Horticulture Research

# 12th IHRC 2025

## INTERNATIONAL HORTICULTURE RESEARCH CONFERENCE

# Abstracts

SANYA, CHINA
OCTOBER 9-12, 2025





**Origin and evolution of floral scent biosynthesis**

Feng Chen

*Department of Plant Sciences, University of Tennessee*

E-mail: fengc@utk.edu

## Abstract

A central hypothesis for explaining the extraordinary evolutionary success and vast radiation of flowering plants is the establishment of mutualistic relationships between flowers and their insect pollinators. Among the many floral traits that facilitate these interactions, floral scent stands out as a critical chemical cue, enabling insects to recognize, locate, and repeatedly visit flowers. Understanding how floral scent first originated and subsequently diversified is therefore a fascinating and fundamental question in plant evolutionary biology. One promising path toward answering this question lies in examining the earliest-diverging lineages of flowering plants, particularly the ANA grade, which includes Amborella, Nymphaeales, and Austrobaileyales. These basal groups hold unique insights into the ancestral states of floral traits. In this talk, we will approach the topic from multiple perspectives: floral scent chemistry, pollination syndromes, and the biochemical and evolutionary dynamics of specific scent-producing genes in basal angiosperms.

# Postharvest regulation of leaf senescence in Chrysanthemum

Yuna Yang[1], Shiyu Sun[1], Jiafu Jiang[1], Sumei Chen[1], Fadi Chen[1], Likai Wang[1]

[1] *State Key Laboratory of Crop Genetics & Germplasm Enhancement and Utilization, Key Laboratory of Flower Biology and Germplasm Innovation, Ministry of Agriculture and Rural Affairs, College of Horticulture, Nanjing Agricultural University, Nanjing 210095, P.R. China*

Email: wlk@njau.edu.cn

## Abstract

Leaf senescence affects crop yield and quality, as well as the ornamental and economic value of ornamental plants. Recent studies have revealed that N6-methyladenosine ($m^6A$) modification of mRNA plays an important role in plant growth, development, and stress responses. However, its molecular mechanism in regulating leaf senescence remains unclear. In this study, we found that the level of mRNA $m^6A$ modification significantly decreases during leaf senescence of cut chrysanthemum. Specifically, the reduction of $m^6A$ modification in the 3' UTR region of the *EIN2* gene leads to increased translation efficiency, thereby accelerating the leaf senescence process. In contrast, $m^6A$ modification located in the 5' UTR region induces the downstream shift of H3K4me3 histone modification, thereby inhibiting gene expression and regulating senescence. This regulatory mechanism is conserved in *Arabidopsis*, rice, and chrysanthemum. Further research demonstrates that the $m^6A$ methyltransferase MTA and the H3K4me3 methyltransferase ATX1 mediate the shift of H3K4me3 modification through dynamic competitive binding to RNA polymerase II. This study reveals a novel mechanism by which $m^6A$ modification participates in the regulation of leaf senescence through dual pathways: regulating gene translation efficiency (3' UTR region) and chromatin modification (5' UTR region). It also provides a theoretical foundation for developing new senescence-resistant germplasm and technologies for senescence regulation.

# Distinct attenuation mechanisms of jasmonate signaling in ripening fruits and wounded leaves

Lei Deng[1,2,*], Tianxia Yang[1,3], Chuanlong Sun[1,4], Qinyang Wang[1,3], Chuanyou Li[1,2,*]

[1] *Taishan Academy of Tomato Innovation, Shandong Agricultural University, Tai'an 271018, Shandong, China*

[2] *College of Life Sciences, Shandong Agricultural University, Tai'an 271018, Shandong, China*

[3] *Key Laboratory of Seed Innovation, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, Beijing 100101, China*

[4] *College of Horticulture Science and Engineering, Shandong Agricultural University, Tai'an 271018, Shandong, China*

E-mail: ldeng@sdau.edu.cn

## Abstract

Ripe fruits are more susceptible to necrotrophic pathogens than unripe fruits. Although this phenomenon is widespread across different fruit species and results in substantial economic losses, the underlying mechanism is still poorly understood. Previous studies revealed that ethylene (ET) is a key signal controlling climacteric fruit ripening and that jasmonate (JA) regulates plant resistance to necrotrophs. We investigated the function of tomato cytochrome P450 94 (*CYP94*) family genes in JA signaling and report here that ET-mediated ripening suppresses JA-mediated defense by promoting the deactivation of bioactive JA-Ile. ETHYLENE-INSENSITIVE 3 (EIN3)/EIN3-LIKE (EIL) transcription factors directly activated *CYP94C1* to convert JA-Ile to its inactive form 12-COOH-JA-Ile, thereby attenuating JA signaling during fruit ripening. Mutation of *CYP94C1* led to increased resistance of ripe fruits to the necrotrophic pathogen *Botrytis cinerea* without affecting the ripening process. Additionally, the master transcription factor MYC2 directly activated two other CYP94 members *CYP94B1* and *CYP94B2* to convert JA-Ile to its less active form 12-OH-JA-Ile, thereby attenuating JA signaling in wounded leaves. Simultaneous mutation of *CYP94B1* and *CYP94B2* increased the resistance of leaves to B. cinerea. Thus, differences in the expression and enzymatic activities of *CYP94* family gene members precisely control JA-mediated defense responses in tomato.

**Fruit Hollowness and Browning in Plum and Associated Molecular Mechanism**

Honghong Deng, Junwei Liu, Ziyang Zhang, Xi Liang, Binqi Li, Tingting Xie, Yuanyuan Wang, Shishi Luo, Aoqi Xue, Ying Yu, Faxing Chen*

*College of Horticulture, Fujian Agriculture and Forestry University*

E-mail: denghonghong2010@163.com

## Abstract

Plums, as important stone fruits, are highly valued for their rich nutritional content and associated health benefits. However, they are susceptible to a physiological disorder known as fruit hollowness and browning (HB), which severely compromises fruit quality and marketability. This study investigated HB development in 'Huangguan' plums sampled at multiple stages after full bloom. HB onset began at veraison, as confirmed by paraffin sectioning, with HB incidence reaching 37.73% at maturity. The fruit shape index was significantly higher in HB-affected plums compared to non-HB fruits. HB tissues exhibited positive staining with Saffranin O and Fast Green, and the affected area expanded during fruit development. Total lignin content increased progressively throughout fruit development and ripening, with significantly higher levels detected in HB fruits than in non-HB fruits ($P \leq 0.001$). Lignin monomers in HB fruits were primarily composed of guaiacyl lignin (22.6%) and syringyl lignin (10.8%). Differential expression of key genes involved in lignin biosynthesis suggested the presence of a complex regulatory network. Proteomic analysis identified 941 and 1,249 differentially expressed proteins between HB and non-HB fruits at the veraison and ripe stages, respectively. Functional enrichment analysis highlighted significant changes in critical metabolic pathways, especially phenylpropanoid biosynthesis (map00940), which is central to lignin production. Key transcription factors (*PsNAC43* and *PsMYB43*) were identified through weighted gene co-expression network analysis. Both factors localized to the nucleus and dual-luciferase assays showed that *PsNAC43* significantly activates the promoters of *PsMYB43* and *PsPAL1*. Transient transformation experiments further indicated that *PsNAC43* and *PsMYB43* upregulate the expression of *NtPAL1*—a key gene in the lignin biosynthesis pathway—and enhance PAL enzyme activity. In summary, lignin accumulation is closely linked to HB development in plums. *PsNAC43* and *PsMYB43* activate the expression of *PsPAL1*, elevate PAL enzyme activity, and thereby contribute to the formation of HB in plum fruit.

# Genetic and Molecular Control of Lettuce Bolting Resistance: From Gene Discovery to Breeding Applications

Guanghui An[1], Yong Wang[1], Yetong Qi[2], Wei Shao[3]

[1] *College of Horticulture, Henan Agricultural University, Zhengzhou, 450046, China*

[2] *Xianghu Laboratory, Hangzhou, 311231, China*

[3] *National Key Laboratory for Germplasm Innovation & Utilization of Horticultural Crops, College of Horticulture and Forestry Sciences, Huazhong Agricultural University, Wuhan, 430070, China;*

E-mail: agh@henau.edu.cn

## Abstract

A major challenge in lettuce production is premature bolting induced by extreme climatic conditions, particularly high temperatures, which severely compromises crop marketability. This issue also forces plant factories to maintain artificially low cultivation temperatures, resulting in substantial energy waste. Consequently, enhancing bolting resistance has become a critical breeding objective in lettuce. We have employed forward genetics to identify and clone key regulatory genes governing bolting time. Three genes were genetically cloned, *LsKN1*, *LsBLH2*, and *LsOFP6*, and their unique molecular mechanisms were elucidated. *LsKN1* encodes a knotted-1-like homeobox protein, and was activated by a CACTA-like transposon which was inserted into the first exon of *LsKN1* in the late bolting lettuce. *LsBLH2* encodes a BEL1-like homeodomain protein and has a 1-bp deletion in exon 1, which leads to a premature stop codon in the late bolting lettuce. *LsOFP6* encodes an ovate family protein and has a nonsense mutation in exon 1, which leads to a premature stop codon in the late bolting lettuce. Interestingly, these three genes exhibit genetic interaction and protein-protein interaction. *LsKN1*, *LsBLH2*, and *LsOFP6* synergistically modulate the transition from vegetative to reproductive growth by synergistically regulating gibberellin biosynthesis and signaling pathways. We have developed multiple lettuce germplasms with enhanced bolting resistance through leveraging natural variation in these genes. These advances provide a robust scientific foundation for breeding high-quality and bolting-resistant lettuce cultivars suitable for plant factory production. Furthermore, our results exemplify the power of leveraging natural genetic diversity to address key agricultural challenges.

# Integrated genomic and transcriptomic analyses reveal the genetic and molecular mechanisms underlying hawthorn peel color and seed hardness diversity

Jiaxin Meng[1], Yan Wang[1], Rongkun Guo[1], Jianyi Liu[1], Kerui Jing[1], Jiaqi Zuo[1], Yanping Yuan[2], Fengchao Jiang[1,*], Ningguang Dong[1,*]

[1] *Beijing Engineering Research Center for Deciduous Fruit Trees, Key Laboratory of Biology and Genetic Improvement of Horticultural Crops (North China), Ministry of Agriculture and Rural Affairs, Institute of Forestry and Pomology, Beijing Academy of Agriculture and Forestry Sciences, Beijing 100093, China*

[2] *College of Landscape Architecture and Arts, Northwest A&F University, Yangling, Shaanxi 712100, China*

E-mail:Jiaxin Meng: mengjiaxin@baafs.net.cn

## Abstract

Hawthorn (Crataegus pinnatifida) fruit peel color and seed hardness are key traits that significantly impact economic value. We present here the high-quality chromosome-scale genomes of two cultivars, including the hard-seed, yellow-peel C. pinnatifida “Jinruyi” (JRY) and the soft-seed, red-peel C. pinnatifida “Ruanzi” (RZ). The assembled genomes comprising 17 chromosomes were 809.1 Mb and 760.5 Mb in size, achieving scaffold N50 values of 48.5 Mb and 46.8 Mb for JRY and RZ, respectively. Comparative genomic analysis identified 3.6–3.8 million single nucleotide polymorphisms, 8.5–9.3 million insertions/deletions, and approximately 30 Mb of presence/absence variations across different hawthorn genomes. Through integrating differentially expressed genes and accumulated metabolites, we filtered candidate genes CpMYB114 and CpMYB44 associated with differences in hawthorn fruit peel color and seed hardness, respectively. Functional validation confirmed that the CpMYB114-CpANS regulates anthocyanin biosynthesis in hawthorn peels, contributing to the observed variation in peel color. CpMYB44-CpCOMT was significantly upregulated in JRY and was verified to promote lignin biosynthesis, resulting in the distinction in seed hardness. Overall, this study reveals the new insights into understanding of distinct peel pigmentation and seed hardness in hawthorn and provides an abundant resource for molecular breeding.

# Section-Wide Potato NLRome Empowers Discovery of Late Blight Disease Resistance Genes

Luyao Wang[1,8], Hongbo Li[1,5,8], Yuhang Ke[2,8], Ping Zhu[1,2], Yuying Li[1], Pei Wang[1,3], Haohao Liu[2], Yajie Li[2], Jingqi Chen[6], Shaoqun Zhou[1], Li Wan[6], Savithramma P. Dinesh-Kumar[7], Suomeng Dong[2*] & Sanwen Huang[1,4*]

[1] *National Key Laboratory of Tropical Crop Breeding, Shenzhen Branch, Guangdong Laboratory of Lingnan Modern Agriculture, Genome Analysis Laboratory of the Ministry of Agriculture and Rural Affairs, Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, China*

[2] *Department of Plant Pathology, Nanjing Agricultural University, Nanjing, China*

[3] *School of Life Sciences, Yunnan Key Laboratory of Potato Biology, Yunnan Normal University, Southwest United Graduate School, Kunming, China*

[4] *National Key Laboratory of Tropical Crop Breeding, Chinese Academy of Tropical Agricultural Sciences, Haikou, China*

[5] *College of Horticulture Science and Engineering, Shandong Agricultural University, Tai'an, China*

[6] *State Key Laboratory of Plant Trait Design, CAS Center for Excellence in Molecular Plant Sciences, Shanghai Institute of Plant Physiology and Ecology, Chinese Academy of Sciences, Shanghai, China*

[7] *Department of Plant Biology and the Genome Center, College of Biological Sciences, University of California, Davis, CA, USA*

[8] *These authors contributed equally: Luyao Wang, Hongbo Li, Yuhang Ke*

E-mail: lihongbo_solab@163.com

## Abstract

Potato late blight, caused by *Phytophthora infestans* and responsible for the Irish Potato Famine, remains a major threat to global food security[1]. Most late blight resistance (*R*) genes encode nucleotide-binding domain and leucine-rich repeat (NLR) proteins, yet many have been overcome by the rapid evolution of *P. infestans*[2]. Deploying *R* genes through hybrid potato breeding[3-7] offers an opportunity to counteract *P. infestans* evolution. Here, we construct a section-wide NLRome comprising 39,211 *NLR* genes from 31 wild and 21 cultivated potato genomes, representing *Solanum* section *Petota*, the tuber-bearing clade. This includes newly sequenced genomes of seven wild species with strong resistance to late blight. Phylogenomic analysis reveals asymmetric evolutionary patterns that may distinguish sensor and helper NLRs. Mining the NLRome results in the cloning of *Rpi-cph1*, whose ortholog was previously identified only in American black nightshade, and *Rpi-cjm1*, the first toll/interleukin-1 receptor (TIR) domain-containing NLR against late blight. Our findings show that the non-canonical NLR integrated domains (IDs) have potentially evolved from integrating pathogen effector targets[8]. We unveil the evolutionary trajectory of NLR IDs in *Petota*, leading to the discovery of *Rpi-brk1* with a heavy-metal-associated (HMA) domain. We demonstrate that the immune function of *Rpi-brk1* necessitates HMA-effector interactions, and that incorporating the HMA domain into the potato NLR R1 expands its resistance spectrum, thereby proposing a domain "plug-in" strategy to engineer disease resistance in potatoes. These findings offer a paradigm for *R* gene discovery through comparative and evolutionary genomics as well as a new strategy for *R* gene engineering.

# Pan-genome analyses facilitate grape breeding

Hua Xiao[1], Zhongjie Liu[1], Nan Wang[1], Ying Su[1], Qiming Long[1], Yanling Peng[1], Lingfei Shangguan[2], Jinggui Fang[2,3], Yongfeng Zhou[1,4]

[1] *National Key Laboratory of Tropical Crop Breeding, Shenzhen Branch, Guangdong Laboratory of Lingnan Modern Agriculture, Key Laboratory of Synthetic Biology, Ministry of Agriculture and Rural Affairs, Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, China*

[2] *College of Horticulture, Nanjing Agricultural University, Nanjing, China*

[3] *College of Horticulture, Qingdao Agricultural University, Qingdao, China*

[4] *National Key Laboratory of Tropical Crop Breeding, Tropical Crops Genetic Resources Institute, Chinese Academy of Tropical Agricultural Sciences, Haikou, China*

E-mail: xiaohua01@caas.cn

## Abstract

Write your abstract here. The abstract should be one paragraph of 300 words or less using 12pt Times New Roman font without figures or tables. The limitations inherent in detecting variations within a single reference genome hinder the identification of crucial variants associated with breeding traits and a comprehensive analysis of agronomic trait inheritance. Advanced pangenome-based approaches emphasize broader efforts aimed at discovering genetic variants in crop breeding. Here, we construct a graph-based pangenome reference, Grapepan v.1.0, utilizing 29 assemblies, including 22 haplotype-resolved assemblies. By integrating high-quality resequencing data from 466 grapevine cultivars, we develop a comprehensive variation map that includes 8,591,919 single nucleotide polymorphisms (SNPs), 513,969 insertions and deletions (InDels), and 236,449 structural variations (SVs). Integrating SVs into a genome-wide association study (GWAS), we identify 148 significant association loci for 29 agronomic traits, encompassing approximately 5.58% of the genome, of which 122 loci are newly discovered. This highlights the substantial role of SVs in the genetic regulation of multiple traits. Additionally, significant genetic correlations were detected among the 29 traits, ranging from 0.41 to 0.97. By combining variants (SVs and SNPs) identified by GWAS with genomic selection methods, the prediction accuracy for traits is significantly improved, showing at least a 16% improvement relative to previous reports for grapevines. Overall, our Grapepan v.1.0 variation map provides highly accurate and comprehensive variation data, significantly enhancing the genomic prediction for key agronomic traits, which will greatly facilitate grapevine genomic breeding.

# EccDNAs transfer between plants: a new way for gene exchange and biotechnological innovation

Aijun Zhang[1], Han Zhou[1], Xiaoyang Wan[1], Tingjin Wang[1], Liping Chen[1, *]

[1] *Department of Horticulture, College of Agriculture and Biotechnology, Zhejiang University, Hangzhou 310058, China*

E-mail: chenliping@zju.edu.cn

## Abstract

Extrachromosomal circular DNAs (eccDNAs) are derived from the plant's own genome, arising through processes such as replication errors, chromosomal rearrangements, and repair of double-strand breaks. They are highly heterogeneous in size—ranging from small gene fragments to complete functional genes—as well as in sequence composition and genomic origin. EccDNAs have been detected in a wide range of plant tissues, and growing evidence shows that they can carry complex genetic elements. In this study, using both interspecific grafting (in Brassica) and intergeneric grafting (in Solanaceae), we provide the first direct evidence that eccDNAs can move from one graft partner to another and be inherited by their progeny. The transferred eccDNAs include intact genes together with their regulatory sequences, and some are transcriptionally active in recipient tissues. Our grafting experiments demonstrate that eccDNAs can traverse the graft junction through the vascular system, thereby challenging the conventional view that grafting is restricted to the exchange of physiological signals. These findings support the notion that grafting may serve as an overlooked yet effective route for horizontal gene transfer (HGT) in plants. The dynamic properties of eccDNAs—such as reversibility, plasticity, and responsiveness to environmental cues—distinguish them from conventional transgene fragments and highlight their potential as a novel genetic vehicle in crop improvement. Moreover, eccDNAs may avoid some of the regulatory and biosafety concerns associated with transgenic and gene-edited crops. Harnessing this system could open new opportunities for incorporating traits from wild or related species, thereby contributing to the development of highly stress-tolerant and broadly adaptable agricultural systems to meet climate and resource challenges.

# Utilizing genome editing to regulate the synthesis of active ingredients in biomedicinal orchids and the development of new methods for addressing methylated targets

Zhicai Wang, Meili Zhao, Zhenyu Yang, Jian Li, Meina Wang

*Key Laboratory of National Forestry and Grassland Administration for Orchid Conservation and Utilization, the National Orchid Conservation Center of China and the Orchid Conservation & Research Center of Shenzhen, Shenzhen 518114, China*

E-mail:wangzc@cnocc.cn

## Abstract

Dendrobine is a sesquiterpene alkaloid specific to several *Dendrobium* orchids. It has gained significant attention recently due to its numerous biological functions, including antiviral, anticancer, antioxidant, and liver-protective properties. The demand for dendrobine is increasing both in research and industrial applications. However, the extraction from *Dendrobium nobile*, which contains high levels of dendrobine, remains insufficient. Therefore, there is an urgent need to diversify biological resources. *Dendrobium catenatum* is the most widely cultivated medicinal orchid. Its transformation is relatively easy, and genome sequencing has been conducted. Additionally, it has a comparatively short life cycle and produces high biomass. Many precursors for dendrobine synthesis are present, making it an ideal plant chassis for dendrobine production. Firstly, we analyzed RNA sequence data from multiple *Dendrobium* species and identified 648 differentially expressed genes. Secondly, we discovered several key genes responsible for dendrobine synthesis, including *CMEAO*. We then stacked seven key genes and coexpressed them to enhance dendrobine production. Additionally, we utilized the CRISPR-Act system to transcriptionally activate key endogenous genes to boost dendrobine synthesis. These methods achieved a 30.1% improvement in dendrobine production in transiently transformed leaves and a 35.6% increase in stable transgenic plants. Furthermore, we will report our recent discoveries related to enhanced genome editing at methylated sites.

