included part of cellulose and fiber was increased in transgenic poplars compared to WT poplars. We will plan to experiment about analyzed for its biomass composition and enzymatic digestibility after chemical pretreatment. We expected to RabG3bCA transgenic poplars have dramatically increase biofuel productivity. So we propose that RabG3bCA transgenic poplars have high commercial utilization.

***Corresponding Author email :** omkim@korea.ac.kr

[DP3]

SHORT-ROOT Controls Cell Elongation Through the Transcriptional Regulation of Cell Wall Genes in the *Arabidopsis* Hypocotyl

Souvik Dhar[1], Eun Kyung Yoon[2], Jun Lim*[1]

[1]Department of Systems Biotechnology, Konkuk University, Seoul 05029, Korea, [2]Department of Agricultural Biotechnology, National Institute of Agricultural Sciences, Jeonju, 54874, Korea

It has been well known that SHORT-ROOT (SHR) plays key roles in formative and proliferative cell divisions in *Arabidopsis* roots and shoots. However, it is unknown about its regulatory role in cell elongation. To address whether SHR is involved in the control of cell elongation, we performed detailed phenotypic analyses of the hypocotyl growth in the loss of SHR function mutants. As what we observed in their roots, *shr* mutants displayed significantly short hypocotyls. Further analysis revealed that the shorter hypocotyl phenotype of *shr* was due to reduced elongation of individual hypocotyl cells. Through genome-wide expression profiling and chromatin immunoprecipitation PCR (ChIP-PCR) experiments, we found that SHR directly regulates the expression of genes involved in cell wall modification. Intriguingly, overexpression of some of SHR-direct target genes in cell wall modification suppressed, in part, the shorter hypocotyl phenotype of *shr*. Furthermore, we found that the restriction of SHR movement from the vascular tissues to the endodermis was still able to rescue the *shr* phenotype in the hypocotyl. This finding likely uncouples the SHR regulatory role in cell elongation from that in radial patterning of the hypocotyl. Taken together, we propose that the mobile transcription factor SHR plays an important role in cell elongation via direct transcriptional regulation of cell wall modification genes in the *Arabidopsis* hypocotyl.

***Corresponding Author email :** jlim@konkuk.ac.kr

[DP4]

Molecular Genetic Analysis of the *Arabidopsis mild insensitivity to ethylene (mine)* Mutant Reveals the Crosstalk of Ethylene and Auxin in the Root

Gyuree Kim[1], Eun Kyung Yoon[2], Jun Lim*[1]

[1]Department of Systems Biotechnology, Konkuk University, Seoul 05029, Korea, [2]Department of Agricultural Biotechnology, National Institute of Agricultural Sciences, Jeonju, 55365, Korea

As sessile organisms, plants have evolved to adjust their growth and development to environmental changes. The plant hormones have been known to play important roles in the interactions between environmental and developmental signals. Here, we describe a novel mutant of Arabidopsis thaliana, named *mild insensitivity to ethylene* (*mine*), identified by its insensitivity to the ethylene precursor, ACC (1-aminocyclopropane-1-carboxylic acid), in root growth inhibition. Interestingly, the *mine* mutant showed root-specific insensitivity to ethylene in the dark. By contrast, the light-grown *mine* seeding displayed root growth retardation, even in the absence of ethylene. To date, it has been known that ethylene and auxin interact to control root growth. To investigate whether the *MINE* gene plays a role in the interaction between the two hormones in the root, we performed genetic and physiological analyses. Intriguingly, we found that ethylene-induced auxin biosynthesis is likely impaired in the *mine* root. To further understand the molecular mechanism underlying the crosstalk between ethylene and auxin in the root, we identified the *MINE* locus using TAIL-PCR, since the mutant was initially isolated in an activation-tagged population. Through genetic and genomic complementation assays, we found that the *MINE* gene encodes a pyridoxine/pyridoxamine 5'-phosphate oxidase (PDX3), which is known to be involved in the biosynthesis of auxin. Currently, we are investigating the role of MINE in the interplay between ethylene and auxin in the root.

***Corresponding Author email :** jlim@konkuk.ac.kr

[DP5]

The GRAS Transcription Factor SCARECROW-LIKE 28 is Involved in the Transcriptional Regulation of Microtubule Dynamics in the *Arabidopsis* Root

Jeong-eun Choe[1], Eun Kyung Yoon[2], Jun Lim*[1]

[1]*Department of Systems Biotechnology, Konkuk University, Seoul 05029, Korea,* [2]*Department of Agricultural Biotechnology, National Institute of Agricultural Sciences, Jeonju, 55365, Korea*

GRAS proteins belong to a plant-specific transcription factor (TF) family, named after three founding members [GIBBERELLIC ACID INSENSITIVE (GAI), REPRESSOR OF GA1-3 (RGA) and SCARECROW (SCR)]. In the *Arabidopsis* genome, a total of 33 members have been annotated, and about a half of them have been characterized in detail. Of uncharacterized GRAS TFs, we have focused on SCARECROW-LIKE 28 (SCL28), due to its tissue-specific expression in the root. To investigate its regulatory role in the root, we characterized loss-of-function mutants (*scl28*) and *SCL28* overexpression plants (*SCL28-OX*). Under standard growth conditions, we found no obvious phenotype in both *scl28* and *SCL28-OX* roots. In the presence of microtubule destabilizing drugs, we found that *scl28* and *SCL28-OX* seedlings showed opposite phenotypes in the root. For example, *scl28* displayed insensitivity to propyzamide (PPZ), whereas *SCL28-OX* seedlings were sensitive in root growth. Furthermore, in the presence of salt stress and PPZ treatment, the root growth phenotypes of *scl28* and *SCL28-OX* were more evidently discernible. We also found that SCL28 is localized in the nucleus and able to activate a marker gene in transient expression assays, suggesting that SCL28 likely acts as a transcriptional activator. To identify downstream target genes of SCL28, genome-wide transcriptome analyses are under way. Taken together, through molecular, genetic, physiological and pharmacological analyses, we have revealed a novel role of SCL28 in the transcriptional regulation of microtubule dynamics in the *Arabidopsis* root.