# Integrating Multi-Dimensional Signatures: Molecular Determinants and Application of Genes Associate with Fruit Cracking in Tomato

Fangling Jiang[1], Xingchen Liu[1], Junde Li[1], Xiaojun Qian[1], Zhen Wu[1], Rong Zhou[1], Jingxuan Li[1], Xiaolong Qin[1], Xufang Liang[1]

[1] *College of horticulture, Nanjing Agricultural University, Nanjing 210095, China;*

Presenting author(s) e-mail：jfl@njau.edu.cn

## Abstract

Horticultural fruits are susceptible to cracking, a severe physiological disorder that deteriorates fruit appearance and quality, which significantly compromises their marketability and commercial value. In this research, we collected 320 tomato genotypes and analyzed cracking-related phenotypes. Utilizing multi-omics (whole transcriptome sequencing, Riob-seq etc.) as well as QTL and GWAS, genes related to cracked fruits were identified, which include hormones, cell wall, water channel proteins, reactive oxygen species, waxes associated genes, as well as transcription factors. Adopted CRISPR and related techniques, the functions of relevant genes were identified. Cracking-resistant tomatoes were also created using CRISPR as well as EMS mutation. Through germplasm innovation, efficient cultivation management, spraying growth regulatory substance and setting up physical barriers, an effective prevention and management technology for fruit cracking was finally established.

# Natural variation of AcEGY3 mediates chloroplastic ROS homeostasis to confer kiwifruit thermotolerance

Chengcheng Ling, Jun Yang, Songhu Wang*

*Anhui Province Key Laboratory of Horticultural Crop Quality Biology, School of Horticulture, Anhui Agricultural University, Hefei 230036, China;*

E-mail: wangsonghu@ahau.edu.cn

## Abstract

The *Actinidia* genus of perennial woody plants includes the popular kiwifruits consumed worldwide. Most kiwifruit species are naturally distributed in warm, moist environments but are vulnerable to high levels of heat stress. Here, our map-based cloning identifies a natural variation underlying the heat stress sensitivity differences between varieties HY0809 and MH-Z11 belonging to the respective kiwifruit species *Actinidia chinensis* and *Actinidia eriantha*. We find that the thermotolerance of HY0809 plants is caused by higher expression of *AcEGY3*, encoding a chloroplastic protein that promotes $H_2O_2$ accumulation in chloroplast, leading to the induction of stress tolerance genes, including those encoding heat shock proteins (HSPs). We show that this higher *AcEGY3* expression is driven by the heat stress-inducible AcGATA1 transcription factor that interacts with a GATA binding site in the natural variation of the *AcEGY3* promoter, which is absent in the corresponding *AeEGY3* promoter from MH-Z11 plants. AcGATA1 enhances *AcEGY3* expression via cooperative interaction with HSFA2-2, a transcription factor that mediates a base level heat stress-induction of both *AcEGY3* and *AeEGY3,* thus causing higher heat stress-induced $H_2O_2$ accumulation in HY0809. Finally, we show that this GATA binding site is also present in *EGY3* promoters of other thermotolerant kiwifruit species, as well as in thermotolerant *Actinidia eriantha* genotypic variants, indicating that it is a major contributor to the natural variation in kiwifruit thermotolerance.

# Mining Key Heat Tolerance Genes in Grapes and Their Regulatory Mechanisms

Lijun Wang[1, 2], Haiyang Chen[1, 2, 3], Haibo Liu[1, 2, 3], Xinna Liu[1, 2, 3]

[1] *State Key Laboratory of Plant Diversity and Specilaty Crops and Beijing Key Laboratory of Grape Science and Enology, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China.*

[2] *China National Botanical Garden, Beijing 100093, China*

[3] *University of Chinese Academy of Sciences, Beijing 100049, China*

E-mail: ljwang@ibcas.ac.cn

## Abstract

With the increasing challenges posed by global warming and climate change, heat stress has become a significant threat to the sustainable production of grapevines. However, the genetic basis of grapevine thermotolerance remains poorly understood. Here, we combine genome-wide association study with transcriptomic profiling to identify *TTC4* (thermotolerance on chromosome 4), a gene encoding a WRKY transcription factor, as a key determinant of thermotolerance in grapevine. TTC4 directly activates two thermotolerance related genes, *HSP18.1* and *APX3*.We also identify a heat-suppressed repressor SPL13 (SQUAMOSA-promoter binding protein-like 13) that cannot bind to the GTAT element ($TTC4^{T(7631)}$) in intron 2 of *TTC4*, but can bind to the natural variant, GTAC ($TTC4^{C(7631)}$). Grapevine accessions with $TTC4^{C/C(7631)}$ genotype exhibit significantly lower thermotolerance compared to those with the $TTC4^{T/T(7631)}$ and $TTC4^{C/T(7631)}$ genotypes. This fine-tuned regulation contributes to thermotolerance divergence among grapevine populations. In addition, we identified and characterized heat stress transcription factor HSFA2, which is a positive regulator in grape heat tolerance. The transcriptional activation activities of VdHSFA2 from heat sensitive *Vitis vinifera*) are higher than VvHSFA2 from heat tolerant *Vitis davidii*, which is associated with the variation of single amino acid (Thr315Ile) in AHA1 motif between VdHSFA2 and VvHSFA2. Moreover, VdHSFA2 can regulate more target genes than VvHSFA2. These key genes should hold significant potential as a genetic resource for breeding thermotolerant grapevine varieties.

# Plant grafting: molecular mechanisms and applications

Ming Feng[1]

[1] *College of Horticulture Science and Engineering, Shandong Agricultural University, Tai'an, China.*

E-mail: yyfengming@163.com

## Abstract

People have grafted plants since antiquity for propagation, to increase yields, and to improve stress tolerance. This report presents the mechanisms and applications of plant grafting and a comprehensive study on conifer graft healing. Here, we developed a micrografting method in conifers using young tissues that allowed quick and efficient grafting between both closely related species and distantly related genera. Conifer graft junctions rapidly connected vasculature and differentially expressed thousands of genes including auxin and cell wall-related genes. By comparing these genes to those induced during *Arabidopsis thaliana* graft formation, we found a common activation of cambium, cell division, phloem and xylem-related genes. A gene regulatory network analysis in Norway spruce (*Picea abies*) predicted that *PHYTOCHROME A SIGNAL TRANSDUCTION 1* (*PAT1*) acted as a core regulator of graft healing. This gene was strongly upregulation during both spruce and *Arabidopsis* grafting, and *Arabidopsis* mutants lacking *PAT*-genes failed to attach tissues or successfully graft. Complementing *Arabidopsis* PAT mutants with the spruce *PAT1* homolog rescued tissue attachment and enhance callus formation. Together, our data demonstrate an ability for young tissues to graft with distantly related species and identifies the PAT gene family as conserved regulators of graft healing and tissue regeneration.

# Characterizing a PHR-directed gene network uncovers key transcriptional and post-transcriptional regulators of phosphate starvation responses in tomato

Dongbo Lin[1,2], Peng Tian[1], Xiaoen Zhu[1], Zeteng Lin[1], Ziwei Li[1], Yongxia Zhang[1], Beixin Mo[1], Xuemei Chen[3,4], Tengbo Huang[1*]

[1]*Guangdong Provincial Key Laboratory for Plant Epigenetics, College of Life Sciences and Oceanography, Shenzhen University, Shenzhen, Guangdong 518071, China.*

[2] *College of Coastal Agricultural Sciences, Guangdong Ocean University, Zhanjiang, Guangdong 524088, China.*

[3] *State Key Laboratory of Protein and Plant Gene Research, School of Life Sciences, Peking-Tsinghua Joint Center for Life Sciences, Peking University, Beijing, 100871, China.*

[4] *Beijing Advanced Center of RNA Biology (BEACON), Peking University, Beijing, 100871, China.*

E-mail: tengbohuang@szu.edu.cn

## Abstract

Phosphorus (P) is an essential nutrient for plants, and its scarcity, especially in the form of soluble phosphate (Pi), induces phosphate starvation responses (PSRs). Our research delves into the molecular mechanisms that regulate the adaptation of tomato plants to Pi deficiency, highlighting the role of two PHR transcription factors, *SlPHR3* and *SlPHR4*, as central regulators of Pi metabolism and related developmental/physiological processes, particularly PSRs. Notably, our investigation into the downstream transcriptional network regulated by *SlPHR3* and *SlPHR4* leads to the discovery of previously-unidentified transcriptional and post-transcriptional regulators that may mediate the function of *SlPHR3*/*SlPHR4*. We characterized the roles of these key genes and revealed their importance in the regulation of Pi metabolism and PSRs in tomato. Our findings shed light on understanding the adaptive strategies of plants to Pi starvation, and also open avenues for the identification of important genes involved in Pi metabolism and PSRs, which can be leveraged to improve Pi use efficiency in agricultural crops.

# A truncated B-box zinc finger transcription factor confers drought sensitivity in modern cultivated tomatoes

Jinhua Li [*,1], Guo Ai[2], Yaling Wang[1], Yin Ding[1], Xiaomeng Hu[1], Yan Liang[1], Qingxia Yan[1], Ke Wu[1], Huang Rong[2], Chunrui Chen[1], Bo Ouyang[2], Xingguo Zhang[1], Yu Pan[1], Lang Wu[1], Zonglie Hong[3], Junhong Zhang[*,2]

[1] *Key Laboratory of Agricultural Biosafety and Green Production of Upper Yangtze River (Ministry of Education), College of Horticulture and Landscape Architecture, Southwest University, Chongqing 400715, P. R. China;*

[2] *National Key Laboratory for Germplasm Innovation & Utilization of Horticultural Crops, Huazhong Agricultural University, Wuhan 430070, P. R. China*

[3] *Department of Plant Sciences, University of Idaho, Moscow, Idaho 83844, USA.*

E-mail: ljh502@swu.edu.cn; zhangjunhng@mail.hzau.edu.cn

## Abstract

Enhancing drought tolerance in crops and understanding the underlying mechanisms have been subject of intense research. The precise function and molecular mechanisms of B-box zinc finger proteins (BBX) remain elusive. Here we report a natural allele of *BBX18* (*BBX18*$^{TT}$) that encodes a C-terminal truncated protein. While most wild tomato germplasms contain the *BBX18*$^{CC}$ allele and show more drought tolerant, modern cultivated tomatoes mostly belong to *BBX18*$^{TT}$ and are more drought sensitive. Knockout of *BBX18* leads to improved drought tolerance in transgenic plants of cultivated tomato. Ascorbate peroxidase 1 (APX1) is identified as a BBX18-interacting protein that acts as a positive regulator of drought resistance in tomato. Chromatin immunoprecipitation sequencing analyses reveal that BBX18 binds to a unique *cis*-acting element of the *APX1* promoter and represses its gene expression. This study provides valuable insights into the molecular mechanism underlying drought resistance mediated by the BBX18-APX1 module in plants.

# Proanthocyanidin Biosynthesis in Grapevine and Its Responses to Light and Salinity Stresses

Keji Yu [1,2*], Jing Cheng[1,2], Richard A. Dixon [4], Changqing Duan [1,2]

[1] *Center for Viticulture and Enology, College of Food Science & Nutritional Engineering, China Agricultural University, Beijing 100083, China*

[2] *Key Laboratory of Viticulture and Enology, Ministry of Agriculture and Rural Affairs, Beijing 100083, China.*

[3] *BioDiscovery Institute and Department of Biological Sciences, University of North Texas, Denton, TX 76203, USA*

E-mail: yukeji@cau.edu.cn

## Abstract

Grape berry derived proanthocyanidins (PAs) are natural flavan-3-ol polymers that contribute bitterness, astringency and color stability to red wines. However, the molecular mechanisms governing PA biosynthesis and its regulation by environmental signals in grapevine are not fully understood. Recently, we identified cysteine conjugates of (+)-catechin and (-)-epicatechin (Cys-C and Cys-EC) and oligomeric ascorbate conjugates of (+)-catechin (AsA-$[C]_n$) as extension unit intermediates for PA non-enzymatic polymerization in grapevine. In addition to catalyzing leucocyanidin, the two leucoanthocyanidin reductases (VvLARs) from grapevine also convert Cys-C and Cys-EC into (+)-catechin and (-)-epicatechin, respectively, thereby regulating PA polymerization. Using a grape callus system, we demonstrate that high light intensity promotes the accumulation of (+)-catechin-type PAs, while flavonol and anthocyanin levels remain relatively unaffected. Combining biochemical and genetic evidence, we further show that VvMYB86 enhances PA biosynthesis but suppresses the anthocyanin pathway under high light conditions by targeting a set of flavonoid genes, including the two VvLARs, in grapevine. Under salinity stress, metabolite and transcriptome analyses reveal that PA and anthocyanin biosynthesis are induced simultaneously in grape cells. The activation of PA biosynthesis under salinity stress is associated with the upregulation of classical flavonoid activators and the downregulation of a newly identified PA-negative transcriptional factor, VvbHLH93. These findings advance our knowledge of PA biosynthesis regulation and provide targets for tailoring PA composition in grape berries.

# Combining AI-powered analytics with multi-scale phenotyping to study yield and quality traits in controlled environment and under field conditions

Ji Zhou

*CAS Center for Excellence in Molecular Plant Sciences, Shanghai Institute of Plant Physiology & Ecology, Chinese Academy of Sciences*

E-mail: jzhou@cemps.ac.cn

## Abstract

Artificial intelligence (AI) and deep learning (DL) are enabling novel biological discoveries in horticultural crop research. Complex traits such as seed vigour, 3D tree-level morphological features, blossom clustering, and fruit formation can now be quantified or modelled by AI-powered solutions, empowering breeders and researchers to identify desirable traits under complex field conditions, in a scalable and effective manner. In the talk, the speaker will start with multi-spectral seed imaging and germination analysis, which is potentially important to study seed vigour for horticultural crops. Then, large-scale LiDAR- and drone-based tree phenotyping will be introduced, which was used to examine growth dynamics at key developmental phases for apple and pear orchards using a combined DL and 3D computer vision (CV) approach. At the end of the talk, the speaker will introduce AI-powered 3D trait analysis developed by his lab for strawberry, followed by mapping genetics underlying desired characteristics that are key to yield and quality for strawberry production.

# Toward revealing the molecular mechanism of yellow apples by analyzing transcriptomic and metabolic profiles of peels

Pingxing Ao[1], Yan Ma[1], Kang Luo[1], Yue Xu[1], Hongjia Zhang, Yun Zheng[1,2,*]

[1] *College of Landscape and Horticulture, Yunnan Agricultural University, Kunming, Yunnan 650201, China*

[2] *College of Big Data, Yunnan Agricultural University, Kunming, Yunnan 650201, China*

E-mail: zhengyun@fudan.edu.cn

## Abstract

Apple is a woody tree in the Rosaceae family, genus Apple. It has been widely reported that MYBs are critical regulators for red color of apple peel by activating the expression of anthocyanin biogenesis genes. However, it is still not clear what is the molecular mechanism for the yellow color of apple peel. In order to investigate key genes and metabolites responsible for yellow coloration of apple peel, three strains of apples, ``Venus Gold (Ype)" with yellow peel, ``Yanfu8 (Mpe)" with medium red peel, and ``Red love" with dark red peel, were selected in this study. Transcriptomic and metabolomic profiles were obtained for the peels of the three apple strains, respectively. After analyzing the transcriptomic profiles, our results suggest that LAR1 is a critical gene for the yellow color of Ype peel. Analysis of metabolomic profiles revealed that the abundances of Catechin and Epicatechin in Ype peel was higher than that of Rpe, indicating an important reason for the yellow color of Ype peel. Furthermore, when comparing volatile metabolites from Ype, Mpe and Rpe, hundreds of volatile metabolites show significantly differently abundances, suggesting that apples with different peel colors have different odors. In summary, our results provide new insights into the yellow coloration for the peels of some apple cultivars.

# Multiomics analysis of flowers reveal that enlarged epidermic cells of petals as the major reason of large flowers of *Gypsophila paniculata* L.

Yue Xu[1], Kang Luo[1], Hongjia Zhang[1], and Yun Zheng[1,2]

[1] *College of Landscape and Horticulture, Yunnan Agricultural University, Kunming, Yunnan, China*
[2] *College of Big Data, Yunnan Agricultural University, Kunming, Yunnan, China*

E-mail: zhengyun@fudan.edu.cn

## Abstract

*Gypsophila paniculata* L. is an important cut flower, often used as decorating flower or processed into eternal (or dry) flower. *Gypsophila paniculata* L. with double petals (GPDP) is of very high ornamental and economical value. However, the flower of GPDP is very small, with average diameters of 10 to 14 mm, which severely hinders the elevation of its economic value. To understand the molecular mechanism of the enlargement of flowers of GPDP, we produced RNA-Seq, small RNA-Seq and degradome profiles of petals of two GPDP cultivars, one with small flowers of diameters of 10 mm and the other with flowers of diameters of 14 mm. Our results indicate that the gene expression profiles of these two GPDP cultivars are very different. We also characterized the miRNA-mediated gene network in *Gypsophila paniculata* L. Importantly, we found that the epidermic cells of petals were significantly enlarged in the cultivar with large flowers. Our results also suggest that an MYB gene is a key regulator for the enlargement of epidermic cells in the GPDP cultivar with large flowers. In summary, our results indicate that the enlargement of epidermic cells of petals is the main reason for the larger flowers of *Gypsophila paniculata* L., which provide clues for breeding of more valuable cultivars of baby's breath.

# A glimpse to the overall perspective of the RNA-based gene duplication in the plant kingdom

Zhenglin Zhu[1]

[1] *School of Life Sciences, Chongqing University, No.55 Daxuecheng South Road, Shapingba, Chongqing, 401331, China*

E-mail: zhuzl@cqu.edu.cn

## Abstract

An overall view of the RNA-based gene duplication, retropositon, in plant species is important. For these, we performed a comprehensive identification of retrocopies in 122 plant species, ranging from Chondrus crispus to eudicots. The crop Camelina sativa has an exceptionally high retroposition frequency (4499 retrocopies), followed by Brassica napus (3236 retrocopies) and Camellia sinensis (1963 retrocopies). Like mammals, there is a high number of plant retrocopies with a small Ks. We examined the flanking regions of retrocopies and performed comprehensive correlation analyses between the number of retrocopies and possible influence factors. The results suggest that long interspersed nuclear element 1 (LINE-1 or L1) retrotransposons may contribute to the producing of plant retrocopies predominantly. Analyses on available transcriptome data and predicted function annotation show a weak bias of plant retrogenes towards stress response. These findings should enrich the understanding of the RNA-based gene duplication in plant species.