***Corresponding Author email :** jlim@konkuk.ac.kr

[DP6]

Modulation of the Timing of the Asymmetric Cell Divisions of the *Arabidopsis* Root Ground Tissue via H_2O_2 Homeostasis

Sejeong Jang[1], Ji Won Choi[1], Jun Lim*[1]

[1]*Department of Systems Biotechnology, Konkuk University, Seoul 05029. KOREA*

The *Arabidopsis* root ground tissue (GT) initially has two layers: endodermis (inside) and cortex (outside). Later in post-embryonic development, the endodermis undergoes additional periclinal asymmetric cell divisions (ACDs) to generate the endodermis and the new cortex, which is designated as the middle cortex (MC), due to its location between the endodermis and the pre-existing cortex. The plant hormone gibberellins (GA) and GRAS transcription factors, including SHORT-ROOT (SHR), SCARECROW (SCR), SCARECROW-LIKE 3 (SCL3), and DELLAs play key roles in the regulation of the timing and extent of MC formation. Recently, it has been shown that hydrogen peroxide (H2O2) precociously induces the formation of the MC layer in the root GT. However, the molecular interactions linking redox homeostasis, GA signaling, and the SHR/SCR module have never been established. Here, we have tried to elucidate the interconnected regulatory networks of these components in the control of MC formation. We have confirmed that H2O2 generation facilitates the formation of the MC layer in the root GT. Furthermore, we provide strong evidence that the transcriptional regulation of redox homeostasis by both hormonal and developmental pathways plays an important role in the modulation of the timing of the ACDs for MC formation. Taken together, our findings provide new insights to how redox homeostasis is achieved by the hormonal and developmental pathways for correct cell/tissue patterning in plants.

***Corresponding Author email :** jlim@konkuk.ac.kr

[DP7]

Starch is polymerized mainly from ADP-glucose synthesized from plastidic hexose phosphates in rice pollen

Sang-Kyu Lee[1], June-Seob Eom[2], Seok-Hyun Choi[1], Dae-Woo Lee[1], Jong-Seong Jeon*[1]

[1]Graduate School of Biotechnology & Crop Biotech Institute, Kyung Hee University, Yongin 17104, Korea, [2]Carnegie Institute for Science, 260 Panama Street, Stanford, CA 94305, USA

To elucidate the starch synthetic pathway and the role of this reserve in rice pollen, we characterized mutations in the plastidic phosphoglucomutase, *OspPGM,* and the plastidic large subunit of ADP-glucose (ADP-Glc) pyrophosphorylase, *OsAGPL4*. Both genes were upregulated in maturing pollen, a stage when starch begins to accumulate. Progeny analysis of self-pollinated heterozygous lines carrying the *OspPGM* mutant alleles, *osppgm-1* and *osppgm-2*, or the *OsAGPL4* mutant allele, *osagpl4-1,* as well as reciprocal crosses between wild type (WT) and heterozygotes revealed that loss of *OspPGM* or *OsAGPL4* caused male sterility, with the former condition rescued by introduction of the WT *OspPGM* gene. While iodine staining and transmission electron microscopy analyses of pollen grains from homozygous *osppgm-1* lines produced by anther culture confirmed the starch null phenotype, pollen from homozygous *osagpl4* mutant lines, *osagpl4-2* and *osagpl4-3,* generated by CRISPR/Cas system, accumulated small amounts of starch, which were sufficient to produce viable seed. Such *osagpl4* mutant pollen, however, was unable to successfully compete against WT pollen, validating the important role of this reserve in fertilization. Our results demonstrate that starch is polymerized mainly from ADP-Glc synthesized from plastidic hexose phosphates in rice pollen, and that starch is an essential requirement for successful fertilization in rice.

***Corresponding Author email :** jjeon@khu.ac.kr

[DP8]

OsWRKY42 Regulates OsMTId Expression and Reactive Oxygen Species in Rice

Kieu Vo[1], Muho Han[1], Chi-Yeol Kim[1], Jong-Seong Jeon[1]

[1]Graduate School of Biotechnology and Crop Biotech Institute, Kyung Hee University, Yongin 446-701, Korea

Reactive oxygen species (ROS) induction is a significant event for programed cell death (PCD) in response to pathogen attack. Here, we isolated a rice (*Oryza sativa* L.) WRKY gene which is highly upregulated in senescent leaves, denoted *OsWRKY42*. Analysis of *OsWRKY42*-GFP expression and its effects on transcriptional activation in maize protoplasts suggested that the OsWRKY42 protein functions as a nuclear transcriptional repressor. *OsWRKY42*-overexpressing (*OsWRKY42OX*) transgenic rice plants exhibited an early leaf senescence phenotype with accumulation of the ROS hydrogen peroxide and a reduced chlorophyll content. Expression analysis of ROS producing and scavenging genes revealed that the metallothionein genes clustered on chromosome 12, especially *OsMT1d*, were strongly repressed in *OsWRKY42OX* plants. An *OsMT1d* promoter:LUC construct was found to be repressed by *OsWRKY42* overexpression in rice protoplasts. Finally, chromatin immunoprecipitation analysis demonstrated that OsWRKY42 binds to the W-box of the *OsMT1d* promoter. Our results thus suggest that OsWRKY42 represses *OsMT1d*-mediated ROS scavenging and thereby promotes leaf senescence in rice.

***Corresponding Author email :** jjeon@khu.ac.kr

[DP9]

Brassinosteroids affect ABA action in stomatal closure in Arabidopsis

Yunmi Ha[1], Dami Yang[1], Hyun-Young Shin[1], Kyoung Hee Nam[1]

[1]*Department of Biological Sciences, Sookmyung Women's University, Seoul 04310, Republic of Korea*

In various physiological processes such as germination and cell elongation, brassinosteroids (BRs) oppositely work with abscisic acid (ABA). However, in a stomatal movement, these two plant hormones show a bit complex relationship. In addition to the known fact that ABA induces stomatal closure, in this study, we demonstrated that brassinolide (BL), the most bioactive BR, also induces stomatal closure when treated alone. However, when high concentration of BL (more than 500nM) was treated together with ABA, BL inhibited ABA-induced stomatal closure. We further demonstrated that direct treatment of H2O2 and SNP that are downstream signaling components in guard cell movement, reversed the BL effects on inhibition of ABA-induced stomatal closure, resulting in normally closed stomata upon ABA. We also showed that a BR signaling mutant, *bri1-301*, failed to inhibit ABA-induced stomatal closure in response to BL. And *BRI1*-overexpressing transgenic plants were hypersensitive to ABA in stomatal closure. These results indicated that, despite a negative roles of BL in ABA action in guard cell, BR signaling capacity is still required for the plant to keep ABA sensitivity.

***Corresponding Author email :** khnam514@sookmyung.ac.kr

[DP10]

Characterization of Three Zinc-finger Protein Genes Expressed in Root Cap Border Cells of Arabidopsis

Sang-Kee Song[1], Hyeon Woong Jang[1], Myeong Min Lee*[2]

[1]*Department of Biology, Chosun University, Gwangju 61452, Korea,* [2]*Department of Systems Biology, Yonsei University, 50 Yonsei-Ro, Seodaemun-Gu, Seoul 120-749, Korea*

To identify genes regulating the root development of Arabidopsis, the *UAS* activation tags were introduced in the background of *Q2610* where GAL4-VP16 is highly expressed around the root tip. A dominant mutant developing short and twisted root together with compromised epidermal patterning was isolated and named as *defective root development 2-D* (*drd2-D*). The T-DNA insertion site was localized in the promoter region of a zinc finger (ZF) protein gene by the TAIL-PCR analysis. The *drd2-D* phenotype was recapitulated by the ectopic expression of the *ZF* gene under the regulation of 5x *UAS* promoter in the *Q2610* background. The ectopic expression of *ZF* gene under the regulation of enhancers such as *J0571* suppressed the growth of above-ground organs as well. The *ZF* gene expression was specifically found in the border cells of root cap and this expression pattern was independent of SOMBRERO, an important root cap regulator. The *ZF* gene expression was also observed in the tissues undergoing maturation including cortex, vasculature, and hydathodes. Two related *ZF* genes exhibited expression patterns similar to the *ZF* gene and their misexpression led to growth defects. These results suggest that these *ZF* genes might play roles in the maturation of undifferentiated cells redundantly.

***Corresponding Author email :** mmlee@yonsei.ac.kr

[DP11]

Characterization of Weak Shoot Defective Mutants in the Background of *poltergeist* Mutant in Arabidopsis

Young Bin Yun[1], Yo Han Kim[1], Hyeon-Yong Park[1], Sang-Kee Song*[1]

[1]*Department of Biology, Chosun University, Gwangju 501-759, Korea*

Partial loss-of-function mutants might be used for understanding the fine functions of a gene of which a null mutation leads to severe phenotypes such as lack of organ development or seedling lethality. *poltergeist* (*pol*) mutant provide a proper genetic background to screen for such weak or partial loss-of-function mutants possessing minute defects in shoot meristem hardly found in the wild-type background. Here, two weak mutants have been isolated in the background of *pol-6* by the introduction of activation tags or by genetic crosses with putative weak T-DNA insertion mutant alleles. *filamentous gynoecium-1* (*fig-1*) mutant raised from the introduction of 35S enhancers in *pol-6* is a recessive mutant developing sporadic filament-like gynoecium. The T-DNA insertion site leading to *fig-1* was localized in the promoter region of a homeodomain transcription factor gene by a next generation sequencing analysis. Another mutant possessing a T-DNA insertion close to the promoter region of the gene exhibited a *fig-1* related phenotype. Together, *pol-like1* (*pll1*) mutants possessing a T-DNA insertion in the promoter region of *PLL1* were screened and crossed to *pol-6* and new *pol-6* and *pll1-2~pll1-4* double mutant combinations were isolated. Among them *pol-6 pll1-4* exhibited very weak phenotype thereby developed above-ground organs spontaneously without grafting unlikely to strong double mutants such as *pol-1 pll1-1* and *pol-6 pll1-1* displaying seedling lethality. The mutation site of *pll1-4* leading to weak double mutant phenotype was determined. These results suggest that *pol* mutant would provide a proper background for the screening of weak mutants of genes involved in shoot meristem development.