# eQTL meta-analysis reveals conserved and population-specific regulatory variation underlying nutritional trait evolution and domestication in tomato

Jiantao Zhao[1,#], Xin Wang[1,6,#], Jing Zhang[2], Yimin Xu[1], Ryan McQuinn[1], Je Min Lee[1], Itay Gonda[1], Ari Feder[1], Denise M. Tieman[3], Sanwen Huang[4], Harry Klee[3], James J. Giovannoni[1,5], Zhangjun Fei[1*]

[1] *Boyce Thompson Institute, Cornell University, Ithaca, NY, USA*

[2] *College of Horticulture, Northwest A&F University, Yangling, Shaanxi, China*

[3] *University of Florida, Horticultural Sciences, Gainesville, FL, USA*

[4] *National Key Laboratory of Tropical Crop Breeding, Shenzhen Branch, Guangdong Laboratory of Lingnan Modern Agriculture, Genome Analysis Laboratory of the Ministry of Agriculture and Rural Affairs, Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, Guangdong 518120, China*

[5] *U.S. Department of Agriculture-Agricultural Research Service, Robert W. Holley Center for Agriculture and Health, Ithaca, NY, USA*

[6] *Current address: National Key Laboratory for Germplasm Innovation & Utilization of Horticultural Crops; Hubei Hongshan Laboratory; College of Horticulture and Forestry Sciences, Huazhong Agricultural University, Wuhan, China*

*#These authors contributed equally.*

E-mail: jz426@cornell.edu

## Abstract

Deciphering how genetic variation shapes transcriptional regulation is key to understanding the regulatory architecture of complex crop traits. Here, we present a comprehensive meta-analysis of expression quantitative trait loci (eQTLs) across five diverse tomato populations. We construct a genome-wide atlas of 40,928 eQTLs, comprising 9,463 *cis*- and 31,465 *trans*-eQTLs, and reveal thousands of loci detectable only through meta-analysis. A substantial fraction of these eQTLs is associated with transcription factors (TFs), including a co-regulated cluster of two TFs and one transcriptional regulator targeted by a shared *trans*-eQTL hotspot harboring *Solyc03g124068*, a key enzyme in gibberellin biosynthesis. Genes underlying nutritional quality traits, including sugars, organic acids, carotenoids, and folates, are predominantly controlled by strong *cis*-eQTLs. We further delineate a *MYB12* (*Solyc01g079620*)-centered regulatory network coordinating flavonoid metabolism that also extends to folate biosynthesis. Remarkably, MYB12 functions as a *trans*-regulator linking flavonoid and folate pathways, contributing to the reduced folate content characteristic of pink-fruited tomatoes. Together, this eQTL meta-analysis provides a high-resolution regulatory map of transcriptional variation in tomato and highlights key regulators of fruit nutritional quality, providing biological insights and practical targets for crop improvement.

# Brassinosteroid-mediated positive regulatory network governing leaf droopiness for mechanical harvesting revealed through integrated GWAS and ATAC-seq analysis

Haoran Liu[1*], Lingxiao Duan [1], Liang Chen[1], Jiedan Chen[1], Jianqiang Ma[1]

[1] *State Key Laboratory of Tea Plant Germplasm Innovation and Resource Utilization, Tea Research Institute of the Chinese Academy of Agricultural Sciences, Hangzhou 310008, China*

E-mail: hrliu@tricaas.com

## Abstract

Due to a labor shortage, the mechanical harvesting of tea plantations has become a focal point. However, mechanical harvest efficiency was hampered by droopy leaves, leading to a high rate of broken tea shoots and leaves. Here, we dissected the genetic structure of leaf droopiness in tea plants using genome-wide association studies (GWAS) on 146 accessions. A set of 16 quantitative trait loci (QTLs) containing 54 SNPs and 34 corresponding candidate genes associated with droopiness were then identified. Two genes nearby lead SNPs, *CsEXL3* (*EXORDIUM-LIKE 3*) and *CsTPR* (*TETRATRICOPEPTIDE REPEAT*), were identified as droopiness related genes based on co-joint analysis with transcriptome and ATAC-seq, respectively. CsEXL3 and CsTPR were then proved to positively or negatively regulate brassinosteroid -induced droopiness by using silencing tea plant, respectively. Additionally, brassinosteroid signal factor CsBES1.2 was proved to participate in *CsEXL3* or *CsTPR*-induced droopiness and vascular cell malformation via using the *CsBES1.2*-silencing tea plant. Notably, CsBES1.2 bound on the E-box of *CsEXL3* promoter to transcriptionally activate *CsEXL3* expression as CUT&TAG based ChIP-qPCR and ChIP-seq suggested *in vivo* and EMSA and Y1H indicated *in vitro*. CsBES1.2 also bound the E-box of *CsTPR* promoter to repress the expression of *CsTPR*, as demonstrated by ChIP-qPCR etc. The whole genome sequencing (WGS) indicated that a single base mutation (T to A) in the promoter of *CsTPR*, which strengthened the inhibitory effect of CsBES1.2 on the expression of *CsTPR* via enhancing the binding affinity to the E-box. Altogether, our findings suggest that the positive regulation network of brassinosteroids on mechanical harvesting related leaves droopiness through the positive and negative transcriptional regulation of CsBES1.2 on the expression of *CsEXL3* and *CsTPR*, respectively.

# Genomic evolution and regulation of key agronomic traits in *Curcuma* species

Xuezhu Liao[1]

[1] *College of Life Sciences and Oceanography, Shenzhen University*

E-mail: 1113monian@live.cn

## Abstract

The genus *Curcuma* is renowned for its medicinal and culinary value, attracting widespread attention due to the abundance of bioactive compounds in its rhizomes that exhibit anticancer, anti-inflammatory, and antioxidant effects. Species within this genus exhibit highly variable chromosome base numbers and widespread polyploidy (ranging from 2*X* to 15*X*), suggesting extensive genomic structural variation. Against this background, we assembled high-quality haplotype-resolved genomes for three *Curcuma* species (*C. longa*, *C. petiolata*, and *C. alismatifolia*) and identified chromosomal inversions at multiple levels: between species, between haplotypes within species, and among populations within species. We further investigated the impact of these inversions on the evolution of terpenoid biosynthesis. In addition, we also investigated the development and dormancy of rhizomes — a key asexual reproductive organ and harvestable storage structure in Zingiberaceae plants — and identified a set of critical genes. These findings establish a genetic foundation for the molecular breeding of *Curcuma* species.

# Citrus-derived antimicrobial metabolites confer resistance to Huanglongbing and Canker

Yuantao Xu[1,2], Kun Yang[1], Wang Zhang[1], Qiang Xu[1,2]

[1] *National Key Laboratory for Germplasm Innovation & Utilization of Horticultural Crops, Huazhong Agricultural University, No. 1, Shizishan Street, Hongshan District, Wuhan 430070, Hubei, China*

[2] *Hubei Hongshan Laboratory, No. 1, Shizishan Street, Hongshan District, Wuhan 430070, Hubei, China*

E-mail: xuyuantao@mail.hzau.edu.cn

## Abstract

Citrus huanglongbing (HLB, mainly caused by *Candidatus* Liberibacter asiaticus, *C*Las) and citrus canker (caused by *Xanthomonas citri* ssp. *Citri*, *Xcc*), are two of the most devastating diseases threatening global citrus production. Developing sustainable and effective management strategies is a critical priority for the citrus industry. Plant secondary metabolites play pivotal roles in defense responses to biotic or abiotic stresses. Recently, we identified disease-resistant citrus germplasm through comprehensive disease resistance evaluation and molecular screening. Comparative metabolomic profiling of resistant and susceptible germplasm revealed a suite of antimicrobial secondary metabolites, including polyphenol, coumarins, and phytohormone, that were significantly up-regulated in resistant varieties. Antibacterial assays showed that chlorogenic acid effectively inhibits *C*Las, while melatonin, daphnetin and esculetin exhibit strong inhibitory activity against *Xcc*. Field trials further confirmed their efficacy in suppressing disease symptoms and limiting pathogen spread. These findings highlight the potential of citrus-derived antimicrobial metabolites as eco-friendly agents for managing HLB and canker, and provide valuable foundation for integrating metabolic resistance into breeding programs and disease control strategies.

# Super Pangenome of Vitis Empowers Identification of Downy Mildew Resistance Genes for Grapevine Improvement

Li Guo[1], Xiangfeng Wang[1], Dilay Hazal Ayhan[1], Mohammad Saidur Rhaman[1], Ming Yan[1], Jianfu Jiang[2], Dongyue Wang[1], Wei Zheng[1,3], Junjie Mei[1,3], Wei Ji[1,3], Jian Jiao[1,4], Shaoying Chen[1], Jie Sun[1], Shu Yi[1], Dian Meng[1], Jing Wang[1], Mohammad Nasim Bhuiyan[1], Guochen Qin[1], Linling Guo[1], Qingxian Yang[1], Xuenan Zhang[1], Haisheng Sun[1], Chonghuai Liu[2], Xing Wang Deng[1,5], Wenxiu Ye[1]

[1] *Shandong Key Laboratory of Precision Molecular Crop Design and Breeding, Peking University Institute of Advanced Agricultural Sciences, Shandong Laboratory of Advanced Agricultural Sciences in Weifang, Shandong 261325, China*
[2] *Zhengzhou Fruit Research Institute, Chinese Academy of Agricultural Sciences, Zhengzhou 450004, China*
[3] *College of Horticulture, Shanxi Agricultural University, Taigu 030801, China*
[4] *College of Horticulture, Henan Agricultural University, Zhengzhou 450046, China*
[5] *School of Advanced Agriculture Sciences and School of Life Sciences, State Key Laboratory of Protein and Plant Gene Research, Peking University, Beijing 100871, China*

E-mail: wenxiuye@pku-iaas.edu.cn

## Abstract

Grapevine (*Vitis*) is one of the oldest domesticated fruit crops with great cultural and economic importance. Here we assembled and annotated haplotype-resolved genomes of 72 global *Vitis* accessions including 25 wild and 47 cultivated grapevines, among which genomes for 60 grapevines are newly released. Haplotype-aware phylogenomics disentangled the mysterious hybridization history of grapevines, revealing the enormous genetic diversity of the *Vitis* genus. Pangenomic analysis reveals that European cultivars, more susceptible to the destructive disease downy mildew (DM), have a smaller repertoire of resistance genes in the NLR family encoding the TIR-NBARC-LRR domain. Through extensive structural variation (SV) characterization, phenotyping, DM-infection transcriptome profiling of 113 Vitis accessions, and SV–expression quantitative trait loci analysis, we have identified over 63 SVs and their relevant genes significantly associated with DM resistance, exemplified by a lysine histidine transporter, *VvLHT8*. This haplotype-resolved super pangenome of the *Vitis* genus will accelerate breeding and enrich our understanding of the evolution and biology of grapevines.

# A calcium-decoding module translates volatile DMNT into jasmonic acid-mediated herbivore resistance in *Camellia sinensis*

Mengyao Chai, Hao Ni, Yahong Zheng, Ruixue Wang, , Chuankui Song, Tingting Jing

*National Key laboratory for tea Plant Germplasm innovation and Resource Utilization, Anhui Agricultural University, 230036 Hefei, Anhui, P. R. China.*

E-mail: jtt0127@163.com

## Abstract

Plants employ sophisticated volatile-mediated signaling mechanisms to defend against herbivore attacks; however, the molecular pathways underlying these processes remain poorly understood. Herein, we unveil the molecular mechanism by which the herbivore-induced volatile (E)-4,8-dimethyl-1,3,7-nonatriene (DMNT) activates jasmonic acid (JA) biosynthesis in tea plants (*Camellia sinensis*). We demonstrate that DMNT triggers early signaling events in tea plants, characterized by a rapid influx of $Ca^{2+}$ in mesophyll cells, which subsequently initiates a signaling cascade involving the CsCAMTA3 (calmodulin-binding transcription activator, CAMTA). Using luciferase reporter assays, yeast one-hybrid analysis, electrophoretic mobility shift assays, and antisense oligonucleotide silencing, we demonstrated that CsCAMTA3 directly binds to the CG-box element in the promoter region of the key transcription factor CsWRKY70 and activates its expression. Subsequently, CsWRKY70 binds to the W-box motif in the *CsLOX3* (*Lipoxygenase, LOX*) promoter, driving the expression of the JA biosynthetic gene and promoting the accumulation of JA and Jasmonoyl-Isoleucine JA-Ile. Silencing *CsCAMTA3* or *CsWRKY70* significantly inhibits JA biosynthesis and reduces tea plant resistance to tea geometrid (*Ectropis obliqua*). Furthermore, the suppression of both *CsCAMTA3* and *CsWRKY70* significantly diminished the ability of tea plants to synthesize JA and JA-Ile in response to DMNT signal induction. Our findings reveal a $Ca^{2+}$-CAMTA3-WRKY70-LOX3 signaling cascade induced by DMNT, broadening the theoretical framework of plant–plant communication.

# Ligand-induced degradation of ethylene receptor modulates plant response to ethylene

Qincui Zhao[1], Nan Ma[1], Xiaofeng Zhou[1]

[1] *Beijing Key Laboratory of Development and Quality Control of Ornamental Crops, Department of Ornamental Horticulture, China Agricultural University, Beijing 100193, China.*

E-mail: zhouxiaofeng@cau.edu.cn

## Abstract

Gaseous phytohormone ethylene regulates various aspects of plant development. Ethylene is perceived by ER membrane-localized receptors, which are inactivated upon binding with ethylene molecules, thereby initiating ethylene signal transduction. Here, we report that a novel E3 ligase RING finger for Ethylene receptor Degradation (RED) and its E2 partner UBC32 ubiquitinate ethylene-bound receptors for degradation through an ER associated degradation (ERAD) pathway in both *Rosa hybrida* and *Solanum lycopersicum*. The depletion of RED or UBC32 leads to hypersensitivity to ethylene, which is manifested as premature leaf abscission and petal shedding in roses, as well as the dwarf plants and accelerated fruit ripening in tomatoes. Disruption of the conserved ethylene binding site of receptors prevents RED-mediated degradation of the receptors. Our study discovers an ERAD branch that facilitates the ethylene-induced degradation of receptors, and provides insights into how the plant's response to ethylene can be controlled by modulating the turnover of ethylene receptors.

# Key Gene Mining for Blue Petals and Floral Scent in Water Lilies

Zhang Liangsheng

*Zhejiang University*

## Abstract

The study employed Nymphaea (blue star water lily) and Eustoma grandiflorum (lisianthus) as experimental materials, and successfully mined the key genes that regulate the formation of blue petals. The discovery of these genes provides core research targets for in-depth analysis of the molecular mechanism underlying blue flower organ development, and lays a molecular foundation for the subsequent improvement of blue flower varieties.Meanwhile, with Nymphaea (blue star water lily) and Victoria cruziana (Cruz water lily) as research objects, the key genes involved in the synthesis of volatile fatty acid-type floral scent substances were identified. There is a significant organ-specific difference in scent release between the two species: the floral scent substances of Nymphaea (blue star water lily) mainly originate from petal tissues, while those of Victoria cruziana are concentrated in the inner whorl stamen appendages. The essence of this difference is a direct result of gene subfunctionalization during evolution.

# Nocturnal burst emissions of germacrene D from the open disk florets of pyrethrum flowers induce moths to oviposit on a nonhost and improve pollination success

Jinjin Li[1], Manqun Wang[2], Maarten A. Jongsma[3], Hao Hu[1,4] and Caiyun Wang[1]

[1] *National Key Laboratory for Germplasm Innovation & Utilization of Horticultural Crops, Huazhong Agricultural University, Wuhan, 430070, China*

[2] *Hubei Insect Resources Utilization and Sustainable Pest Management Key Laboratory, College of Plant Science and Technology, Huazhong Agricultural University, Wuhan, 430070*

[3] *Business Unit Bioscience, Wageningen Plant Research, Droevendaalsesteeg 1, 6708 PB, Wageningen, the Netherlands*

[4] *School of Life Sciences, Peking-Tsinghua Joint Center for Life Sciences, Peking University, Beijing, 100871, China*

E-mail: hao.hu@pku.edu.cn; wangcy@mail.hzau.edu.cn

## Abstract

Recent studies show that nocturnal pollinators may be more important to ecosystem function and food production than is currently appreciated. Here, we describe an agricultural field study of pyrethrum (Tanacetum cinerariifolium) flower pollination. Pyrethrum is genetically self-incompatible and thus is reliant on pollinators for seed set. Our pollinator exclusion experiment showed that nocturnal insects, particularly moths, significantly contribute to seed set and quality.We discovered that the most abundant floral volatile, the sesquiterpene (-)-germacrene D (GD), is key in attracting the noctuid moths Peridroma saucia and Helicoverpa armigera. Germacrene D synthase (GDS) gene expression regulates the specific GD production and accumulation in flowers, which, in contrast to related species, lose the habit of closing at night. We did observe that female moths also oviposited on pyrethrum leaves and flower peduncles, but found that only a small fraction of those eggs hatched. Larvae were severely stunted in development, most likely due to the presence of pyrethrin defense compounds. This example of exploitative mutualism, which blocks the reproductive success of the moth pollinator and depends on nocturnal interactions, is placed into an ecological context to explain why it may have developed.

# Metabolomic and transcriptomic analyses provide insight into the variation of floral scent and molecular regulation in different cultivars and flower development of Curcuma alismatifolia

Chao Song[a] , Jingpu Tian[b †], Dejin Xie[c], Shengnan Lin[a], Yingxue Yang[a,c], Xiaoni Zhang[a,c], Xuezhu Liao[c*], Zhiqiang Wu [a,c*]

[a] *Kunpeng Institute of Modern Agriculture at Foshan, Shenzhen Branch, Guangdong Laboratory of Lingnan Modern Agriculture, Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, 518124, China.*

[b] *Department of Architecture and Design, Hunan University of Science and Technology, Xiangtan, 411201, China.*

[c] *Shenzhen Branch, Guangdong Laboratory of Lingnan Modern Agriculture, Key Laboratory of Synthetic Biology, Ministry of Agriculture and Rural Affairs, Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, China.*

E-mail: chaosong0506@qq.com

## Abstract

*Curcuma alismatifolia* is an important ornamental plant of significant economic value, while the floral fragrance has been rarely investigated, leading to a lack of knowledge about the floral scent. By performing metabolomic and transcriptomic analyses, we investigated the variation of 906 volatile organic compounds (VOCs) in florets of eight *C. alismatifolia* cultivars and four different developmental stages of 'Chiang Mai Pink' (CMP). The metabolite profiling revealed that the terpenoid group (213 out of 906) was the predominant VOC, accounting for 33.5% and 43.4% of total VOC contents in the florets of different cultivars and developmental stages, respectively. Sweet and woody were the predominant odors not only in different cultivars but also during developmental stages. The varied intensities of other odors contributed to forming odor diversities in *C. alismatifolia* floret. We uncovered seven terpenoid synthetase (*TPS*) genes and four *MYB* genes of significant association with the biosynthesis of terpenoids in eight cultivars and floret development, respectively. We performed an activity assay on four selected *TPS* genes and identified that Chr15HA1352 and Chr15HA2528 are responsible for the biosynthesis of $\alpha$-farnesene. The significant association between the *MYB* gene (*Chr03HA28*) and seven terpenoids can be observed among different cultivars and during different developmental stages. These findings highlight the varying floral scents in different cultivars and floret development and suggest the potential roles of identified *TPS* and *MYB* genes in the biosynthesis of terpenoids in *C. alismatifolia*.

# Regulatory module of RFO hydrolysis by alkaline α-galactosidase CsAGA2 controls cucumber development

Huan Liu

*Department of Horticulture, Zhejiang University, Hangzhou 310058*

E-mail: huan-liu@zju.edu.cn

## Abstract

Seed size and weight are pivotal agronomic traits that link plant sexual reproduction with subsequent seedling establishment and are affected by embryo development and sugar filling. However, the molecular mechanism that controls sugar translocation and the timing between early embryo development and subsequent seed filling in cucumber remains poorly understood. Here, we report that the alkaline α-galactosidase 2 (*CsAGA2*) is expressed in the placental vascular bundles and funicular phloem and is responsible for the hydrolysis of Raffinose Family Oligosaccharides (RFOs). Further study proved that FUSCA3 (CsFUS3) activates and Auxin Response Factor 9 (CsARF9) represses the expression of *CsAGA2*. Interaction of CsFUS3 and Sucrose Non-fermenting-1 (Snf1)-Related Protein Kinase 1α1 (CsSnRK1α1) further enhanced *CsAGA2* expression, while CsARF9 recruitment of the TOPLESS (CsTPL) corepressor further weakened *CsAGA2* expression. Transgenic *CsAGA2*-RNAi (RNA interference), *CsFUS3*-RNAi (*csfus3*-crispr) and *CsARF9*-OE (overexpression) cucumber lines showed severe seed abortion rates, likely caused by reduced sugar supply during embryo development and seed filling stages. These results demonstrated the critical collaboration of these genes in delivering sugars for development of seeds. Our findings revealed that cucumber CsAGA2 is an essential enzyme that is switched on by CsFUS3 to precisely regulate embryo development and to provide the vast quantities of sugars needed for seed filling (15-25 DAP). As the seeds mature, *CsAGA2* expression is decreased by CsARF9 to gradually reduce the sugar supply to seeds.

**Long male peduncles and the evolution of sex chromosomes in papaya**

Yongmei Zhou[1], Ziqin Pang[2], Wen Wang[1], Ray Ming[1]

*[1] Center for Genomics and Biotechnology, Fujian Agriculture and Forestry University, Fuzhou, Fujian 350002, China*

*[2] National Engineering Research Center for Sugarcane, Fujian Agriculture and Forestry University, Fuzhou, 350002 China*

## Abstract

Papaya is a fruit crop with XY sex chromosomes. The development of long male peduncles (Mp) is critical for the transition from gynodioecy to dioecy. We identified *CpMp*, a Y-linked SVP paralog (*CpSVP-Yp*), as the gene controlling peduncle length through comparative genomics and complementation tests. *CpMp* promotes peduncle elongation by activating *CpYUC6*, an auxin biosynthesis gene, increasing IAA levels. Elevated IAA enhances GA production, triggering *CpGASA6*, which drives cell division and elongation. Overexpression of *CpGASA6* confirmed its role and revealed feedback inhibition on IAA synthesis. Two upstream regulators of *CpMp* were identified: *CpTRAB1*, a GA-repressed activator, and *CpGATA8*, a *CpMp*-induced repressor, forming feedback loops. Notably, male trees produce 400 times more pollen than hermaphrodites, providing a fitness advantage and establishing dioecy in papaya. Together with two sex-determination genes, *CpMp* was essential for dioecy and sex chromosome evolution, offering new insights and genetic resources for papaya improvement.

# Transcriptional regulation of miR528-PPO module by miR156 targeted SPLs orchestrates chilling response in banana

Xiangjin Kong[1,2,3], Kuan Peng[1,2,3], Youxia Shan[1,2], Ze Yun[1,2,3], Tamas Dalmay[4], Xuewu Duan[1,2,3], Yueming Jiang[1,2,3], Hongxia Qu[1,2,3*] and Hong Zhu[1,2,3*]

[1] *Guangdong Provincial Key Laboratory of Applied Botany, Key Laboratory of South China Agricultural Plant Molecular Analysis and Genetic Improvement, South China Botanical Garden, Chinese Academy of Sciences, 510650 Guangzhou, China*

[2] *South China National Botanical Garden, Guangzhou 510650, China*

[3] *University of Chinese Academy of Sciences, Beijing 100049, China*

[4] *School of Biological Sciences, University of East Anglia, Norwich, UK*

E-mail: kongxiangjin@scbg.ac.cn

## Abstract

Banana is sensitive to cold stress and often suffers from chilling injury with browning peel and failure to normal ripening. We have previously reported that banana chilling injury is accompanied by a reduction of miR528 accumulation, alleviating the degradation of its target gene *MaPPO* and raising ROS levels that cause peel browning. Here, we further revealed that the miR528-MaPPO cold-responsive module was regulated by miR156-targeted SPL transcription factors, and the miR156c-MaSPL4 module was also responsive to cold stress in banana. Transient overexpression of miR156c resulted in a more severe chilling phenotype by decreasing the expression of *MaSPL4* and miR528. Conversely, the browning was alleviated in STTM-miR156c silencing and OE-*MaSPL4* samples. Furthermore, DNA affinity purification sequencing and *MaSPL4*-overexpressing transcriptome jointly revealed that MaSPL4 may mediate the transcription of genes related to lipid metabolism and antioxidation, in addition to the miR528-MaPPO module, demonstrating MaSPL4 as a master regulator in the fruit cold response network. In summary, our results suggest that the miR156c-MaSPL4 module can mediate the chilling response in banana by regulating the miR528-MaPPO module and multiple other pathways, which provides evidence for the crosstalk between TFs and miRNAs that can be used for the molecular breeding of fruit cold tolerance.

# Evolution and functional divergence of glycosyltransferase genes shaped the quality and cold tolerance of tea plants

Jingming Wang[1], Yutong Hu[1], Yuling Wang[1]， Danyang Guo[1], Chuankui Song[1,*]

[1] *State Key Laboratory of Tea Plant Biology and Utilization, Anhui Agricultural University, 130 Changjiang Ave W., Hefei, Anhui 230036, People's Republic of China*

E-mail: wangjm0611@163.com

## Abstract

Plant uridine diphosphate–dependent glycosyltransferases (UGTs) play a key role in plant growth and metabolism. Here, we examined the evolutionary landscape among UGTs in 28 fully sequenced species from early algae to angiosperms. Our findings revealed a distinctive expansion and contraction of UGTs in the G and H groups in tea (*Camellia sinensis*), respectively. Whole-genome duplication and tandem duplication events jointly drove the massive expansion of UGTs, and the interplay of natural and artificial selection has resulted in marked functional divergence within the G group of the sinensis-type tea population. In Cluster II of group G, differences in substrate selection (e.g. abscisic acid) of the enzymes encoded by UGT genes led to their functional diversification, and these genes influence tolerance to abiotic stresses such as low temperature and drought via different modes of positive and negative regulation, respectively. UGTs in Cluster III of the G group have diverse aroma substrate preferences, which contribute a diverse aroma spectrum of the sinensis-type tea population. All Cluster III genes respond to low-temperature stress, whereas UGTs within Cluster III-1, shaped by artificial selection, are unresponsive to drought. This suggests that artificial selection of tea plants focused on improving quality and cold tolerance as primary targets.

# Ethylene promotes anthocyanin synthesis in 'Viviana' lily via the LvMYB5-LvERF113-LvMYB1 module

Yibing Zhang[1†], Lijing Chen[1*]

[1] *Key Laboratory of Agriculture Biotechnology, Key Laboratory of Protected Horticulture (Ministry of Education), College of Biosciences and Biotechnology, Shenyang Agricultural University, Liaoning 110161, China;*

E-mail: zhangyibing@stu.syau.edu.cn; chenlijing@syau.edu.cn

## Abstract

Ethylene (ET) influences the synthesis of anthocyanins, although its regulatory effects can differ significantly across various plant species. In apples (Malus domestica), ET promotes anthocyanin synthesis, whereas in Arabidopsis thaliana, it inhibits its accumulation. Our research showed that ethephon (Eth), an ET derivative, promotes anthocyanin synthesis in 'Viviana' lilies, which has great potential in the cut flower industry. The regulatory mechanism whereby ET influences anthocyanin synthesis in lilies remains unclear. In this study, we screened and characterized an ET-induced ET response factors (ERFs), LvERF113, with inhibitory function. Our analyses suggested that LvERF113 could inhibit the negative regulatory function of LvMYB1 at transcriptional and posttranslational levels, promoting anthocyanin synthesis in 'Viviana' lily tepals. In addition, LvERF113 is positively regulated by LvMYB5, forming the LvMYB5-LvERF113-LvMYB1 module controlling anthocyanin synthesis by ET in 'Viviana' lily. These findings offer new insights into the ET regulatory network of anthocyanin synthesis and provide a theoretical basis for the application of ET derivatives in the cut flower industry.

# Alginate Oligosaccharide Promoted the Nutrient Uptake and Growth of Cucumber Seedlings Under Suboptimal Temperature Conditions

Xu Guo[1], Yun Li[1], Kai Fan[1], Lingru Guo[1], Zhenchuan Mao[2], Leiping Hou[1], Yaling Li[1],* and Longqiang Bai[1]*

[1] *College of Horticulture, Shanxi Agricultural University, Jinzhong, 030801, China;*

[2] *Chinese Academy of Agricultural Sciences, Beijing, 100081, China*

E-mail: xuguogz@163.com

## Abstract

Due to its sensitivity to cold temperatures, cucumber growth is substantially constrained by suboptimal temperature stress in northern China's off-season production systems. Suboptimal temperatures severely repress the nutrient absorption, growth, and yield formation of vegetables in solar greenhouses during winter and early spring in China. Alginate oligosaccharides (AOSs) are anionic acidic polysaccharides derived from brown algae, known for promoting plant growth and alleviating abiotic stress. In this study, we aimed to investigate the effects of different nutrient solution concentrations combined with AOS on the growth and nutrient uptake of cucumber seedlings under suboptimal temperatures (15/8 ℃, day/night). Potted'Jinchun 4'cucumber seedlings grown in coconut coir were treated with 0.5×, 1.0×, or 1.5× strength of Hoagland solution alone (N0.5, N1, N1.5), or with 30 mg·$L^{-1}$ AOS (A0.5, A1, A1.5). The results showed that the growth attributes and nitrogen (N) accumulation of cucumber plants of N1 and N1.5 were significantly higher than those of N0.5. Additionally, plants of A0.5 exhibited significantly higher plant height, chlorophyll a content, root surface area, root volume, root vitality, N metabolism enzyme (NR, GDH, GS) activities, and N accumulation, than those under N0.5, N1, or N1.5. Moreover, compared to A0.5, the net photosynthetic rate, total root length, root surface area, root N content, leaf nitrate reductase activity, root glutamate dehydrogenase activity, and N accumulation of A1 and A1.5 were significantly higher than those of A0.5. Correlation analysis revealed strong linkages between root morphology traits and tissue N content. In summary, under suboptimal temperature conditions, the application of AOS improved cucumber seedlings' nutrient absorption and growth more efficiently than merely raising nutrient levels, as it enhanced root surface area, root vitality, and N metabolic enzyme activities.

# *CPK10* regulates secondary xylem development in poplar

Wenya Yuan[1], Mengzhu Lu[1], Jin Zhang[1]*

[1] *National Key Laboratory for Development and Utilization of Forest Food Sources, College of Forestry and Biotechnology, Zhejiang A&F University,Hangzhou,China*

E-mail: ywy19990109@126.com

## Abstract

Calcium ions ($Ca^{2+}$) function as key secondary messengers in numerous plant signal transduction pathways, including those regulating wood formation. Nevertheless, the specific $Ca^{2+}$ sensors modulating this process remain largely unknown. Calcium-dependent protein kinases (CPKs), a family of serine/threonine kinases, act as central $Ca^{2+}$ sensors involved in plant growth, development, and stress adaptation. Here, we identified *PtCPK10* from *Populus trichocarpa* as a putative regulator of xylem development. Histochemical GUS staining in transgenic poplar carrying $Pro_{PtCPK10}$*::GUS* revealed strong expression in expanding xylem tissues. Overexpression of *PtCPK10* led to a 12% increase in plant height, a 9% increase in stem diameter, and a 15% expansion in xylem width, along with significantly thickened xylem fiber cell walls. Conversely, knockout of PagCPK10 and its homolog *PagCPK11* in hybrid poplar (*Populus alba* × *P. glandulosa*, '84K') resulted in a dwarf phenotype with reduced xylem width and thinner fiber cell walls—phenotypes opposite to those of overexpression lines. Using yeast two-hybrid screening against a wood development-related transcription factor library, we identified four putative interactors of PtCPK10: MYB4, ZFP11a, ZFP11b, and a C2H2 ZFP. These interactions require further validation via BiFC and Co-IP assays. Subsequent studies will focus on identifying PtCPK10-specific phosphorylation targets and sites to elucidate its mechanistic role. Overall, this study highlights *PtCPK10* as a positive regulator of wood formation and provides a foundation for improving wood quality in poplar.

# MdHMGB15-MdXERICO-MdNRP Module Mediates Salt Tolerance of Apple by Regulating the Expression of Salt Stress-related Genes

Ranxin Liu[1], Shanshan Li[1], Qianyu Yue[1], xiaofei Wang[1*], chunxiang You [1*]

[1] *National Research Center for Apple Engineering and Technology, College of Horticulture Science and Engineering, Shandong Agricultural University, 271018, China.*

E-mail: 1411886357@qq.com

## Abstract

Soil salinity is an important limiting factor for plant growth. Salt stress, one of the most detrimental factors affecting crop output and plant growth, affects cells in both osmotic and ionic or ion-toxic ways. Plants must maintain ROS homeostasis, modify osmotic potential, and balance cellular ions in order to combat salt stress. Genetic and physiological and biochemical studies have demonstrated that various salt stress responses and regulatory factors of plants can regulate salt tolerance via multiple and complex stress signaling pathways. As a RING-type E3 ubiquitin ligase, MdXERICO is highly responsive to salt stress and can enhance salt tolerance of plants. However, the molecular mechanism for the response of MdXERICO to salt stress remains unclear. This study aims to dissect the molecular mechanisms for MdXERICO to regulate plant response to salt stress. This study revealed that MdXERICO interacted with MdNRP and improved salt tolerance of apple by ubiquitinating and degrading MdNRP via the 26S proteasome pathway. Moreover, the HMG box-containing transcription factor MdHMGB15 interacted with the MdXERICO promoter, thereby activating its expression and enhancing the salt tolerance of apple. Overall, this study investigated the tolerance of apple to salt stress by the MdHMGB15-MdXERICO-MdNRP module, providing references for future studies of salt tolerance of plants.

# Combined analysis of BSA and RNA-Seq to develop the candidate genes related to deciduous trait in Kiwifruit (Actinidia arguta)

Qina Zhang

*Zhengzhou University*

E-mail: zhang_qina99@163.com

## Abstract

*Actinidia arguta*, has a unique fruit flavor and is rich in vitamin C, possessing relatively high economic and nutritional values. The time of deciduous is important to its dormancy and tree resistance, especially to cold resistance. *A. arguta*, which is a deciduous fruit, displayed different deciduous traits in different accessions. Here, we performed BSA and RNA-seq using the *A. arguta* F1 populations to locate the deciduous trait. The results of BSA showed that the loci related to the deciduous trait were in a 2.04 Mb interval on chromosome Chr26. Moreover, through a comprehensive analysis of BSA and RNA-Seq data, 15 genes with differential expression in the parents (RB-3 and KL) were identified. These include RNA helicase, calcium-dependent protein kinase, nuclear factor Y, stress-related protein, and importin. After further analyzing their expression levels in the yellow-green leaves of the parents by RT-qPCR, 4 candidate genes related to the deciduous trait were determined, which include Importin-5 (IPO-5), Calcium-dependent protein kinase 7 (CDPK7)、Zinc finger A20 and AN1 domain-containing stress-associated protein 8 (SAP8) and DEAD-box ATP-dependent RNA helicase 52. In addition, we selected 4 individuals with high deciduous rates (Y01-Y04) and 4 individuals with low deciduous rates (G01-G04) from the two extreme pools of the progeny population to verify their expression trends. The results showed that the expression levels of these 4 genes in individuals with high deciduous rates were significantly higher than those in individuals with low deciduous rates, indicating that these 4 genes are candidate genes controlling the deciduous trait in plants. This study laid a foundation for the genetic mechanism of deciduous regulation and for developing its cold resistance genes in *A. arguta*.

# TEA5K: a high-resolution and liquid-phase multiple-SNP array for molecular breeding in tea plant

Dingding Liu[1†], Chenyu Zhang[1†], Yuanyuan Ye[1], Piao Mei[1], Yang Gong[1], Zhen Liu[2], Chao Sun[3], Xuecheng Zhao[1], Shiqi Ding[1], Jiedan Chen[1*], Liang Chen[1,4*] and Chunlei Ma[1*]

[1] *Tea Research Institute of the Chinese Academy of Agricultural Sciences; State Key Laboratory of Tea Plant Germplasm Innovation and Resource Utilization, Hangzhou 310008, China*

[2] *Tea Research Institute, Hunan Academy of Agricultural Sciences, Changsha 410125, China*

[3] *Tea Research Institute, Yunnan Academy of Agricultural Sciences, Kunming 650221, China*

[4] *Yunnan Key Laboratory of Tea Germplasm Conservation and Utilization in the Lancang River Basin, Lincang 677000, China*

E-mail: 13790050536@163.com

## Abstract

High-throughput genotyping technology has become an indispensable tool for advancing molecular breeding and genetic research in plants, facilitating large-scale exploration of genomic variation. In this study, we developed a highly efficient multiple-SNP array, the TEA5K mSNP array, which comprises 5,781 liquid-phase probes based on the Genotyping by Target Sequencing (GBTS) system. Using this array, we genotyped 231 developed tea cultivars, revealing that genetic similarity within the same cultivar ranged from 92.53–97.95%, whereas genetic similarity between different cultivars generally remained below 82.36%. Furthermore, utilizing this array, we constructed a high-density genetic map consisting of 3,274 markers, covering a total genetic distance of 2,225.19 cM, with an average marker interval of 0.76 cM. The high-resolution genetic map facilitated the identification of multiple QTLs linked to eight amino acid components, as well as two molecular markers strongly associated with the albinoleaf trait in the 'Huangjinya' cultivar, both mapped to chromosome eight. Moreover, we applied the array to analyze the population structure and phylogenetic relationships of 519 tea germplasm, classifying them into three major groups: wild accessions, landraces, and modern cultivars. Notably, modern cultivars exhibited lower genetic diversity compared to landraces. Additionally, we observed substantial genetic differentiation between wild resources and modern cultivars, with minimal to no gene flow from wild populations into domesticated cultivars. These findings suggest that modern tea breeding faces an "improvement bottleneck," a challenge similar to that encountered in other perennial crops.In conclusion, the TEA5K mSNP array is presented as a flexible, cost-effective, and low-maintenance genotyping tool that significantly enhances both genetic research and molecular breeding in tea plants. By providing a robust platform for genome-wide analysis and facilitating the identification of key QTLs, this tool offers valuable insights for improving the genetic diversity and agronomic performance of tea cultivars.

# Genome-wide association study revealed the regions associated with important agronomic traits in melon

Chengcheng Shen[1,3], Yushun Li[1], Wenjiao Wang[2], Na Zhang[3], Xuejun Zhang[1], Bin Liu[1]

[1] *Biological Breeding Laboratory, Xinjiang Uygur Autonomous Region Academy of Agricultural Sciences, Urumqi, China*

[2] *College of Horticulture, Shanxi Agricultural University, Jinzhong, China*

[3] *College of Horticulture, China Agricultural University, Beijing, China*

Presenting author(s) e-mail: shenchengcheng0608@163.com

## Abstract

Melon (*Cucumis melo* L.) exhibits considerable phenotypic diversity and holds a significant share in the global consumer market. The identification of causal genes underlying key agronomic traits is essential for the targeted genetic improvement and breeding of melon varieties with enhanced yield and quality. As a powerful approach, genome-wide association studies (GWAS) have facilitated the discovery of numerous genes associated with important traits such as monoecy, flesh color, peel color, acidity, rind sutures, and carpel number. Nevertheless, many traits critical to productivity remain unexplored at the genetic level. In this study, we assembled a diverse collection of 1,244 melon accessions originating from over 42 countries. Utilizing high-coverage whole-genome sequencing and multi-environment phenotypic investigations, we performed GWAS on approximately 30 horticulturally important traits. Our analysis revealed several genomic regions significantly associated with these traits. Subsequent haplotype analysis and examination of gene expression patterns further pinpointed potential causal mutations and candidate genes. These findings enhance our understanding of the molecular mechanisms governing melon growth and development, and provide valuable genetic resources for molecular breeding, thereby accelerating the development of improved melon varieties.

# Establishment of CRISPR/Cas9 Editing System Based on Efficient Somatic Embryogenesis of *Eustoma grandiflorum*

Xingyu Zhou

*Yunnan University*

E-mail: 2792610550@qq.com

## Abstract

*Eustoma grandiflorum* belonging to Gentianaceae, is a well-known cut flower, which also called ‘prickle-less rose’. Compared with the roses, the number of commercial cultivars of *E. grandiflorum* is limited, probably related to the narrow genetic background of this species. CRISPR-based genome editing to manipulate crop traits is a straightforward and rapid system, which is a powerful tool for crop breeding. However, efficient CRISPR system was reported rarely in ornamental plants. Here, we report an efficient genome editing system for *E. grandiflorum* based on somatic embryogenesis. Two CRISPR/Cas9 vectors, pCAMBIA1300-pYAO: Cas9 and AtU6-DiKO, were recruited to modify the *EgPDS* gene by constructing three single-target sites and a double targets, respectively. Seven albino seedlings of 'Champagne' and 16 albino seedlings of 'light purple' were identified by Agrobacterium-mediated infection of calli, indicating *PDS* was knock-out. Both of them were positive seedlings through PCR identification, and the sequencing results showed that various types of amino acid base were generated, including deletion, substitution and insertion. Those results suggested that we successfully established a CRISPR/Cas9 editing system in *E. grandiflorum* based on somatic embryogenesis in two cultivars. The CRISPR system provides an important theoretical basis and technical support for the breeding of ornamental varieties and the innovation of germplasm resources.