***Corresponding Author email :** drakblue@naver.com

[DP12]

IPS (IRREGULAR PHYLLOTAXIS) Encoding a SPC24 Homolog, a Member of the NDC80 Kinetochore Complex, Affects Development by Cell Division Regulation in Arabidopsis

Jinwoo Shin[1], Goowon Jeong[1], Ilha Lee*[1]

[1]*National Leading Research Laboratory of Plant Developmental Genetics, School of Biological Sciences, Seoul National University, Seoul 151-742, Korea*

A kinetochore, a protein structure on the centromere of the chromosome, mainly functions in chromosome movement during cell division for providing attachment sites of microtubules. The four-protein NDC80 complex (NDC80, NUF2, SPC24 and SPC25) in outer kinetochore has a critical role in chromosome alignment and segregation. Even though the NDC80 complex is well conserved from yeast to human, functional studies were not reported in Arabidopsis. Here, we characterized a recessive T-DNA insertion mutant, *ips-1 (irregular phyllotaxis-1),* which showed irregular phyllotatic pattern and ectopic development of SAM by up-regulation of meristem identity genes. We cloned the gene and found that *IPS* is a *SPC24* ortholog. *ips-1* showed growth retardation, defects in embryo development, phragmoplast formation and DNA aneuploidy. *IPS* was ubiquitously expressed and co-localized in centromere with CENH3. *ips-1* seemed to be a weak allele, thus we made null mutants by TALEN and CRISPR/Cas9 and confirmed the zygotic embryo-lethal phenotype like *nuf2-1*. Also IPS interacted with other NDC80 components in yeast two hybrid assay and BiFC, we expected that NDC80 complex could function in Arabidopsis as similar way in other organisms with few differences. This study provides cornerstones of studying the differences between plants and animal kinetochore related cell division.

***Corresponding Author email :** ilhalee@snu.ac.kr

[DP13]

Screening transcriptional regulators of *VIN3*, a gene required for sensing long-term winter cold for flowering in *Arabidopsis*

Goowon Jeong[1], Jinwoo Shin[1], Ilha Lee*[1]

[1]*National Research Laboratory of Plant Developmental Genetics, Department of Biological sciences, Seoul National University, Seoul 151-742, Korea*

Vernalization is one of the most important mechanisms to determine the timing of flowering in plants. In Arabidopsis, PRC2-mediated repression of strong floral repressor gene *FLOWERING LOCUS C* (*FLC*) is critical for vernalization. PHD finger domain protein VERNALIZATION INSENSITIVE3 (VIN3) was reported as one of the key component in PRC2-mediated *FLC* repression. When plant is exposed to vernalization, expression of *VIN3* gradually elevates according to period. However, how plant perceives long term cold is not known yet. To reveal upstream components of *VIN3*, we generated a transgenic line with the minimal promoter of *VIN3* fused to *GUS* reporter gene and performed EMS mutagenesis. We have isolated several mutants which show increased or decreased expression of *VIN3* after vernalization treatment. One of these mutants, *hyperactivation of VIN3 1 (hov1)* shows increased expression of *VIN3* compared to wild type. When vernalization is saturated, however, hov1 shows similar expression level of *VIN3* compared to wild type. We cloned *HOV1* by positional cloning combined with whole-genome resequencing, and introduction of *HOV1* transgene to *hov1* rescued its mutant expression of *VIN3*. Based on these result, *HOV1* seems to act as a transcriptional repressor of *VIN3* in normal condition.

***Corresponding Author email :** ilhalee@snu.ac.kr

[DP14]

Exploring genetic diversity among rice germplasms based on their physiological trait responses to salinity

Bheema Lingeswar Reddy Inja Naga[1], Sung-Mi Kim[1], In Sun Yoon[1], Taek-Ryoun Kwon*[1]

[1]*Genetic engineering division, National Institute of Agricultural Sciences, Jeonju, South Korea*

Salinity is one of the major abiotic stresses that severely affect crop production throughout the world; especially rice plant which is generally categorized as a typical glycophyte as it cannot grow in the presence of salinity. Previous studies on rice plants have focused on salt tolerance mechanisms; but there are not many studies which have focused on studying diversity among germplasms based on specific physiological traits for salt tolerance. Most diversity studies for salt tolerance in rice have been done with limited number of germplasm accessions. This study has systematically analyzed phenotypic data of 191 germplasms in two different (moderate and high) salt concentrations apart from control (without NaCl supplement) to check different salt tolerance mechanisms and also to identify better germplasms in response to salinity. We have focused our diversity study based on physiological traits such as sodium concentration, potassium concentration, K^+/Na^+ ratio which are reported to be major components determining salt tolerance. This study also shows the correlation among various phenotypic traits. This study identified germplasms which can perform better in the presence of salinity based on single trait and also combination of different physiological traits. The salt tolerant germplasm can be taken forward into developing better varieties by conventional breeding and exploring genes for salt tolerance.