# Research on the Differentiation Technology and Mechanism of Callus from Different Explants of Pinus sylvestris var. mongolica

Lijuan Gao[1]

[1] *Qingdao Agricultural University, Qingdao 266109, P. R. China*

E-mail: 2640175492@qq.com

## Abstract

Pinus sylvestris var. mongolica has a low reproductive efficiency. Tissue culture technology can not only make up for the shortcomings of traditional reproduction but also provide core support for resistance breeding and germplasm innovation by establishing an efficient regeneration system. Currently, there is still a problem of difficult differentiation in tissue culture technology. This study starts with the seeds of superior trees of Pinus sylvestris var. mongolica and uses tissue culture technology as a means to focus on the differentiation technology and mechanism of callus. It observes the morphology and differentiation process of callus at different stages, and measures the contents of superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), soluble sugar (SS), and starch in callus at different growth stages, as well as the activities of endogenous hormones such as auxin (IAA), gibberellin (GA), and abscisic acid (ABA). It also conducts Library-free DIA quantitative proteomics analysis to explore the best differentiation scheme of callus from different explants (mature embryos, female gametophytes, and cotyledonary stem segments) of Pinus sylvestris var. mongolica. The callus induction rate of mature embryos and female gametophytes was significantly improved. The metabolic activity of the callus after the third subculture was determined to be higher through physiological index and endogenous hormone measurement, which was more conducive to callus differentiation. In the study of Pinus sylvestris var. mongolica stem segments with cotyledons, it was found that the longer the interaction time between the explants and the callus, the stronger the differentiation ability. The number of differentially expressed proteins was less when the interaction time was longer, and the callus was closer to the differentiated callus than that with a shorter interaction time. It was controlled by a ternary balance system of energy metabolism- protein homeostasis - hormone response.

# CsUGT89A2 enhances tea plant resistance to Toxoptera aurantia by mediating flavonoid glycosides biosynthesis

Xingrong Zhou[1†], Dingli Chen[1†], Fengyun Tian[1], Jie Ma[1], Mei Chen[1], Youshudi Xie[1], Changli Yang[1], Yanglin Liang[1], Houhong Xiao[1], Xue Dong[1], Di Yang[2], Yingqin He[1], Xinlong Dai[1]*, Yan Li[3]*

[1] *College of Tea Science, Guizhou University, Guiyang 550025, Guizhou Province, China.*

[2] *School of Pharmaceutical Sciences, Guizhou University, Guiyang 550025, China.*

[3] *Guizhou Tea Research Institute, Guizhou Academy of Agricultural Sciences, Guiyang 550006, Guizhou, China.*

E-mail: xldai@gzu.edu.cn; rhu5@gzu.edu.cn

## Abstract

Tea plant [*Camellia sinensis* (L.) O. Kuntze] is a globally important crop but is severely threatened by *Toxoptera aurantia* infestations, which impact yield and safety. However, the response of tea plants to aphid feeding remains largely unexplored. This study investigates the feeding behavior of *T. aurantia* on different cultivars and identifies ‘Huangjinya’ and ‘Qiancha 1’ as susceptible and resistant cultivars, respectively. Transcriptome analysis revealed that *CsUGT89A2* was significantly upregulated in response to *T. aurantia* infestation. *In vitro* biochemical assays demonstrated that *CsUGT89A2* encodes a flavonoid 7-glycosyltransferase that catalyzes the conversion of flavonoids and UDP-glucose into flavonoid 7-*O*-glucosides. *In vivo*, silencing *CsUGT89A2* significantly reduced flavonoid glycoside accumulation. To further clarify the role of *CsUGT89A2* in tea plant resistance to *T. aurantia*, we used tobacco and tea flowers to evaluate aphid feeding and reproduction under chemical treatment, gene silencing, and gene overexpression conditions. Statistical analysis showed that, compared with flavonoids, the application of flavonoid 7-*O*-glycosides significantly reduced *T. aurantia* reproductive capacity. Furthermore, compared with the control, overexpression of *CsUGT89A2* significantly reduced the reproductive ability of aphids, while its silencing increased reproductive rates. Overall, our findings demonstrate that *CsUGT89A2* mediates flavonoid glycosylation and enhances insect resistance in tea plants by increasing flavonoid glycoside levels, offering new insights into the role of flavonoid glycosides in the insect resistance of *C. sinensis*.

# Chromosome-Level Genome Assembly of Kiwifruit (Actinidia arguta) Reveals Mechanisms of Anthocyanin Regulation

Yukuo Li[1,2], Zhe Song[1,3], Xu Zhan[1,4], Xiaohan Li[1,3], Lingshuai Ye[1], Miaomiao Lin[1,2], Ran Wang[1], Hailei Huang[5],Jian Guo[6], Leiming Sun[1], Hong Gu[1], Jinyong Chen[1], Jinbao Fang[1] and Xiujuan Qi[1,2]

[1] *National Key Laboratory for Germplasm Innovation & Utilization of Horticultural Crops, Zhengzhou Fruit Research Institute, Chinese Academy of Agricultural Sciences, Zhengzhou 450009, China*

[2] *Zhongyuan Research Center, Chinese Academy of Agricultural Sciences, Xinxiang 453500, China*

[3] *School of Agricultural Sciences, Zhengzhou University, Zhengzhou 450001, China*

[4] *College of Horticulture and Plant Protection, Henan University of Science and Technology, Luoyang 471000, China*

[5] *Shiyan Economic Crops Research Institute, Shiyan 442099, China*

[6] *College of Horticulture Science and Engineering, Shandong Agricultural University, Tai'an 271018, China*

E-mail: lxhan1007@163.com

## Abstract

Actinidia arguta is a newly emerged, commercially cultivated Actinidia species. A. arguta has a beautiful appearance and is rich in anthocyanin, and is thus highly welcomed by consumers. However, the mechanism of anthocyanin regulation in A. arguta remains unclear. In this study, we assembled the nearly complete genome of the first red A.arguta cultivar, 'Tianyuanhong', with an N50 of 21 Mb. Comparative genome analysis revealed a role of the expansion/contraction of gene families in the species-specific trait formation of A. arguta. Through verification of transient overexpression and stable transformation, RNA-seq analysis revealed a key bHLH transcription factor, AaBEE1, which negatively regulates anthocyanin biosynthesis. DAP-seq analysis combined with Y1H, EMSA, Chip-qPCR and LUC sug-gested that AaBEE1 binds to the G-box of the AaLDOX promoter and suppresses its expression. Overall, we assembled the genome of A. arguta and clarified its AaBEE1-AaLDOX module-mediated molecular mechanism of anthocyanin regulation.

# Phytochromes modulate calmodulin-like protein-mediated PIF1 suppression to optimize seed germination under stress

Jing Kang[1], Xiaoyang Liu[1], Xiaowei Ye[1], Hongling Li[2], Sheng Xu[2], Shuai Zhao[1], Shangwei Zhong[2], Hui Shi[1*]

[1] *College of Life Sciences, Capital Normal University, Beijing 100048, China*

[2] *State Key Laboratory of Protein and Plant Gene Research, School of Life Sciences and School of Advanced Agricultural Sciences, Peking University, Beijing 100871, China*

E-mail: hui.shi@cnu.edu.cn; kangjing0202@163.com

## Abstract

Seed germination is precisely regulated by environmental cues. Light is essential for small-seeded species like *Arabidopsis* and lettuce. Conversely, abiotic stresses such as salinity and osmotic stress severely inhibit germination, posing significant challenges to agriculture. However, the mechanisms underlying how seeds integrate light and stresses to achieve optimal germination remain largely elusive. Here, we demonstrate that light significantly promotes seed germination under salt stress and mannitol-induced osmotic stress, and the photoreceptor phytochrome B (phyB)-mediated PHYTOCHROME-INTERACTING FACTOR 1 (PIF1) acts as a central regulator governing seed stress tolerance. We identify a salt and mannitol-stimulated calcium sensor calmodulin-like (CML) protein physically interacts with PIF1 in a calcium-dependent manner and acts upstream of PIF1 to promote seed germination under salt and osmotic stresses. We further show that CML-PIF1 interactions suppress PIF1's self-interaction, oligomerization and DNA-binding capabilities, thereby relieving PIF1's repression on seed germination, while the interactions between PIF1 and mutated CML significantly reduce and CML mutation releases its suppression effect on self-interaction and oligomerization of PIF1. Moreover, photo-activated phyB also interact with CML and promote the CML-PIF1 interaction. Our studies shed light on the mechanistic framework by which the phyB-CML-PIF1 module integrates light and stress signals to initiate seed germination.

# The influence of SlCYP704B1 gene in the P450 family on male sterility in tomato

Ding Yingfeng[1],Shen Hui[1], Ding Yumei[2], Bao Huihui[1],Chen Fengqiong[1], Wang Zhiqiang[1], HeZhao Yilan[1], Yang Zheng'an[1]

[1] *College of Landscape and Horticulture, Yunnan Agricultural University, Kunming 650201, Yunnan, China*

[2] *College of Food Science and Technology, Yunnan Agricultural University, Kunming 650205, Yunnan, China.*

E-mail: 1872248406@qq.com

## Abstract

The tomato (*Solanum lycopersicum* L.) is an economically important crop cultivated worldwide. The development of male sterility is highly valuable for tomato breeding, as the use of sterile males reduces the cost of $F_1$ hybrid seed production. The P450 family represents one of the largest transcription factor families in plants and plays extensive roles in various biological processes, including growth, development, and stress resistance. Previous studies have shown that the *SlCYP704B1* gene is essential for tapetum development in Arabidopsis and rice, where it participates in sporopollenin biosynthesis and pollen exine formation. To investigate the function of *SlCYP704B1* and assess its potential as a candidate gene for inducing male sterility in tomato, we employed CRISPR/Cas9-mediated genome editing strategies via *Agrobacterium*-mediated transformation. The full-length *SlCYP704B1* gene (1710 bp) was cloned from tomato pollen cDNA. Pollen staining revealed that the proportion of non-viable pollen in *SlCYP704B1*-knockout plants reached 100%, significantly higher than that in wild-type plants (less than 10%; $P<0.01$). The sterile pollen exhibited wrinkled or abnormal morphology, whereas wild-type pollen grains were oval and plump. Compared with the wild type, the *SlCYP704B1* knockout positive strain showed normal floral organ differentiation, but the anther development was abnormal and curved, and the ovary was unable to enlarge to form a seeded fruit. Furthermore, qRT-PCR analysis showed that *SlCYP704B1* expression was highest in early young stamens among different tomato tissues.These results indicate that *SlCYP704B1* plays a critical role in pollen development and may contribute to male sterility in tomato, providing new insights into the molecular mechanisms underlying this trait.

# Transcriptomic Analysis Reveals a Regulatory Network of Adventitious Root Formation in Blueberry and Significance of *VcWOX4a* in Lateral Root Formation

Botian Zheng[1], Fanglong Li[1], Jiaqi Zhu[1], Guiliang Tang[2], Lulu Zhai[1], Jingying Wang[1], Tianye Liang[1], Xuyan Li[1*], Shaomin Bian[1*]

[1] *College of Plant Science, Jilin University, Changchun, Jilin 130062, China*
[2] *Department of Biological Sciences, Life Science and Technology Institute, Michigan Technological University, Houghton, MI, USA*

*E-mail: shmbian@jlu.edu.cn; xuyanli@jlu.edu.cn*

## Abstract

Despite the widespread use of stem cuttings for propagation, blueberry (*Vaccinium* spp.) adventitious root formation is hindered by its fibrous root system. Transcriptomic analysis across four morphological stages of AR development in blueberry stem bases revealed dynamic gene expression patterns: upregulation of *WOX11*, *EXPA4*, *EXPA8*, and *PILS1* during primordium initiation; changes in *LBD36*, *MYB93*, and *LRP1* during proliferation; and specific activation of *EXPB15* and *CYP735A1* during elongation. Comparative transcriptomics between perennial and juvenile cuttings uncovered stage-dependent regulatory mechanisms. Furthermore, 39 *VcWOX* genes were identified, with functional analysis demonstrating that *VcWOX4a* inhibits lateral root development, highlighting the role of *WOX* genes in shaping blueberry root architecture.

# Genome-wide identification of the bHLH transcription factor family and the regulatory roles of *PbbHLH74* in response to drought stress in *Phoebe bournei*

Ningning Fu[a,1], Li Wang[a,2], Qinglin Sun[a,3], Qiguang Wang[a,4], Yuting Zhang[a,b,5], Xiao Han[a,b,6], Qi Yang[a,b,7], Wenjun Ma[c,8], Zaikang Tong[a,b,9], Junhong Zhang[a,b,10]

[a] *State Key Laboratory of Subtropical Silviculture, School of Forestry & Biotechnology, Zhejiang A&F University, Lin'an, Hangzhou 311300, Zhejiang, P. R. China*

[b] *Zhejiang Key Laboratory of Forest Genetics and Breeding, Zhejiang A&F University, Lin'an, Hangzhou 311300, Zhejiang, P. R. China*

[c] *State Key Laboratory of Tree Genetics and Breeding, Key Laboratory of Tree Breeding and Cultivation of State Forestry Administration, Research Institute of Forestry, Chinese Academy of Forestry, National Innovation Alliance of Catalpa Bungei, Beijing, China*

E-mail: 2024202012006@stu.zafu.edu.cn, 20206020411010@stu.zafu.edu.cn, 369288208@qq.com, wgg137574662380@163.com, zhangyt@zafu.edu.cn, hanx2017@zafu.edu.cn, qiyang@zafu.edu.cn, zktong@zafu.edu.cn, zhangjunhong@zafu.edu.cn

## Abstract

*Phoebe* species constitute a large portion of subtropical forestry, which are key players in biomass resources. However, abiotic stresses such as drought stress severely limit the growth and development of *P. bournei*, and even lead to its death. It has been shown that basic helix-loop-helix (*bHLH*) as the second largest transcription factor family plays essential roles in response to multiple stresses in plants. However, little information of *bHLH* family is available in *P. bournei*. In this study, 130 *PbbHLH*s were identified and classified into 24 subfamilies. Then, the *bHLH* domain, conserved motifs and gene structures, evolutionary patterns and protein structural features were probed. The expression levels of 17 *PbbHLH*s were differentially induced by PEG and ABA by RT-qPCR analysis, indicating that they may be involved in drought stress response. Characterization of the drought candidate gene *PbbHLH74* showed that it was transcriptionally active and localized in the nucleus. Heterologous transformation of *PbbHLH74* into yeast improved cellular tolerance to drought stress. Meanwhile, overexpression of *PbbHLH74* in *Arabidopsis* showed higher seed germination, plant biomass and expression levels of stress-related genes under drought conditions. Through the hairy root technique, overexpression of *PbbHLH74* in *P. bournei* improved drought tolerance by enhancing root development and expression levels of genes involved in ABA-dependent and ROS scavenging pathways. Moreover, PbbHLH74 might positively regulate the expression of *PbPOD* by Y1H and dual-luciferase reporter assays. Overall, these results elucidated the structure and evolution of the *PbbHLH* family, in which *PbbHLH74* could be applied to molecular assisted breeding for drought tolerance in *P. bournei*.

# The ERF transcription factor VcERF061 activates *VcF3'5'H4* expression and promotes light-induced anthocyanin biosynthesis in blueberry

Ting Yan

*College of Forestry, Guizhou University*

E-mail: 1741195688@qq.com

## Abstract

Light-induced anthocyanin synthesis in blueberries involves complex transcriptional regulation. This study investigates the role of the transcription factor VcERF061 in regulating the key anthocyanin biosynthesis gene *VcF3'5'H4* in rabbiteye blueberry 'Powderblue'. Bioinformatics analysis and cloning results show that *VcERF061*, a nucleus-localized member of the AP2/ERF family with a total length of 855 bp, may interact with the *VcF3'5'H4* promoter, which contains multiple light-responsive elements. Yeast one-hybrid and split-luciferase complementation assays demonstrated that VcERF061 binds directly to the *VcF3'5'H4* promoter. Electrophoretic mobility shift assays further confirmed specific binding to an ACGGGTCCCC motif. Subcellular localization revealed nuclear accumulation of VcERF061, consistent with its function as a transcription factor. Overexpression of *VcERF061* in blueberry calli and tobacco leaves significantly upregulated *VcF3'5'H4* expression and increased anthocyanin accumulation under light. These results demonstrate that VcERF061 activates *VcF3'5'H4* expression by directly binding to its promoter, thereby promoting light-induced anthocyanin biosynthesis in blueberry. This study provides mechanistic insight into the regulatory role of ERF transcription factors in light-mediated anthocyanin accumulation.

# CiNF-YB18, Encoding a Nuclear Factor Y Family Protein, Regulates Fruit Enlargement in Pecan (Carya illinoinensis)

Nan Gao[1], Zhengjia Wang[2], Zhengfu Yang[3]

*National Key Laboratory for Development and Utilization of Forest Food Resources, Zhejiang A&F University, Hangzhou 311300, China.*

E-mail: gn@stu.zafu.edu.cn

## Abstract

The pecan (*Carya illinoinensis*) is a globally recognized tree nut and a significant woody oil-bearing species. Fruit size is a crucial determinant of yield in economic forests, with considerable variation observed among different pecan varieties. However, the molecular mechanisms underlying this trait remain elusive. We previous studies found the period from pollination to rapid expansion is the critical phase determining fruit size, and quantitative phosphoproteomic analysis revealed numerous phosphorylated sites, peptides, and proteins potentially implicated in regulating fruit enlargement. Among them, a nuclear factor Y family proteinwas differentially phosphorylated during fruit development. In this study, we analyzed the pecan NF-Y gene family and preliminarily investigated its regulatory role during pecan fruit development. The main results are as follows:1. Based on the pecan genome, a total of 41 CiNF-Y genes were identified, comprising 11 *CiNF-YA*, 18 *CiNF-YB*, and 12 *CiNF-YC* members. 2. *CiNF-YB18* was differentially phosphorylated with high expression levels in leaves, female flowers, and during early fruit development. Overexpression of *CiNF-YB18* led to increased rosette leaf size and number, as well as larger mature seeds and enhanced thousand-seed weight. 3. CiNF-YB18 was predominantly localized in the nucleus. Mutations at its phosphorylation sites did not alter its subcellular localization, and the protein lacked transcriptional activation activity. Both yeast two-hybrid and bimolecular fluorescence complementation (BiFC) assays confirmed that CiNF-YB18 interacts with CiNF-YC1/3/5/8/10 to form heterodimers. Furthermore, CiNF-YC1/3/5/8/10 were shown to interact with CiNF-YA4.Additionally, the phosphomimetic mutant CiNF-YB18$^{S20D}$ failed to interact with CiNF-YC1/3/5/8/10, whereas the dephosphorylation-mimetic mutant CiNF-YB18$^{S20A}$ retained interaction specifically with CiNF-YC8/10. These results indicate that phosphorylation modification modulates the interaction between CiNF-YB18 and its partner proteins. Taken together, this study elucidate the molecular mechanism of CiNF-YB18 mediated by phosphorylation regulating the early fruit enlargement and provide theoretical support for improving the yield of pecan.

# Squalene acts as a feedback signaling molecule in facilitating bidirectional communication between tea plants

Jieyang Jin[1], Wenkai Du[1], Yutong Hu[1], Xiaoyan Tang[1], Chuankui Song[1]

[1] *National Key Laboratory for Tea Plant Germplasm Innovation and Resource Utilization, Anhui Agricultural University, 230036 Hefei, Anhui, P. R. China.*

E-mail: sckfriend@163.com

## Abstract

Plants respond to environmental stimuli by releasing volatile organic compounds (VOCs), which play diverse roles in plant-to-plant interactions. Previous studies have primarily focused on how receiving plants respond. However, little is known about how these receivers communicate back to the emitter plants and their subsequent impacts. Our findings indicated increased plant tolerance when neighboring plants were present, suggesting bidirectional plant communication. Furthermore, we established a model to explore the role of signals from receiver plants, identifying squalene as a crucial feedback signal enhancing the cold tolerance in emitter plants by up-regulating *CsCBF5* expression. Further analysis using yeast one-hybrid analysis coupled with inhibition of brassinosteroid pathways suggested that squalene-induced castasterone (CS) accumulation directly activated *CsCBF5* expression modulated by *CsBES1/BZR1*. Overall, these results demonstrated the role of the squalene-CS-*BES1/BZR1-CBF5* pathway in the bidirectional communication between plants, expanding our understanding of plant interactions.

# An InDel variant in the promoter of the NAC transcription factor *MdNAC18.1* regulates apple fruit ripening

Qianyu Yue[1,2], Yinpeng Xie[2], Fengwang Ma[2], Qingmei Guan[2]

[1] *Shandong Agricultural University*

[2] *Northwest A&F University*

E-mail: qyue@sdau.edu.cn; qguan@nwsuaf.edu.cn

## Abstract

Apple (Malus domestica), as one of the most widely cultivated and consumed fruits worldwide, exhibits a wide range of maturity periods that are crucial for meeting market demands and consumer preferences. However, the apple industry faces the challenge of a narrow cultivar structure, and the identification of genes related to fruit maturity and the understanding of their regulatory mechanisms remain limited. Here, we report MdNAC18.1 was identified as being tightly correlated to fruit ripening Through genome-wide association studies (GWAS). A 58-bp insertion/deletion (Indel-58 bp) in the promoter region of *MdNAC18.1* was found to causes variation in apple fruit ripening. In the early-ripening genotype MdNAC18.1E, the deletion of Indel-58 weakens the inhibitory effect of MdAGL11 on MdNAC18.1, thereby promoting fruit ripening. In contrast, in the late-ripening genotype MdNAC18.1L, the insertion of Indel-58 enhances this inhibitory effect, delaying fruit maturation. Specifically, the deletion of Indel-58 bp reduces the binding affinity of MdAGL11, a negative regulator of fruit ripening, to the *MdNAC18.1* promoter, thereby promoting *MdNAC18.1* transcription and accelerating fruit ripening. This natural variation in MdNAC18.1 provides important genetic insight into the regulation of apple fruit ripening.

# The Coupling of Light and Potassium Supplements Optimize Strawberry Fruit Performance Through Plant Nitrogen Uptake, Assimilation and Translocation to Fruit

Xiangnan Xu

*Beijing Academy of Agriculture and Forestry Sciences*

E-mail: 15101654599@163.com

## Abstract

The greenhouse strawberry usually had undesirable quality in deep-winter due to inefficiency nitrogen use, so this experiment investigated if light and potassium supplements could alleviate this drawback and the mechanism beneath. The plants were subjected to two light conditions coupling with two leaf sprays, yielding 4 different groups: 1) control (NDN), 2) 10 mM KCl spray (NDK), 3) 3h supplemental light (SDN) and 4) 10 mM KCl spray + 3h supplemental light (SDK). The supplemental light enhanced leaf nitrate uptake (36.5 %), fruit nitrogen concentration (8.9 %), per fruit nitrogen accumulation (41.3 %) and single fruit mass (50.1 %); the KCl spray decreased leaf nitrogen concentration (4.0 %) but improved the activity of leaf nitrate reductase (60.0 %), nitrite reductase (12.9 %) and glutamine synthase (19.3 %), and fruit sugars concentration (19.7 %). Comparing with NDN, SDK increased both fruit size (38.6 %) and sugars content (23.8 %). The leaf transcriptomic analysis suggested the differentially expressed genes in SDK vs NDN were significantly enriched in nitrogen metabolism, alanine, aspartate and glutamate metabolism, cyanoamino acid metabolism and tryptophan metabolism based on the KEGG pathways. The key genes were screened out from these pathways and examined by qRT-PCR test, and the results confirmed that the SDK had up-regulated the leaf expression of Nitrate reductase, Ferredoxin-nitrite reductase and (NADH) glutamate synthase. Hence, the combination of supplemental light and potassium spray enhanced plant source-sink nitrogen coordination through nitrogen uptake, assimilation, translocation and key genes expression, thereby improving strawberry fruit performance in deep-winter greenhouse.

# Evaluate Water Use Efficiency in Mungbean (*Vigna radiata* (L.)): Genotypic Variation and Phenotyping Approaches

Yayue Zhong [1], Vijaya Singh [2], Mark Dieters [1], Kaye Basford [1], Yash Chauhan [3], Malcolm Hunter [1], Vivi Arief [1]

[1] *School of Agriculture and Food Sustainability, The University of Queensland, QLD, Australia*

[2] *Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, QLD, Australia*

[3] *Department of Agriculture and Fisheries, Kingaroy, QLD, Australia*

E-mail: joyce.zhong@uq.edu.au

## Abstract

Mungbean (*Vigna radiata* (L.)) is a short-duration legume valued for its nutritional quality, contribution to sustainable cropping, and relatively low water requirement compared with other summer crops. However, yield in mungbean is often constrained by limited water availability, underscoring the importance of improving water use efficiency (WUE). Accurate and repeatable phenotyping methodologies are essential to accelerate breeding for WUE. We applied Hunter's inverted-bottle pot (IBP) system to phenotype WUE and related traits in nine Australian commercial varieties and 76 Australian Grain Genebank accessions under well-watered glasshouse conditions. The IBP system allows precise estimation of water use and repeatable screening across many genotypes. Two indices of WUE were calculated: WUE_bio (shoot dry weight per unit water use) and WUE_yield (seed weight per unit water use). Substantial genotypic variation was detected. WUE_bio ranged from 8.85 to 11.73 g/kg and WUE_yield from 0.41 to 1.97 g/kg. Genotype rankings differed between indices, but some accessions combined both high WUE_bio and WUE_yield. Among commercial cultivars, 'Berken' ranked highest in WUE_yield (1.97 g/kg) and showed the greatest harvest index (0.20). Flowering duration also varied markedly, from 35 to 80 days after sowing, while total water use ranged from 12,000 g to over 30,000 g across genotypes. These contrasting strategies indicate that both phenology and water-use dynamics contribute to observed differences in WUE. Our findings establish the IBP system as a repeatable and scalable approach for WUE phenotyping in mungbean and reveal substantial genetic variability for this trait. These results provide a foundation for targeted selection and breeding of mungbean cultivars with improved water-use efficiency and yield stability in dryland environments.

# Characterization of nitrate utilization efficiency in tea plants based on leaf chlorate sensitivity

Wenjing Zhang, Lizhi Long and Jianyun Ruan

*Tea Research Institute, Chinese Academy of Agricultural Sciences, Hangzhou 310008, China*

E-mail: Zhangwenjing@tricaas.com

## Abstract

As leaf-harvested perennial crops, tea plants (*Camellia sinensis*) have a high nitrogen (N) demand to sustain synthesis of amino acids in young tissues. However, as nitrate accounts for 51%–63% of leached nitrogen in tea plantations, enhancing nitrate utilization efficiency (NiUE) represents a potential target for establishing a sustainable tea production system. While NiUE phenotyping in herbaceous plants is well-established, rapid assessment tools for woody plant remain underdeveloped. Ten representative tea plant cultivars from various tea regions in China were selected. Fresh branches of 10 cultivars were hydroponically cultivated and subjected to potassium nitrate ($KNO_3$) and potassium chlorate ($KClO_3$) treatments. The sensitive cultivar, Zhenong 117 (ZN117), showed a decrease in SPAD and Fv/Fm values following $KClO_3$ treatment, while the tolerant cultivar, Teiguanyin (TGY), exhibited minimal significant changes. After 5 days of cultivation, the $^{15}N$ concentration and proportion in new shoots of ZN117 were significantly higher (1.56-fold and 1.13 fold) than those in TGY. Based on the time-course transcriptomes, ZN117 exhibited faster transcriptional reprogramming, with bHLH/SBP TFs and NRT2.4 transporters upregulated during early stress (6–24 h). Field trials demonstrated significant increases in total N and free amino acids in ZN117 young shoots with increased N application. The mobilization of N derived from winter-applied $^{15}N$-urea, applied to mature leaves of the two contrasting genotypes was further tracked during spring shoot flushes.The decline of $^{15}N$ content in mature leaves occurred more rapidly in ZN117 than in TGY, and ZN117 exhibited 2.24-fold higher $^{15}N$ in content new shoots compared to TGY. This study pioneers a chlorate-sensitivity-based methodology to evaluate NiUE in tea plants and deciphers genotype-specific responses to N rates.We present the identified biomarkers (*bHLH, SBP, NRT2.4*) offer targets for molecular breeding, while genotype-specific N response curves inform precision fertilization strategies. This scalable method holds promise for nutrient-utilization-efficient cultivar screening across tree crops.

# Population genomics provide insights into the local adaptation and future climate-induced vulnerability of a subtropical endangered tree

Yan Liu[1,2]

[1] *National Key Laboratory for Development and Utilization of Forest Food Resources, Zhejiang A&F University, Hangzhou, 311300, China.*

[2] *Zhejiang Key Laboratory of Forest Genetics and Breeding, Zhejiang A&F University, Hangzhou, 311300, China.*

E-mail: liuy12345671@gmail.com

## Abstract

Elucidating the genomic underpinnings of adaptive variation is important for guiding tree conservation and management in the context of global climate change. Nevertheless, little information is available concerning the genetic mechanisms driving climate adaptation in *Phoebe bournei*, a near-threatened subtropical tree species endemic to China. Whole-genome resequencing was conducted on 362 individuals from 27 natural populations across the geographical range of the species. Genome-environment association analyses were employed to identify 1,556 climate-associated variants and 167 candidate genes associated with temperature and precipitation variables. Through functional annotation and expression profiling, pivotal genes, including *TRX-M4* and *FBD1,* were identified as integral to drought and heat stress responses, with adaptive alleles displaying distinct geographic frequency distributions. Divergent evolutionary trajectories were deduced among populations, with southeastern populations distinguished by elevated genetic diversity and strong signatures of local adaptation. Nevertheless, projections derived from RONA and gradient forest models suggest that these southeastern populations will face substantial genomic offset under future climate scenarios, signaling heightened vulnerability. This study provides the first genome-wide perspective into the adaptive evolution of *P. bournei* and offers a robust foundation for its conservation and climate-resilient management.

# The CLE45 peptide maintains quality, delays senescence, and extends shelf life through regulating fruit energy metabolism in kiwifruit

Sibo Jiao[1], Huizhen Bao[1], Bowen Duan[1], Yilin Li[1], Junpeng Niu[1,*], Guodong Wang[1,*]

[1] *College of Life Sciences, Engineering Research Center of High-Valued Utilization of Fruit Resources in Western China, Ministry of Education, Shaanxi Normal University, Xian 710119, China*

E-mail: adenine404@foxmail.com

## Abstract

Known as peptide hormones, small signaling peptides play diverse functional roles in plants, yet their application in fruit preservation remains largely underexplored. Here, the effect of the CLAVATA3/EMBRYO SURROUNDING REGION-RELATED 45 (CLE45) peptide on the postharvest shelf life of 'Xuxiang' kiwifruit (*Actinidia deliciosa*) was studied. The CLE45 peptide effectively maintained fruit morphology and firmness in a concentration-dependent manner. CLE45 peptides reduced the accumulation of soluble solids, slowed the degradation of titratable acid, and mitigated the decrease of lightness and hue. Furthermore, CLE45 treatments suppressed relative conductivity, respiratory rate, and ethylene release rate. The flavor and taste were also modulated by exogenous CLE45 peptides. Comprehensive evaluation of distinct quality parameters indicated that 5 μmol $L^{-1}$ CLE45 provided the optimal preservation effect on delaying senescence and deterioration. Further assays indicated that 5 μmol $L^{-1}$ CLE45 preserved cellular ultrastructure, inhibited starch degradation and total sugar accumulation, and enhanced energy metabolism. These observations were supported by the alterations of key enzymes and genes involved in glycolysis, the tricarboxylic acid cycle, the pentose phosphate pathway, gluconeogenesis, and the electron transport chain. Targeted metabolomics identified pivotal pathways and differential metabolites associated with energy metabolism that is correlated with 5 μmol $L^{-1}$ CLE45-mitigated postharvest deterioration. Altogether, exogenous application of the CLE45 peptide can maintain the quality of 'Xuxiang' kiwifruit by preserving morphology, color, texture, flavor, taste, nutrition, physiological stability, and ultrastructural integrity, thereby delaying senescence and extending shelf life. Hence, our study proposes a fresh outlook on the potential application of peptide hormones in postharvest fruit preservation.

# Regulatory Mechanism of the C2H2 Zinc Finger Protein PagZFP12 in Modulating Xylem Development of Poplar

Minhui Li[1], Mengzhu Lu[1], Jin Zhang[1]*

[1] *National Key Laboratory for Development and Utilization of Forest Food Sources, College of Forestry and Biotechnology, Zhejiang A&F University, Hangzhou, Zhejiang 311300, China*

E-mail: lmh03242023@163.com

## Abstract

C2H2 zinc finger proteins (C2H2-ZFPs) are a pivotal family of transcription factors in plants, modulating gene expression via specific DNA binding and playing key roles in plant growth, development, and stress adaptation. This study focused on the function and regulatory mechanism of C2H2-ZFP PagZFP12 in xylem development of poplar (*Populus alba* × *P. glandulosa*) '84K', aiming to provide a theoretical basis for deciphering the molecular network of woody plant secondary growth and advancing fast-growing, high-quality forest breeding. PagZFP12—a transcription factor strongly negatively correlated with lignin monomer synthase genes—was screened via co-expression analysis of key lignin biosynthesis genes. GUS staining of *proPagZFP12::GUS* transgenic poplars revealed its specific high expression in cambium and developing xylem. Three-month-old *pagzfp12a/pagzfp12b* knockout lines (generated by CRISPR-Cas9) showed increased plant height, thicker ground diameter, expanded pith cavity, narrowed xylem, fewer xylem cell layers, thicker xylem fiber cell walls, expanded cambium, and elevated lignin content compared with the wild-type plants. PagZFP12 localized to the nucleus with transcriptional activation activity; yeast two-hybrid screening identified vascular-specific interactors such as WRKY20 and bZIP21. Subsequent studies will use CUT&Tag to screen its direct downstream targets and yeast one-hybrid to verify transcription factor-target promoter interactions. In conclusion, as a transcriptional activator, *PagZFP12* may negatively regulate poplar cambium activity and xylem development by modulating downstream inhibitory targets. Its functional loss promotes biomass and secondary growth, offering a novel target for poplar wood yield and quality improvement.

**Study on the Regulation of *GhJOX2* in Cotton Verticillium Wilt Resistance**

Chenxi Pang

*Zhengzhou University*

E-mail: pcxixi99@163.com

## Abstract

Verticillium wilt severely threatens cotton production, and developing resistant varieties via genetic engineering is key to its control. Jasmonic acid (JA) induces broad-spectrum disease resistance but inhibits plant growth when overaccumulated; jasmonate oxidase, which hydroxylates JA to 12-hydroxyjasmonic acid, is critical for plant growth. Our lab previously identified a JA oxidase gene *GhJOX2* associated with cotton Verticillium wilt resistance. *GhJOX2* expression was downregulated in 'Zhongzhimian 2' and 'Jimian 11' after Verticillium dahliae inoculation. Silencing *GhJOX2* in cotton enhanced resistance, increased JA content, and upregulated *GhAOS* (JA biosynthesis), *GhJAZ*, *GhPDF1.2*, *GhPR4* (resistance genes), with no significant change in *MYC2* (JA signaling activator). Heterologous overexpression of *GhJOX2* in Arabidopsis increased susceptibility to V. dahliae and downregulated JA/ROS-related genes.Yeast two-hybrid showed GhJOX2 interacted with GhSAMS1 (ethylene biosynthesis); silencing *GhSAMS1* enhanced cotton resistance and upregulated *GhPPO*, *GhNOA*, *GhJAZ1*, *GhCHI*. Yeast one-hybrid revealed *GhJOX2* was positively regulated by *GhTCP9*, whose expression decreased in resistant/susceptible cotton under V. dahliae induction. GhSAMS1 and GhJOX2 co-localized at the cell membrane. In conclusion, *GhJOX2* negatively regulates cotton Verticillium wilt resistance; *GhTCP9* upregulate*s GhJOX2*, and GhJOX2 interacts with GhSAMS1 to modulate JA signaling-related genes for resistance regulation.

# The neglected role of autotoxic self-DNA played in the development of continuous cropping obstacle of *Eustoma grandiflorum*

Di He[1], Chunlian Jin[2], Fan Li[2], Jihua Wang[2], Peihua Zhang[2]

[1] *Yunnan University*

[2] *Floriculture Research Institute, Yunnan Academy of Agricultural Sciences*

E-mail: zhangpeihua@yaas.org.cn

## Abstract

Continuous cropping obstacle (CCO) seriously impedes the quality production of *Eustoma grandiflorum* (lisianthus). Growing evidence suggests that fragmented extracellular self-DNA (ex-DNA) from plant litter plays a crucial role in the development of negative plant-soil feedback, commonly known as CCO. However, the underlying mechanisms involving ex-DNA remain poorly understood.

In this study, we first confirmed the species-specific autotoxicity of ex-DNA through root litter addition (25 $\mu g \cdot g^{-1}$) and ex-DNA exposure (200 $\mu g \cdot mL^{-1}$) from lisianthus itself, alongside several closely (e.g., *Gentiana scabra*, *Gentiana rigescens*) and distantly (e.g., *Dianthus caryophyllus*, *Lactuca sativa*) related species. The results clearly demonstrated that only lisianthus root litter and ex-DNA significantly inhibited the growth and fitness of the tested seedlings. Furthermore, this inhibitory effect diminished with increasing phylogenetic distance from lisianthus. An ecotoxicity test was conducted with a range of ex-DNA concentrations (0, 50, 100, 200, and 400 $\mu g \cdot mL^{-1}$) to further investigate the effective toxic concentration and the underlying mechanisms. The results showed that the autotoxic effect intensified with increasing ex-DNA concentrations, with significant inhibition observed at 50 $\mu g \cdot mL^{-1}$. Subsequent physiological analyses revealed that ex-DNA induced significant oxidative stress and caused root tissue damage in lisianthus plants. Transcriptomic analysis further demonstrated that ex-DNA triggered strong defensive responses (e.g., plant–pathogen interactions and responses to fungi), while also suppressing essential growth processes (e.g., photosynthesis and energy metabolism). Nevertheless, this activated defensive response proved insufficient to protect against pathogens as evidenced by our inoculation test, which showed population buildup of *Fusarium oxysporum* in the rhizosphere increased under higher ex-DNA exposure.

Our findings reveal a potential pathway through which autotoxic self-DNA contributes to the development of CCO and suggest that overcoming CCO in lisianthus requires more concerns on the fate and removal of self-DNA in cultivated soil.

# Influence of Chlorella Sorokiniana and Plant Growth Regulators During the Micropropagation of Callicarpa Peichieniana

Yiteng Zhang [1,2], Manna Guo [1,2], Jinfeng Xu [1,2], Yuping Xiong [1], Junyu Liu [1,2], Guohua Ma [1], Songjun Zeng [1], Kunlin Wu [1,*] and Lin Fang [1,*]

[1] *Guangdong Provincial Key Laboratory of Applied Botany, South China Botanical Garden, Chinese Academy of Sciences, Guangzhou 510650, China*

[2] *University of Chinese Academy of Sciences, Beijing 100049, China*

E-mail: zhangyiteng@scbg.ac.cn

## Abstract

*Callicarpa peichieniana* is an important traditional Chinese medicinal plant with pharmacological benefits for digestive system diseases and wounds, as well as high ornamental value. The goal of this study is to establish an effective in vitro regeneration system in order to satisfy the expanding market demand. Extracts from algae can enhance the proliferation and rooting effect of adventitious buds and can improve the survival rate of transplantation. This study developed an in vitro regeneration system using apical bud explants of *C. peichieniana* associated with *Chlorella sorokiniana* (an alga species). Inter simple sequence repeat (ISSR) molecular markers confirmed the genetic fidelity of the regenerated plantlets. The highest number of adventitious buds (5.00 buds) was induced from the apical buds with 0.5 mg/L 6-BA in a Murashige and Skoog (MS) medium, and the highest proliferation coefficient (5.83) was achieved with 2.0 mg/L 6-BA. A rooting rate of 100% was achieved by using 0.1 mg/L NAA, MS with 50% macroelements, and 20 g/L sucrose, averaging 6.36 roots per explant and a root length of 1.32 cm. In all micropropagation stages, *C. sorokiniana* coexisted and proliferated alongside *C. peichieniana* materials. ISSR showed that the genetic fidelity of *C. peichieniana* regenerated plants was 95.45%. Coconut coir/perlite = 1:1 (*v*/*v*) was identified as the optimal transplantation substrate, achieving a 100% survival rate. The "*C. peichieniana–C. sorokiniana* association" in vitro regeneration system established in this study not only enables the mass production of high-quality regenerated plantlets but provides new ideas and demonstrations for culturing multiple species in the same in vitro system.

# Poplar *RBOHF* Mediate Tension Wood Formation

Dong Ning[1], MengZhu Lu[1]

[1] *National Key Laboratory for Development and Utilization of Forest Food Sources, College of Forestry and Biotechnology, Zhejiang A&F University,Hangzhou,China*

E-mail: 619843803@qq.com

## Abstract

Tension wood (TW) formation in angiosperm trees represents a fundamental adaptive response to gravitational stress, playing a crucial role in providing mechanical support and influencing wood quality. During the early stages of TW development, the superoxide anion ($O_2^{\cdot-}$) speciallyand these bursts accumulates and functions as a key signaling molecule that triggers cambial cell proliferation, differentiation, and ultimately, TW formation. However, the specific members of the RBOH family responsible for $O_2^{\cdot-}$generation and their functional relationship with TW formation remain uncharacterized. In this study, we demonstrate that *RBOHF* is involved in this process. Overexpression of *RBOHF* enhances TW formation and significantly increases the thickness of G-layer in TW fibers. Conversely, *RBOHF* knockout mutants exhibit reduced $O_2^{\cdot-}$accumulation and a marked decrease in G-layer thickness. These results provide the genetic evidence supporting that RBOHF regulates $O_2^{\cdot-}$ levels during TW formation.