***Corresponding Author email :** trkwon@korea.kr

[DP15]

Epigenetic regulation of miR820-mediated drought stress response in rice

So Young Park[1], Hyeon-Chul Park[1], Dong-Hoon Jeong*[1]

[1]*Department of Life Science and Multidisciplinary Genome Institute, Hallym University, Chuncheon, 24252, Republic of Korea*

Epigenetic regulation has been implicated in the plant development and stress responses. The underlying mechanisms of epigenetic regulation include DNA methylation, histone modification, and non-coding RNA-mediated regulation of gene expression. Of these, non-coding small RNAs, including microRNAs and small interfering RNAs, play a crucial role in negative regulation of gene expression at both transcriptional and posttranscriptional levels. In rice, miR820 has been known to be down-regulated by drought stress. It targets *OsDRM2*, which is involved in *de novo* DNA methylation of CG and non-CG sequences in the rice genome through a RNA-dependent DNA methylation mechanism. In this study, we identified a target-mimic of miR820, *ONAC300*, which is up-regulated by drought stress and may regulate the miR820 expression. To explore the function of miR820 during drought stress, transgenic rice plants over-expressing miR820 was generated. The transgenic plants exhibited drought-resistant phenotype compared with wild type plants. In addition, several transposable elements, including *RIRE7*, *CACTA* and *Tos17*, were up-regulated in these transgenic plants. These results might be due to down-regulation of *OsDRM2*, which is responsible for the suppression of those transposable elements. Possible roles of these epigenetic regulation by miR820, *OsDRM2*, and *ONAC300* as well as their agricultural impacts on drought stress resistance will be discussed.

***Corresponding Author email :** dhjeong73@hallym.ac.kr

[DP16]

MicroRNA-mediated regulation of floral organ identity in rice

Hyojin Heo[1], Dong-Hoon Jeong*[1]

[1]*Department of Life Science and Multidisciplinary Genome Institute, Hallym University, Chuncheon, 24252, Republic of Korea*

MicroRNAs (miRNAs) are non-coding small RNAs that regulate target gene expression at the posttranscriptional level. Plant miRNAs have been identified that contribute to a variety of biological process, including flower development. Functional studies examining Arabidopsis miRNAs have identified several miRNAs that play crucial roles in flower development by repressing key regulatory genes in floral organ identity. Although these miRNAs are well conserved in rice, their role in rice flower development had not been well characterized. To better understand the role of rice miRNAs in flower development, small RNA libraries were constructed from rice floral organs including glume, palea/lemma, lodicule, anther, and pistil. We also constructed PARE (Parallel Analysis of RNA Ends) degradome libraries from anther and pistil. Both small RNA and PARE libraries were deeply sequenced by Illumina technology and analyzed to identify differentially regulated miRNAs and to investigate their target cleavage functions. From the small RNA libraries, we have identified the floral-organ preferentially expressed miRNAs. In addition, analysis of PARE data revealed the cleavages of target RNAs in the particular floral organ where these miRNAs are expressed. Of floral-organ preferentially expressed miRNAs, miR5179, which targets an *OsMADS16* that is involved in anther identity, was pistil-preferentially expressed. To determine the biological function of miR5179, transgenic rice plants over-expressing miR5179 were generated. The transgenic plants exhibited the multiple-pistil phenotypes, which is due to down-regulation of *OsMADS16*. Additional studies are underway to investigate the association of floral-organ-preferential miRNAs with the expression of their target genes.

***Corresponding Author email :** dhjeong73@hallym.ac.kr

[DP17]

Reduced chromatin occupancy of CURLY LEAF-containing POLYCOMB REPRESSIVE COMPLEX2 in the *accelerated flowering1-1D mutant of Arabidopsis thaliana*

Inhye Lee[1], Taejoon Kwak[1], Sumin Lee[1], Moon-Soo Soh*[1]

[1]*Division of Integrative Bioscience and Biotechnology, Sejong University, 98, Gunja-Dong, Gwangjin-Gu, Seoul, Republic of Korea, 143-747*

Polycomb repressive complex2 (PRC2) mediates deposition of H3K23me3, a repressive histone mark in the euchromatic region. In plants, CURLY LEAF (CLF) and SWINGER encode partially redundant histone methyltransferases, being a component of PRC2, which modify histone methylation states. Although it is well characterized that PRC2 functions in various developmental and environmental contexts, little information about how it is regulated is available. Recently, we identified a genetic mutant in Arabidopsis, *accelerated flowering1-1D* (*afl1-1D*) that exhibits pleiotropic developmental defects associated with altered H3K27me3, reminiscent of *clf* mutant. The phenotypic similarity led us to hypothesize that *afl1-1D* may impact on the CLF activity. We found that *afl1-1D* mutation did not alter the transcript level of CLF. Nuclear expression of CLF-GFP fusion protein was also normal in *afl1-1D* mutant. Interestingly, the chromatin occupancy of CLF was reduced in the *afl1-1D* mutant, compared to wildtype. The defect of recruiting PRC2 were found in various chromatins, suggesting that *afl1-1D* might represent a component regulating chromatin recruitment of PRC2 in a non-gene-specific way. Molecular cloning of *afl1-1D* would reveal a novel mode of regulation of plant PRC2, thereby providing insight into how epigenetic regulation is controlled.

[DP18]

Promotion of plant growth by Fe chelating crab shell

Ji Young Hwang[1], Ju-sung Oh[2], Sang Eun Jun[1], Jin Young Moon[1], Nam Jo Park[3], Eun Ju Sohn[3], Gyung-Tae Kim*[1]

[1]*Department of Molecular Biotechnology, Dong-A university, Busan 604-714, Korea2Dept. of Genetic Engineering, Dong-A university, Busan 604-714, Korea3Bioaaplications Inc., Pohang 604-714, Korea*

This study presents promotion of plant growth and yield by a new fertilizer using crab shell powder (CSP) and iron oxide derived from spent pickling liquor. CSP consisting of organic material as chitin and spent pickling liquor containing iron oxide are considered as useful bio-materials because of its positive effects and cost. Fe is one of major elements involved in photosynthesis chlorophyll synthesis, and oxidation-reduction process. Therefore, we sought a new application of CSP and spent pickling liquor on plant cultivation. To investigate the effect of CSP and iron oxide on crop growth, we performed chelation of Fe^{2+} of iron oxide on CSP and Fe-chelated CSP was treated in soil. We analyzed the leaf growth of lettuce in soil containing Fe-chelating CSP. As results, the number, length, width, and weight of leaves in Fe chelating CSP groups increased up to 54%, 18%, 28%, and 183%, compared to an untreated group, respectively. These results indicate that growth of lettuce is promoted by synergic effect of crab shell and chelated Fe. Taken together, we will provide effects of CSP chelating Fe^{2+} of iron oxide on growth and yield of crops as a fertilizer.