# Seed coating agents disrupt soil function and bulb health by reshaping the rhizosphere microbiome and metabolome

Yi Lu[a †], Yuhan Wu[a,b †], Dan Gao[a*], Haobo Zhang[a,b], Zemin Yang[a,c], Huigan Xie[d], Shaobing Fu[d], Haiqing Wang[d], Xiwen Li[a, b, e*]

[a] *Institute of Chinese Materia Medica, China Academy of Chinese Medical Sciences, Beijing 100700, China*

[b] *College of Traditional Chinese Medicine, Jilin Agricultural University, Changchun 130118, China*

[c] *College of Traditional Chinese Medicine, Yunnan University of Chinese Medicine, Kunming 650500, China*

[d] *Nin Jiom Medicine Manufactory (Hong Kong) Limited, Hong Kong 999077, China*

[e] *Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences, Peking Union Medical College, Beijing 100193, China*

E-mail: Xiwen Li, xwli@implad.ac.cn; Dan Gao, dgao@icmm.ac.cn

## Abstract

Seed coating agents, while effective against soil-borne pathogens, may inadvertently disrupt plant-microbe interactions critical for stress resilience. This study investigates how thiabendazole-fludioxonil-metalaxyl-M seed coatings exacerbate bulb rot in Fritillaria cirrhosa through multi-omics analysis. Field trials revealed a 2.8-fold increase in rot incidence with coating use ($P < 0.001$), along with reduced bacterial diversity (25.1% decline in Simpson index) and enrichment of pathogens (such as Fusarium spp.). Microbial network topology analysis revealed severe degradation of microbial interconnectivity, manifested by a 38% reduction in edge numbers and decreased modularity ($P < 0.05$) compared to untreated controls. Metabolomic profiling identified significant enrichment in fundamental metabolic pathways under untreated soil, particularly pyrimidine metabolism and ABC transporter activity, along with marked accumulation of stress indicators such as D-gluconic acid (13.81-fold) and salicylic acid (117.20-fold). The mantel analysis revealed statistically robust associations ($P < 0.01$) between lipid metabolites and both fungal ($r = 0.776$) and bacterial ($r = 0.748$) taxa, while microbial abundance patterns showed significant correlation with soil health indices ($P < 0.01$), demonstrating the critical linkage between belowground biodiversity and ecosystem functioning. Collectively, seed coatings exerted a pronounced disruption on soil health, rhizosphere microbiome structure, and metabolic composition, with these alterations being primarily driven by the coating agents rather than bulb health status. These findings support a mechanistic model wherein seed coatings impair the bulb's innate "calling for help" response by inhibiting chemoattractant-mediated recruitment of beneficial microbiota while selecting those fungicide-tolerant taxa. The study challenges conventional fungicide-dependent management paradigms and advocates for the development of microbiome-compatible transplantation strategies in medicinal bulb crop production systems.

# Stage-specific nutrient management based on ecological stoichiometry improves photosynthesis and berry quality in ‘Powderblue’ rabbiteye blueberry

Fangping Zhong[1], Xiaoli An[2]

[1] *College of Forestry, Guizhou University*

E-mail: 1976164771@qq.com

## Abstract

This study integrated ecological stoichiometry and plant physiology to systematically explore the effects of nitrogen (N), phosphorus (P), and potassium (K) stoichiometric fertilization on leaf photosynthetic characteristics and fruit quality of seven-year-old ‘Powderblue’ rabbiteye blueberry across four key growth stages: flowering (S1), fruit setting (S2), young fruit (S3), and fruit expansion (S4). Graded fertilization treatments (1F, 5F, 10F, 15F, where F = theoretical nutrient demand) were applied to verify fertilizer requirements. Results showed fertilization significantly enhanced photosynthetic capacity: the S1-5F, S2-10F, S3-5F, and S4-5F treatments exhibited optimal photosynthetic performance, with notably higher net photosynthetic rate ($P_n$), stomatal conductance ($G_s$), and intercellular $CO_2$ concentration ($C_i$) than other treatments in the same stage. The S1-5F, S2-10F, S3-15F, and S4-1F treatments effectively increased fruit longitudinal/transverse diameters and single fruit weight. Anthocyanin content was positively correlated with fertilization rate, peaking at 3.55 mg/g under S2-15F, while appropriate flowering-stage fertilization (1F/5F) increased soluble sugar content, reduced titratable acid, and improved sugar-acid ratio. Stoichiometric analysis determined optimal per-plant nutrient demands for S1–S4: N (4.85, 5.89, 3.21, 12.63 g), P (0.17, 0.18, 0.18, 0.40 g), and K (3.54, 3.24, 3.32, 5.28 g), corresponding to ammonium sulfate (22.88, 27.81, 15.13, 59.60 g), superphosphate (0.67, 0.72, 0.71, 1.61 g), and potassium sulfate (7.90, 7.25, 7.43, 11.80 g) respectively. This study provides a scientific basis for blueberry precision fertilization, improving yield/quality while avoiding nutrient waste.

# The amino acid permease SlAAP6 contributes to tomato growth and salt tolerance by mediating branched-chain amino acid transport

Qi Qiang[1], Zhonghui Zhang[2], Xianggui Li[3]

[1] *National Key Laboratory for Tropical Crop Breeding, School of Breeding and Multiplication Sanya Institute of Breeding and Multiplication, Hainan University, Sanya, 572025, China*
[2] *National Key Laboratory for Tropical Crop Breeding, College of Tropical Agriculture 11 and Forestry, Hainan University, Sanya Hainan 572025, China*
[3] *National Key Laboratory for Tropical Crop Breeding, College of Tropical Agriculture 11 and Forestry, Hainan University, Sanya Hainan 572025, China*

E-mail: xianggui.li@hainanu.edu.cn

## Abstract

Branched-chain amino acids (BCAAs) are essential amino acids in tomato (Solanum lycopersicum) required for protein synthesis, which also modulate growth and abiotic stress responses. To date, little is known about their uptake and transport in tomato especially under abiotic stress. Here, the tomato *amino acid permease 6* (*SlAAP6*) gene was identified as an amino acid transporter that restored mutant yeast cell growth on media with a variety of amino acids, including BCAAs. Overexpression of SlAAP6 (*SlAAP6-OE*) in tomato raised the BCAA content and elevated the fresh weight, while *SlAAP6* knockouts (*slaap6*) showed reduced levels of neutral and basic amino acids in seedling tissues and lower total free amino acid distribution to shoots. In comparison to wild type (WT) and slaap6 mutants, *SlAAP6-OE* alleviated root limited growth by elevated BCAA transport and upregulated the expression of root growth-related genes by increasing BCAAs in vivo. As SlAAP6 serves as a positive regulator for BCAA abundance, *SlAAP6-OE* lines showed greater salinity tolerance, while *slaap6* mutants exhibited increased salt sensitivity. The salt tolerance of *SlAAP6-OE* plants was further enhanced by the application of exogenous BCAAs. In addition, BCAA supplementation reduced the accumulation of $H_2O_2$ in root under salt stress conditions. Based on these findings, SlAAP6-mediated uptake and transport of BCAAs facilitated growth and salt tolerance in tomato. By characterizing this key amino acid transporter, this study provides a novel approach to simultaneously enhance tomato nutritional quality, growth and development, and stress resistance through genetic improvement.

# Identification of Genes Involved in Oil Gland Development and Essential Oil Biosynthesis in Citrus

Dongping Pu[1,2,3], Yuan Yu[1,2,3]

[1] *FAFU-UCR Joint Center for Horticultural Plant Biology and Metabolomics*

[2] *Haixia Institute of Science and Technology*

[3] *Fujian Agriculture and Forestry University*

F-mail: 1827989837@qq.com

## Abstract

Oil glands in Citrus (Rutaceae) are secretory cavity structures known for their richness in essential oils. However, the molecular mechanisms underlying the development and expansion of these secretory cavities remain largely unknown. In this study, we observed that citrus cultivars with larger oil gland volumes were more susceptible to sunburn and associated damage. To further investigate, we screened and selected natural mutants of Shatian pomelo with smaller oil cells. By integrating metabolomic and transcriptomic analyses with qRT-PCR validation, we identified key genes associated with oil gland development, expansion, and essential oil biosynthesis. These findings enhance our understanding of oil gland morphogenesis and provide a foundation for improving citrus fruit quality and environmental resilience through molecular breeding.

# A novel strategy for mitigating drought stress in tomato based on the ROS scavenging activity of carbon dots

Feijuan Gao[1], Yuantao Tan[2], Yaoke Duan[3], Hao Sun[3]

[1] *Henan Key Laboratory of Ion-Beam Green Agriculture Bioengineering, School of Agricultural Sciences, Zhengzhou University, Zhengzhou, 450052 China*
[2] *National Key Laboratory of Cotton Bio-breeding and Integrated Utilization, School of Agricultural Sciences, Zhengzhou University, Zhengzhou, 450001 China*

E-mail: gaofeijuan@gs.zzu.edu.cn

## Abstract

Drought stress represents a significant abiotic constraint on global agricultural productivity, adversely affecting plant growth and development, and imposing long-term limitations on crop yield and quality. Conventional approaches such as agronomic management and breeding programs are often resource-intensive and time-consuming, and frequently prove ineffective for rapid drought mitigation during the growing season. Therefore, there is a critical need to develop efficient and practically applicable drought-resistance technologies. In this study, we developed carbon dots (CDs) which can relieve drought stress on tomato (*Solanum lycopersicum* L.) and investigated the mechanism. Using a hydroponic system, tomato seedlings under drought conditions were treated with CDs. The results demonstrated that CDs exhibit not only significant reactive oxygen species (ROS)-scavenging capabilities but also highly efficient root delivery properties. Specifically, CDs were taken up by tomato root cells primarily through active transport-based transmembrane pathways as well as the apoplastic route. Following uptake, CDs effectively reduced the accumulation of superoxide anion ($O_2^{\bullet -}$) and hydrogen peroxide ($H_2O_2$) under drought stress and decreasing membrane lipid peroxidation, as indicated by reduced malondialdehyde (MDA) content. Consequently, CDs preserved cellular membrane integrity, mitigated leaf damage, and ultimately enhanced the drought tolerance of tomato plants. In summary, this study demonstrates that CDs alleviate drought-induced oxidative damage and improve drought resistance through their potent ROS-scavenging activity, providing a theoretical basis for applying carbon-based nanomaterials in agriculture.

# A Novel PbHY5-1-PbBBX1-PbMYB30 Axis and a Functional *PbUFGT9* Promoter SNP Drive Flavonoids Accumulation and Leaf Color Polymorphism in *Phoebe bournei*

Li Wang[a,1], Ningning Fu[a,2], Yuting Zhang[a,b,3], Xiao Han[a,b,4], Qi Yang[a,b,5], Wenjun Ma[c,6], Zaikang Tong[a,b,7], Junhong Zhang[a,b,8]

[a] *State Key Laboratory of Subtropical Silviculture, School of Forestry & Biotechnology, Zhejiang A&F University, Lin'an, Hangzhou 311300, Zhejiang, P. R. China*

[b] *Zhejiang Key Laboratory of Forest Genetics and Breeding, Zhejiang A&F University, Lin'an, Hangzhou 311300, Zhejiang, P. R. China*

[c] *State Key Laboratory of Tree Genetics and Breeding, Key Laboratory of Tree Breeding and Cultivation of State Forestry Administration, Research Institute of Forestry, Chinese Academy of Forestry, National Innovation Alliance of Catalpa Bungei, Beijing, China*

E-mail: 2024202012006@stu.zafu.edu.cn, 20206020411010@stu.zafu.edu.cn, zhangyt@zafu.edu.cn, hanx2017@zafu.edu.cn, qiyang@zafu.edu.cn, zktong@zafu.edu.cn, zhangjunhong@zafu.edu.cn

## Abstract

In the *P. bournei* germplasm collection, newly emerged leaves exhibited three distinct color phenotypes: green, yellow, and red that carry both ecological and ornamental value. With 151 families in the germplasm collection, we selected two individuals with markedly divergent phenotypes from each family, measured their leaf color and growth traits, and conducted whole-genome resequencing. Using a high-quality reference genome, we identified 19,518,953 reliable SNPs. Genome-wide association analysis (GWAS) revealed a key SNP significantly associated with the color parameter $a^*$, which is located in the promoter region of the anthocyanidin glucosyltransferase gene *PbUFGT9* and alters the MYB-binding motif. By combining metabolomic and transcriptomic data and employing weighted gene co-expression network analysis (WGCNA) to uncover the molecular mechanisms of *P. bournei* leaf color formation. In red leaves, PbMYB30 activated *PbPAL1*, *PbCHI1*, and *PbUFGT9* three genes to promote anthocyanin accumulation, while the PbUFGT9 promoter with SNP was weakened transcriptional regulation to reduce its expression and anthocyanin levels. In yellow leaves, PbMYB211 regulated *PbFLS*, diverting flux toward flavonol biosynthesis, thereby generating the yellow phenotype. In addition, PbBBX1 and PbHY5-1 were found to form a transcriptional complex that synergistically activates *PbMYB30*. Notably, $a^*$ values were positively correlated with summer growth, while the distribution of *PbUFGT9* genotypes, linked to the maximum temperature of the hottest month, implied that red-leafed individuals are more likely to exhibit enhanced tolerance to heat and drought. This multi-omics study reveals the key regulatory network underlying leaf color variation in *P. bournei* and proposes the potential of leaf color as a visual marker for identifying drought- and heat-tolerant superior genotypes, providing new theoretical and practical foundations for the breeding of improved *P. bournei* varieties.

# Analysis of lncRNA-mRNA Pairs Induced by *Colletotrichum camelliae* and Novel Regulator *Cslnc170* for *CsLOX4* in Tea Plants

Ting Jiang[12], Cheng Liu[1], Xiaolan Jiang[1], Liping Gao[3*], Tao Xia[1*]

[1] *National Key Laboratory for Tea Plant Germplasm Innovation and Resource Utilization, West 130 Changjiang Road, Hefei 230036 Anhui, China*[1]

[2] *Moith Agricultural Tech Co., Ltd., Chizhou 242899, PR China*

[3] *School of Life Science, Anhui Agricultural University, West 130 Changjiang Road, Hefei 230036 Anhui, China*

E-mail: tjiang0919@163.com

## Abstract

In tea-growing regions, fungal diseases like anthracnose significantly affect tea plant growth. Understanding disease resistance mechanisms and identifying resistance genes can aid in breeding resistant varieties. Non-coding RNAs, including long non-coding RNAs (lncRNAs), play critical roles in regulating plant immunity by influencing target gene expression. Yet, their role in tea plant disease resistance remains underexplored. This study used RNA sequencing to identify differentially expressed lncRNAs and mRNAs in Camellia sinensis following infection with Colletotrichum camelliae. Analysis revealed 524 antisense lncRNA-mRNA pairs and 3,588 cis-acting lncRNA-mRNA pairs, involved in photosynthesis, amino acid biosynthesis, fatty acid metabolism, and secondary metabolism pathways such as flavonoid biosynthesis. Among these, the cis-acting pair Cslnc170-CsLOX4 was identified as a key regulator of disease resistance. The Cslnc170 gene (1,581 bp) lies 9,254 bp downstream of the CsLOX4 gene, a member of the 13-lipoxygenase family. Functional studies showed that Cslnc170 activates CsLOX4 expression via its secondary structure's loop 4 and the CsLOX4 promoter region (930–952 bp). Agrobacterium-mediated overexpression and antisense oligonucleotide-mediated silencing experiments confirmed that the Cslnc170-CsLOX4 pair enhances resistance to anthracnose in tea leaves. These findings provide new insights into the regulatory role of lncRNA-mRNA pairs, offering potential targets for improving disease resistance in tea plants.

# Gap-free genome and efficient transcript purification system reveals the genomes diversity and chlorophyll degradation mechanism in pitaya

Jiaxuan Chen[1+], Fangping Li[1,2*+], Jieying Liu[2], Yuchen Mao[1], Zhenpeng Gan[2], Haifei Hu[2], Irfan Ali Sabir[1], Imran Khan[1], Jiayi Chen[1], Canbin Chen[1], Zhike Zhang[1], Jietang Zhao[1], Guibing Hu[1], Shaokui Wang[2], Yonghua Qin[1*]

*[1] Guangdong Provincial Key Laboratory of Postharvest Science of Fruits and Vegetables and Key Laboratory of Biology and Genetic Improvement of Horticultural Crops (South China), Ministry of Agriculture and Rural Affairs, College of Horticulture, South China Agricultural University, Guangzhou, 510642, China*

*[2] Guangdong Provincial Key Laboratory of Plant Molecular Breeding, State Key Laboratory for Conservation and Utilization of Subtropical Agro-Bioresources, South China Agricultural University, Guangzhou 510642, China.*

E-mail: qinyh@scau.edu.cn; fangping.li@scau.edu.cn

## Abstract

Pitaya is an important perennial herbaceous fruit tree. The color of fruit determines pitaya nutritive (and attractive) value, which is considered as an important objective in breeding improvement. In this study, we reported the first telomere-to-telomere (T2T) gap-free genome of 'Shuangse No. 1' pitaya (Hylocereus polyrhizus; red peel). Two high-quality genomes for 'Dahong' (H. polyrhizus; red peel) and 'Honghuaqinglong' (H. stenopterus; stay-green) were further assembled aiming to explore the genetic diversity of pitaya genomes. In further analysis, we noticed a high proportion of viral contamination in pitaya tissues, which hindered the efficient utilization of transcriptomic data. To address this issue, we analyzed 111 pitaya transcriptome data from different geographic regions to characterize and separate viral components. Then we developed an efficient, novel, and universal transcript purification system for pitaya transcriptomes by applying it to 27 samples from different tissues and species, thereby enhancing the utility for transcriptomic and broader biological research. Combining the purified transcriptomic data with comparative genomic analyses, we identified HuERF72, a transcription factor that potentially regulates chlorophyll degradation in pitaya. Interaction assays and plant transformation elucidated that HuERF72 acts as a repressive transcription factor by directly binding to the promoter of HuSGR1, a key structural gene in the chlorophyll degradation pathway. This study provides high-quality genomic resources and novel methodologies for molecular investigations in pitaya. Additionally, the proposed regulatory network advances our understanding of the transcriptional regulatory mechanisms underlying chlorophyll degradation, offering valuable insights into the genetic improvement of pitaya.

# Development and Application of Genetic Variation Genotyping Algorithms Based on Graph Pan-Genomes

Ze-Zhen Du[1,2,3], Jia-Bao He[1,2,3], Pei-Xuan Xiao[1,2,3], Jianbing Hu[1,2], Ning Yang[2,4], Wen-Biao Jiao[1,2,3*]

[1] *National Key Laboratory for Germplasm Innovation and Utilization of Horticultural Crops, Huazhong Agricultural University, Wuhan, China*

[2] *Hubei Hongshan Laboratory, Wuhan, China.*

[3] *Hubei Key Laboratory of Agricultural Bioinformatics, College of Informatics, Huazhong Agricultural University, Wuhan, China*

[4] *National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China.*

E-mail: jiao@mail.hzau.edu.cn

## Abstract

Accurate variant genotyping is crucial for genomics-assisted breeding. Graph pangenome references can address single-reference bias, thereby enhancing the performance of variant genotyping and empowering downstream applications in population genetics and quantitative genetics. However, existing pangenome-based genotyping methods struggle with large or complex pangenome graphs, particularly in polyploid genomes. Here, we introduce Varigraph, an algorithm that leverages the comparison of unique and repetitive k-mers between variant sites and short reads for genotyping both small and large variants. We evaluated Varigraph on a di-verse set of representative plant genomes as well as human genomes. Varigraph outperforms current state-of-the-art linear and graph-based genotypers across non-human genomes while maintaining comparable genotyping performance in human genomes. By employing efficient data structures including Counting Bloom Filter and bitmap storage, as well as GPU models, Varigraph achieves improved precision and robustness in repetitive regions while managing computational costs for large datasets. Its wide applicability extends to highly repetitive or large genomes, such as those of maize and wheat. Significantly, Varigraph can handle extensive pangenome graphs, as demonstrated by its performance on a dataset containing 252 rice genomes, where it achieved a precision exceeding 0.9 for both small and large variants. Notably, Varigraph is capable of effectively utilizing pangenome graphs for genotyping autopolyploids, enabling precise determination of allele dosage. This work provides a robust and accurate solution for genotyping plant genomes and will advance plant genomic studies and ge-nomics-assisted breeding.