***Corresponding Author email :** kimgt@donga.ac.kr

[DP19]

Elongator, an evolutionary conserved complex regulates leaf patterning and cell proliferation in Arabidopsis leaves

Sang Eun Jun[1], Ji Young Hwang[1], Jin Young Moon, Kiu-Hyung Cho, Raffael Schaffrath, John Bowman, Gyung-tae Kim*[1]

[1]Division of Molecular Biotechnology, Dong-A University, Busan 604-714, South Korea, [2]Gyeongbuk Institute for Bio industry, Andong, 760-380, South Korea, [3]Institut fur Biologie, FG Mikrobiologie, Universitat Kassel, 34132 Kassel, Germany, [4]School of Biological Sciences, Monash University, Melbourne, Victoria 3800, Australia

Elongator (ELP) complex was initially identified as a transcription elongation factor of RNA polymerase II. Later, Elongator has been also reported to be involved in histone acetylation and tRNA modification. Elongator complex consists of six subunits, which form the core complex (ELP1-ELP3) and a second module (ELP4-ELP6), and an associated protein (DRL1/KTI12). Arabidopsis Elongator proteins have been reported to possess evolutionarily conserved domain. In our previous study, loss-of-function mutant of Arabidopsis DRL1 showed defective SAM formation and adaxial leaf patterning. However, the evolutionary conserved function of transcription elongation between plants and other eukaryotes is still unclear. To determine the evolutionally conserved functions of ELP and DRL1 of Arabidopsis, we performed a binding analysis between each ELP subunits and its associated protein DRL1. In addition, we also performed functional study with Arabidopsis gene expression in yeast ELP/TOT-deficient mutants. Our comparative analysis of the function between Arabidopsis and yeast provides insight into the roles of Elongator in the establishment of SAM and leaf polarity.

***Corresponding Author email :** kimgt@donga.ac.kr

[ST1]

Understanding the signaling from the cell wall to the nucleus using an expansin-induced cell wall modification system

Hyodong Lee[1], Hyung-Taeg Cho*[1]

[1]*Department of Biological sciences, Seoul National University, Seoul 151-742, Korea (Republic of)*

A plant cell is surrounded by the cell wall which consists of cellulose microfibrils, matrix polysaccharides and minor proteins and play both mechanical support and restraint for the cell. For a plant cell to proceed division, expansion, and differentiation, intimate interactions between the cell wall and the protoplast would be required. Diverse cell wall-associated transmembrane kinases are indicative of the presence of the interface for these interactions. Mechanical changes in the cell wall, via this interface, would cause diverse downstream cytoplasmic or nuclear events for the cellular processes. Among many cell wall-modifying proteins, expansins are unique in that they reassemble the cell wall without apparent hydrolytic activity and cause cell expansion. We have adopted this expansin-mediated cell-wall modification to identify the events downstream of mechanical cell wall changes. In this study, we have expressed several types of expansin proteins by the glucocorticoid-inducible system in Arabidopsis seedlings and analyzed the time-dependent transcriptome changes. This analysis would give a starting clue to understand the mechanism from cell-wall dynamics to cellular events.

***Corresponding Author email :** htcho@snu.ac.kr

[ST2]

Brassionosteroid-regulated RLCK induce ABA-mediated stomatal closure in *Arabidopsis thaliana*

Tae-Woo Kim[1], Eun-Ji Kim[1], Tae-Ki Park[1], Se-Hwa Lee[1], So-Hee Kim[1], Tae-Wuk Kim*[1]

[1]*Department of Life Science, College of Natural Science, Hanyang University, Seoul 133-791, Korea*

Stomata are epidermal pores that regulate gas exchange and water evaporation in plats. The stomatal movement is tightly regulated by environmental stimuli and abscisic acid. Recent studies demonstrated that brassinosteroid (BR) regulates stomatal development. Here we show that BR also positively regulate stomatal closing in *Arabidopsis* leaves. We found that BR treatment induces the stomatal closing. In ABA-induced stomatal closing assay, BR enhanced ABA activity for stomatal closing. And we examine that CDL1 (CDG1-Like 1: CDL1) mutant showing reduced sensitivity to both ABA and BR in stomatal closing assay. CDL1 is member of RLCK families which mediate BR signaling. The *cdl1* knock out mutant is less sensitive to both ABA-induced and BR-induced stomatal closing. Our results suggest that CDL1 positively regulates the stomatal closing through the crosstalk between BR and ABA signaling.

***Corresponding Author email :** twgibio@hanyang.ac.kr

[ETM1]

Functional analysis of chloroplast-targeted DEAD-box RNA helicases in rice under abiotic stresses

Ghazala Nawaz[1], Hunseung Kang*[1]

[1]*Department of Plant Biotechnology, College of Agriculture and Life Sciences, Chonnam National University, Gwangju 61186, Korea*

Although many chloroplast-targeted DEAD-box RNA helicases (RH) have been determined to play essential roles in *Arabidopsis thaliana*, their functions in crop species, including rice (*Oryza sativa*), are largely unknown. To get insights into the roles of chloroplast-targeted DEAD-box RH in rice, we carried out a genome-wide analysis of RH present in rice and found that the rice genome harbors 12 RHs that contain a putative chloroplast transit peptide. The expression levels of rice RH were modulated by abiotic stresses, including cold, drought, UV, and high salinity, and upon ABA application. To determine the functional roles of RH under various abiotic stresses, rice OsRH53-expressing transgenic *Arabidopsis* plants were generated, and their phenotypes were analyzed. Expression of a rice OsRH53 delayed *Arabidopsis* seed germination by upregulating the expression of *SEEDSP* and LEAP, and inhibited *Arabidopsis* seedling growth under salt or dehydration stress. Moreover, OsRH53 inhibited cotyledon greening and seedling growth of the transgenic *Arabidopsis* plants in the presence of ABA by upregulating ABA signaling-related gene ABI3 and ABI4. Taken together, these results suggest that chloroplast-targeted OsRH53 plays important roles in plant responses to abiotic stresses and ABA.

***Corresponding Author email :** hskang@jnu.ac.kr

[ETM2]

Study on the plant immunity-improving activity of Cu2O

Younghoon Ko[1], Chian Kwon*[1]

[1]*Department of Molecular Biology, Dankook University, Cheonan 31116, Korea*

Plants possess the immune system to protect themselves from the attack of potential pathogens. This plant immunity begins with the detection of a pathogen and terminates with the secretion of immune molecules. Since the pathogen-driven disease occurrence in plants results in severe decrease of crop productivity, it is critical to find an immune-elevating agent that can help plants to better survive and produce. Silver and copper ions have long been used for inhibiting bacterial growth. In this study, the anti-bacterial activity of Cu_2O crystals was examined, based on the previous report that a Cu_2O crystal inhibits the growth of *E. coli*. The 6-facet Cu_2O crystal significantly inhibits the growth of the virulent *Pseudomonas syringae* DC3000 like its effect on E. coli. Interestingly, the 6-facet Cu_2O crystal can also induce the expression of PR1 gene which is a marker gene for plant immune responses. Therefore, it is suggested that the dual activity of Cu_2O crystal in both inhibiting bacterial growth and inducing plant immunity can help to develop a better disease-controlling agent, which may lead to contributing in reducing crop loss by pathogens.

***Corresponding Author email :** chiank@dankook.ac.kr

[ETM3]

Study on an immune function of a protein kinase JINK in Arabidopsis

Eunyoung Lee[1], Chian Kwon*[1]

[1]*Department of Molecular Biology, Dankook University, Cheonan 31116, Korea*

In nature, plants are continuously exposed to many potential pathogens. As sessile, plants entirely depend on innate immunity to defend against pathogens. Plants detect a pathogen by recognizing a pathogen-specific molecule called PAMP (pathogen-associated molecular pattern) using PRR (pattern-recognition receptor). Although many more PAMPs are present in a pathogen, only a limited number of PRRs have been characterized so far in plants. Here, the possible function a new protein kinase JINK (joy in GSRK signaling) was studied in GSRK signaling, which induces late immunity by recognizing -glucan in the virulent *P. syringae* bacterium. *JINK* was found to be mostly co-expressed with *GSRK* in a co-expression database. Activity comparison between normal JINK and the kinase-dead form, in which the nucleotide-binding site was mutated, indicated that JINK is a true protein kinase with autophosphorylation activity. The GST pull-down assay using purified recombinant proteins of GST-GSRK and HA-JINK revealed that GSRK and JINK directly interact at least in vitro. To understand the biological function of JINK, three T-DNA insertion mutant lines, *jink*-1, -2 and 3 were isolated. In addition, to understand the role of JINK in GSRK signaling, transgenic plants overexpressing JINK in either *jink* or *gsk1* mutant plants were also generated.

***Corresponding Author email :** chiank@dankook.ac.kr

[ETM4]

Transcriptome Analysis of *Ranunculus trichophyllus* (Ranunculaceae), an Amphibious Plant

Jinseul Kyung[1], Juhyun Kim[1], Ilha Lee*[1]

[1]*School of Biological Sciences, Seoul National University, Seoul 08826, Korea*

R. trichophyllus var. kadzuensisis is an amphibious plant widely distributed in Korea. The plant inhabits both on land and in the water. When grown under water, the plant develops thin and cylindrical leaves which are called 'aquatic leaves' whereas, on land, the plant produces thick and broad leaves which are called 'terrestrial leaves' Comparative phylogenetic study suggested that the terrestrial ancestor of this specie re-adapted to the aquatic environment. To elucidate the re-adaptation mechanism of the plant, we characterized the transcriptome signatures of *R. trichophyllus*. We performed *de novo* RNA sequencing of 8 libraries, including aquatic leaves, terrestrial leaves, shortly submerged leaves and ethylene-treated leaves which imitates aquatic leaf development. As a result, total 210,686 contigs were sequenced and 12,950 contigs showed differential expression between the libraries. Gene Ontology analysis suggested that several GO terms including 'response to stress', 'transport', 'secondary metabolic pathway', 'homeostasis' are over-represented in aquatic and ethylene-treated leaves, compared to the terrestrial leaves. In contrast, GO terms including 'oxygen binding' and 'transcription factor activity' were significantly over-represented in terrestrial leaves. In addition, expression of stomatal genes, wax biosynthesis genes, transporter genes and leaf development genes were significantly altered in aquatic and ethylene-treated leaves. This result of the transcriptome analysis would provide a clue to the molecular events underlying re-adaptation of plants to aquatic environment.

***Corresponding Author email :** ilhalee@snu.ac.kr

[ETM5]

Evolutionary Change in Hypoxia Response Induces Heterophylly of an Amphibious Plant *Ranunculus trichophyllus*

Myeongjune Jeon[1], Juhyun Kim[1], Ilha Lee*[1]

[1]*School of Biological Sciences, Seoul National University, Seoul 08826, Korea*

Heterophylly, an alteration of leaf form, appears in several plants belong to Ranunculaceae family. Since heterophylly is a characteristic of amphibious plants which have terrestrial ancestor, it has been suggested that heterophylly is an adaptive trait gained in the process of re-adaptation to aquatic habitat. However, what kinds of evolutionary changes cause such phenomenon is largely unknown. Here we report the involvement of hypoxia sensing mechanism for heterophylly in *Ranunculus trichophyllus*, an amphibious plant. *R. trichophyllus* develops cylindrical leaves under water but broad leaves on land. If the oxygen level decreases, the plant changes its leaf shape to aquatic form. Similar to heterophylly induced by submergence, hypoxia-induced leaf form is thought to be the consequence of expressional change in the leaf polarity genes. Some of *KANADI* genes, which mark abaxial development of leaf, were upregulated while an ortholog of *HD-ZIP III* was downregulated in hypoxia. We found that HRPE (hypoxia responsive promoter element) exists on the promoters of *R. trichophyllus KANADI*s. In contrast, the promoters of *KANADI* genes of *Ranunculus sceleratus*, a close terrestrial relative of *R. trichophyllus*, do not have HRPE. This suggests that the evolution of *cis* regulatory elements which alters the range of hypoxia response might explain part of heterophyllous nature of *R. trichophyllus*.

***Corresponding Author email :** ilhalee@snu.ac.kr

[PB1]

Characterization of MRF translation regulatory factors in Arabidopsis

Du-Hwa Lee[1], Seung Jun Park[1], Hyun-Sook Pai*[1]

[1]*Department of Systems biology, Yonsei University, Seoul 03722, Korea*

Plants adjust translation activity depending on cellular energy status, particularly photosynthetic produced sugar level. Target of rapamycin (TOR) is a master regulator of protein synthesis in response to energy/nutrient availability through modulating massive transcription network and phosphorylation of translation regulating components. Although translational regulatory mechanisms of TOR signaling in mammal and yeast were revealed in detail, it was poorly understood in plants. Here, we identified novel downstream target of TOR pathway in *Arabidopsis, MRF* (MA3 domain-containing translation regulatory factor) family genes. *MRFs* were transcriptionally induced by dark and starvation (DS) that was modulated by TOR and only MRF1 phosphorylated *in vivo*, and phosphorylated by S6 kinase1 (S6K1) and S6K2 *in vitro*. We also investigate unknown function of MRF family proteins. MRFs were co-sedimented with ribosomes and interacted with translation initiation factor eIF4A with differential affinity depending on cellular energy status. ^{35}S-methionine labeling suggested decreased and increased nascent protein synthesis in the *MRF* silenced and *MRF1* overexpressed seedlings, respectively, under DS conditions. Furthermore, ribosome association of MRF1 was modulated by the cellular energy status and by the TOR pathway through phosphorylation. These results suggest that MRFs positively modulate translation, especially under energy-deficient conditions, and that MRFs are controlled by the TOR pathway.

***Corresponding Author email :** hspai@yonsei.ac.kr

[PB2]

Mechanisms of cell cycle defects caused by depletion of PeBoW ribosome assembly factors

Seung Jun Park[1], Chang Sook Ahn[1], Du-Hwa Lee[1], Hyun-Sook Pai*[1]

[1]*Department of Systems biology, Yonsei University, Seoul 03722, Korea*

The nucleolar protein pescadillo (PES) controls biogenesis of the 60S ribosomal subunit through functional interactions with Block of Proliferation 1 (BOP1) and WD Repeat Domain 12 (WDR12) in plants. Here, we determined molecular functions of BOP1 and WDR12, and characterized defects in plant cell growth and proliferation caused by a deficiency of PeBoW (PES-BOP1-WDR12) proteins. When BOP1 and WDR12 level was reduced, plants showed developmental arrest and premature senescence phenotypes, similar to *PES* RNAi. Both the N-terminal domain and WD40 repeats of BOP1 and WDR12 were critical for specific associations with 60S/80S ribosomes. In response to nucleolar stress or DNA damage, PeBoW proteins moved from the nucleolus to the nucleoplasm. Leaf growth kinematic analyses revealed that depletion of PeBoW proteins led to significantly suppressed cell proliferation, cell expansion, and epidermal pavement cell differentiation. It also resulted in reduced CDKA activity, causing reduced phosphorylation of histone H1 and retinoblastoma-related (RBR) protein. *PeBoW* silencing caused rapid transcriptional modulation of cell cycle genes, including reduction of *E2Fa* and Cyclin D family genes, and induction of several *KRP* genes, accompanied by down-regulation of auxin-related genes and up-regulation of jasmonic acid-related genes. Taken together, these results suggest that the PeBoW proteins involved in ribosome biogenesis play a critical role in plant cell growth and survival, and their depletion leads to inhibition of cell cycle progression, possibly modulated by phytohormone signaling.

***Corresponding Author email :** hspai@yonsei.ac.kr