# Natural variation in the ClPrx53 promoter regulates plant height in watermelon under waterlogging

Xiaoxiao Guan, Ye Wang, Zhongyuan Hu

*College of Agriculture and Biotechnology, Zhejiang University, Hangzhou 310058, China*

E-mail: 22016161@zju.edu.cn

## Abstract

Waterlogging and other abiotic stresses adversely affect the growth and development of watermelon (*Citrullus lanatus*). In this study, a genome-wide association study (GWAS) was conducted on 178 watermelon accessions with extensive natural variation, leading to the identification of *Clprx53*, a novel quantitative trait locus encoding peroxidase. A 3-bp InDel in the promoter region of *Clprx53* was found to be responsible for differential responses to waterlogging. Accessions with the 3-bp insertion exhibited greater plant height and enhanced waterlogging tolerance compared to those without the insertion. Transgenic watermelon and tobacco plants overexpressing *ClPrx53* showed increased plant height and improved waterlogging tolerance under flooded conditions. Furthermore, virus-induced gene silencing (VIGS) analysis confirmed that *ClPrx53* positively regulates plant height under waterlogging stress. GUS (β-glucuronidase) staining assays demonstrated its responsiveness to flooding. Both in vivo and in vitro assays revealed that CLGAI-LIKE, a DELLA protein, specifically binds to the inserted promoter region of *Clprx53*, thereby enhancing its expression. These findings provide insights into the molecular mechanism underlying waterlogging tolerance and offer potential strategies for improving stress resilience in watermelon.

# Identification and characteristic analysis of PavUGT48 as a novel UDP-glycosyltransferase with dual functions on anthocyanin and amygdalin biosynthesis in sweet cherry

Chen Feng[1,2,3], Qingqing Guo[1,2,3], Chuanbao Wu[1,2,3], Jing Wang[1,2,3], Xiaoming Zhang[1,2,3], Guohua Yan[1,2,3], Yu Zhou[1,2,3], Wei Wang[1,2,3], Zheyong Xue[4,*], Kaichun Zhang[1,2,3,*], Xuwei Duan[1,2,3,*]

[1] *Institute of Forestry and Pomology, Beijing Academy of Agriculture and Forestry Sciences, Beijing 100093, China*

[2] *Key Laboratory of Biology and Genetic Improvement of Horticultural Crops (North China), Ministry of Agriculture and Rural Affairs, Beijing 100093, China*

[3] *Beijing Engineering Research Center for Deciduous Fruit Trees, Beijing 100093, China*

[4] *Heilongjiang Key Laboratory of Plant Bioactive Substance Biosynthesis and Utilization, Northeast Forestry University, Harbin 150040, China*

E-mail: fengclgs@163.com

## Abstract

UDP-glycosyltransferase (UGT) is essential for fruit development, but its specific role in sweet cherry fruits has not been clearly defined. This study identified a new differentially expressed UGT gene, FUN_010048, by analyzing transcriptomic data from various colored regions of bicolored fruits. The 187 UGT family members were identified in the sweet cherry genome of 'Tieton'. Notably, FUN_010084 has been renamed PavUGT48. Expression analysis revealed that PavUGT48 increased during the development of the differently colored sweet cherry fruits. Co-expression network analysis and further experiments confirmed that PavUGT48 generates cyanidin-3-O-glucoside from cyanidin using UDP-glucose (UDP-Glc) as the sugar donor, enhancing fruit coloration. Additionally, PavUGT48 facilitates the formation of amygdalin from prunasin, but it does not react with naringenin, dihydrokaempferol, or kaempferol when UDP-Glc serves as the sugar donor. These findings enhance our understanding of UGT enzymes in sweet cherries and provide a basis for improving fruit quality and developing new varieties.

# Spring cold stress at high altitudes in southeastern Xizang activates *CsABF2* to regulate chlorophyll degradation and phenolic biosynthesis in tea plants

Yipeng Huang[1, *], Didi Jin[1, *], Tianming Jiao[1], Zhenhong Wang[3], Ting Jiang[1], Lei Zhao[4], Xiaolan Jiang[1], Haiyan Wang[2], Yajun Liu[2], Yunsheng Wang[2, #], Liping Gao[2, #], Tao Xia[2, #]

[1] *State Key Laboratory of Tea Plant Germplasm Innovation and Resource Utilization / Key Laboratory of Tea Biology and Tea Processing of Ministry of Agriculture / Anhui Provincial Laboratory of Tea Plant Biology and Utilization, Anhui Agricultural University, West 130 Changjiang Road, Hefei 230036 Anhui, China*

[2] *School of Life Science, Anhui Agricultural University, 130 Changjiang West Road, Hefei 230036 Anhui, China*

[3] *College of Resources and Environment, Xizang Agricultural and Animal Husbandry University, 100 Yucai West Road, Linzhi 860000 Xizang, China*

[4] *College of Horticulture, Qingdao Agricultural University, 700 Changcheng Road, Qingdao, 266109 Shandong, China*

E-mail: Huangyp9703@163.com

## Abstract

The tea plant (*Camellia sinensis*), native to warm and humid low-latitude regions of southwestern China, has expanded to higher altitudes, including southeastern Xizang, where cultivation above 2,500 meters poses challenges due to low accumulated temperatures. However, the impact of high-altitude climatic conditions, particularly temperature, on tea growth remains underexplored. To investigate, weather stations were deployed at three altitudes in southeastern Xizang: Medog (MD, 1,200 m), Zayü (ZY, 1,720 m), and Layue in Bayi District (BY, 2,600 m) to monitor spring temperature fluctuations. Field observations and meteorological data indicated that the milder spring temperatures in MD and ZY facilitated normal budburst and growth, whereas the lower temperatures in BY delayed budburst and resulted in leaf yellowing and browning. Comparative experiments revealed that seedlings exposed to fluctuating low temperatures (10 °C / 4 °C) experienced the most severe cold injury and exhibited the lowest germination rates compared to seedlings under constant-temperature treatments. Transcriptome analysis uncovered differential expression of genes involved in chlorophyll degradation, lignin biosynthesis, and flavonoid pathways under cold stress. Functional characterization of the cold-induced transcription factor *CsABF2* revealed its central role in activating these pathways, as evidenced by antisense oligodeoxynucleotide (AsODN) silencing and promoter activation assays, to activate key downstream genes: *CsSGR1* (chlorophyll degradation), *CsPALa* (phenylpropanoid pathway), and *CsMYB6c* (flavonoid biosynthesis). These results provide mechanistic insights into how spring temperature variability at high altitudes impairs tea plant development and alters quality-related metabolites, offering a molecular basis for improving cold resilience in tea cultivation.

# A SlMYB78‐regulated bifunctional gene cluster for phenolamide and salicylic acid biosynthesis during tomato domestication, reducing disease resistance

Peng Cao[1,2,3], Jun Yang[1,2*], Shouchuang Wang[1,2,3,*]

[1] *School of Breeding and Multiplication (Sanya Institute of Breeding and Multiplication), Hainan University, Sanya 572025, China;*

[2] *College of Tropical Agriculture and Forestry, Hainan University, Haikou 572208, China;*

[3] *Collaborative Innovation Center of Nanfan and High-Efficiency Tropical Agriculture, Hainan University, Haikou 572208, China;*

E-mail: yang9yj@hainanu.edu.cn, shouchuang.wang@hainanu.edu.cn

## Abstract

Plants have evolved complex chemical defense networks to combat pathogens, in which phenolamides and salicylic acid (SA) play key roles in immune responses. However, the mechanisms of their synergistic regulation in biosynthesis remain to be explored. Here, we identified a biosynthetic gene cluster (BGC2) on chromosome 2 that is associated with the biosynthesis of phenolamides and SA, where the key component SlEPS1 has dual catalytic functions for the synthesis of both phenolamides and SA. Overexpression of the key BGC2 component *SlEPS1* in tomato enhanced resistance to the bacterial pathogen *Pst DC3000*, while knockout plants were more susceptible to the disease. Exogenous application of SA and phenolamides revealed that these two compounds enhance plant resistance through synergistic action. Notably, during tomato domestication, a disease-resistant allele of *SlEPS1*, *SlEPS1*$^{HapB}$, was negatively selected, leading to reduced levels of phenolamides and SA in modern varieties and compromised disease resistance. Additionally, SlMYB78 directly regulates the BGC2 gene cluster, enhancing the biosynthesis of phenolamides and SA, and modulating resistance to *Pst DC3000*. Our study, using multi-omics approaches, elucidates the synergistic regulation of phenolamide and SA biosynthesis, providing new insights into the complexity of plant immunity-related metabolism.

# Expression and Functional Analysis of the ABORTED MICROSPORES (*AMS*) Gene in Marigold (*Tagetes erecta* L.)

Xuejing Ma[1,2], Jinhua Tian[1,2], Daocheng Tang[1,2], Qiuyue Liang[1,2] and Nan Tang[1,2,*]

[1] *Plateau Flower Research Centre, Qinghai University, Xining 810016, China;*
[2] *Qinghai Key Laboratory of Garden Plants and Ornamental Horticulture , Xining 810016, China;*

Presenting author(s) e-mail:

## Abstract

Marigold (*Tagetes erecta* L.), an annual herbaceous plant of the genus *Tagetes* L. in the Asteraceae family, which has large, vibrantly colored flowers, superior stress resistance, long flowering period, and extensive cultivar diversity. These attributes make it widely applicable to ornamental landscaping projects. In marigold crossbreeding, emasculation is very difficult to carry out because of its capitulum. Male-sterile dual-purpose lines are usually used as parents to breed heterosis varieties. Therefore, male sterility is an important trait in heterosis utilization and marigold breeding. ABORTED MICROSPORES (*AMS*) is an important gene that regulates tapetum and microspore development. Therefore, the effect of *AMS* on fertility was studied. *TeAMS* was located in the nucleus and exhibited self-activation activity. *TeAMS* was highly expressed in the flower buds of *T. erecta*. The expression of this gene in fertile plants was higher than that in sterile plants, and the expression level gradually increased with the development of flower buds. The expression trend of *TeAMS* in sterile plants was opposite to that in fertile plants. In tobacco (*Nicotiana tabacum* L.), overexpression of the *TeAMS* gene resulted in shortened floral tubes, increased thousand-seed weight, a reduced flowering period, and decreased flower numbers. The pollen viability of transgenic tobacco was significantly lower than that of the wild type, and the pollen grains were smaller and showed irregular shapes. The pollen wall was dry and shrunk. Some pollen germinal furrows were distorted, and a few were almost invisible. Silencing *TeAMS* resulted in a longer flowering period in tobacco, reduced thousand-seed weight, and high pollen viability. Pollen morphology in silenced lines showed no significant differences compared to the wild-type and empty vector controls. Only a few pollen grains were smaller, shriveled, and shrunken. Therefore, the *TeAMS* gene plays an important role in regulating the fertility of marigolds. This study provided a theoretical support for breeding of marigold male-sterile lines.

**Keywords:** marigold (*Tagetes erecta* L.); male sterility; *TeAMS*; transcription factor; functional verification

# Genome-wide association of terpenoid metabolites reveals candidate gene for carrot flavor

Jie Wu[1], Xiaoyu Qi[1], Xiaoping Kong[1,2], Hao Sun[3]

[1] *College of Agriculture and Animal Husbandry, Qinghai University, Xining 810016, China*

[2] *Xining Vegetable Technical Service Center, Xining 810003, China*

[3] *School of Agricultural Sciences, Zhengzhou University, Zhengzhou 450052, China*

E-mail: 360946095@qq.com

## Abstract

Carrots, renowned for their diverse range, stand as a staple among vegetables, yet distinct taste profiles characterize various cultivars. The fleshy roots of carrots boast a wealth of terpenoids, known for their propensity to engender undesirable flavors like bitterness and medicinal notes. This study is a 2-year investigation involving a 4-point survey of flavor traits across 240 carrot varieties. Resequencing was carried out on these varieties planted in Xingquan Village, Huangyuan County in June 2022. Subsequently, an extensive targeted metabolomics analysis was conducted on the mature fleshy roots of 162 selected carrot varieties to identify terpenoid differential metabolites. Leveraging a genome-wide association study based on metabolomics (mGWAS), we conducted a correlation analysis to elucidate the interplay between terpenoid metabolite variations and genomic diversity across carrot cultivars, pinpointing key genes implicated in the biosynthesis of target terpenoids. Metabolomic analysis was conducted on the roots of 162 representative carrot varieties at maturity, identifying two distinct terpene metabolites: 11-O- β -D-glucoside 3-hydroxy-7, 8-dihydro-beta-ionol and dehydrodigitoxin. Leveraging mGWAS analysis, associations between terpene metabolite variations and genomic variances across different carrot varieties were explored. The analysis identified 75 significant SNPs linked to metabolic phenotypes, representing 108 candidate genes, with 37 genes being annotated. Notably, four candidate genes implicated in flavor traits were identified through functional annotations: *Cris1*, *Germ1*, *Cis-A1* and *Dehy1*. Subsequent RT-PCR analysis demonstrated that the expression levels of *Cris1*, *Germ1*, and *Dehy1* were significantly higher in sweet varieties compared to bitter ones, while *Cis-A1* exhibited the opposite trend, with higher expression in bitter varieties than in sweet ones.

# Characteristics of two new marigold （*Tagetes erecta* L.） varieties and construction of DNA fingerprint

Sirangsuo Li[1,2]， Xuejing Ma[1,2], Junwei Guo[1,2], Nan Tang[1,2,*]

[1] *Plateau Flower Research Centre, Qinghai University, Xining 810016, China;*
[2] *Qinghai Key Laboratory of Garden Plants and Ornamental Horticulture , Xining 810016, China;*

Presenting author(s) e-mail:

## Abstract

Marigold (*Tagetes erecta* L.) is an ornamental plant that is widely cultivated all over the world, which has high ornamental and economic value. Determination of the distinctness, uniformity and stability (DUS) of plant cultivars is an important means of protecting the original innovations of breeders, increasing the enthusiasm of breeders and promoting the development of plant breeding. To elucidate the genetic structure and diversity of Marigold varieties, we analyzed 12 marigold varieties (lines) by using 60 pairs of SSR markers and 22 DUS morphological features. In the DUS testing, significant positive or negative correlations among 22 DUS traits were observed. When the Euclidean distance is 10, the 12 marigold varieties (lines) are divided into 4 groups. Ten pairs of SSR markers were screened out from 60 pair of SSR markers. The average polymorphism index (PIC) of the SSR markers was 0.346, which could be used as key SSR markers for genetic diversity analysis and fingerprint map construction of marigold. Genetic diversity analysis of the 12 test materials showed that the average Shannon's information index (I) was 0.651. Cluster analysis revealed that there were correlations between the SSR markers and morphological characteristics, and there were also some diferences. Based on SSR markers, the fingerprint spectra of two new marigold varieties and their control varieties (lines) were successfully constructed. It provides important technical support for the selection and identification of core germplasm of marigold and the breeding of new varieties, which could lay a foundation for subsequent production applications as well as breeding work of new varieties.

**Keywords:** marigold (*Tagetes erecta* L.); DUS descriptors; simple sequence repeats; markers fingerprints; genetic diversity

# Poplar *RBOHF* Mediate Tension Wood Formation

Dong Ning[1], MengZhu Lu[1]

[1]*National Key Laboratory for Development and Utilization of Forest Food Sources, College of Forestry and Biotechnology, Zhejiang A&F University,Hangzhou,China*

E-mail: 619843803@qq.com

## Abstract

Tension wood (TW) formation in angiosperm trees represents a fundamental adaptive response to gravitational stress, playing a crucial role in providing mechanical support and influencing wood quality. During the early stages of TW development, the superoxide anion ($O_2^{\cdot-}$) speciallyand these bursts accumulates and functions as a key signaling molecule that triggers cambial cell proliferation, differentiation, and ultimately, TW formation. However, the specific members of the RBOH family responsible for $O_2^{\cdot-}$generation and their functional relationship with TW formation remain uncharacterized. In this study, we demonstrate that *RBOHF* is involved in this process. Overexpression of *RBOHF* enhances TW formation and significantly increases the thickness of G-layer in TW fibers. Conversely, *RBOHF* knockout mutants exhibit reduced $O_2^{\cdot-}$accumulation and a marked decrease in G-layer thickness. These results provide the genetic evidence supporting that RBOHF regulates $O_2^{\cdot-}$ levels during TW formation.

# The molecular mechanism of PpBBX24 mutation promoting ‘Red Zaosu’ peel coloring

Liuxiao Feng[1,2]

[1]*China Agricultural University*
[2]*Institute Of pomology of CAAS*

E-mail: Liuxiaofeng01@caas.cn

## Abstract

In this study, we systematically elucidated the key molecular mechanisms underlying red peel formation in ‘Red Zaosu’. These findings confirmed that light induced the coloration of ‘Red Zaosu’, which was closely linked to anthocyanin accumulation and the upregulated expression of related genes (*PpCHS*, *PpCHI*, *PpMYB10*, and *PpHY5*). The study focused on the previously identified 14 bp deletion mutant gene Ppbbx24-del. It was found that the mutant protein could promote anthocyanin synthesis and enhance coloring ability by activating the expression of structural genes and regulatory factors. In contrast, the original protein PpBBX24 did not have this function. Subcellular localization analysis showed that the mutation led to the protein being distributed in both the nucleus and the cytoplasm, while the wild-type protein was exclusively localized in the nucleus. Further mechanistic investigations demonstrated that both Ppbbx24-del and PpHY5 could bind to the G-box elements within the promoters of downstream genes and activate transcription. In contrast, the function of PpHY5 was suppressed by PpBBX24. Furthermore, the study revealed that PpBBX24 interacts with the E3 ubiquitin ligase PpPUB59 via its VP domain, leading to its degradation. Owing to the lack of the VP domain, the mutant Ppbbx24-del bypassed the degradation pathway, resulting in a stable promotion of anthocyanin synthesis. This study uncovered the molecular basis underlying the bud mutation responsible for red-skin coloring in pears, offering a crucial theoretical foundation for enhancing pear fruit quality.

**Multi-omics comparative analzes of Chinese and Worldwide oil-bearing roses**

Yaping Kou, Ruidong Jia, Xin Zhao, Huayang Li, Hong Ge*, Shuhua Yang*

*State Key Laboratory of Vegetable Biobreeding, Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences, Beijing 100081, China*

E-mail: kouyaping@caas.cn

## Abstract

Rose essential oils are obtained from rose petal, it has been used to treat health conditions and used in natural beauty treatments for thousands of years. Flower fragrant volatile organic compounds (VOCs) are main compounds for essential oils and it determined the commercial value of the rose. Nowadays, *Rosa* × *damascena, R. centifolia, R. hybrids* 'Morocco', *R. hybrids* 'de Grasse' are used to extract essential oils in worldwide, while *R. rugosa* 'Feng Hua', *R. rugosa* f. *plena*, *R. hybrids* 'Ku Shui' are used in Chinese. Here, we firstly invest floral VOCs profiles of 7 oil-bearing rose mentioned in above context by HS-SPME-GC-MS. Then, the different development stages of flower petals are used to extract essential oils and pigment, carry out RNA-sequence, untargeted-metabolome. The chemical compounds of essential oils, anthocyanins , and flavonols are comparative analysis between *Rosa* × *damascena and R. rugosa* 'Feng Hua'. Chemical profiles of essential oils analyze shows that the content of methyleugenol, geraniol, neroli, geranyl acetate, phenethyl acetate, nonadecane, heneicosane are significantly different. Seven anthocyanins and 19 flavonols glycoside are identified in total, the content of anthocyanin are significantly lower in *R.* × *damascena*, while flavonols are higher compaired with *R. rugosa* 'Feng Hua'. Wide-targeted metabolomes and RNA-sequence comparative analysis indicate that DXRs play key roles on carbon distribution and biosynthesis of terpenoids skeleton and caused different in content of terpenoids VOCs in two roses. This study provide more reference for rose molecular breeding.

# The glutaredoxin (CitGRX9)-mediated decrease of CitCAT1 activity negatively regulates Huanglongbing tolerance via reconstructs $H_2O_2$ homeostasis in citrus

Xiangyang Li[1,2,3], Minyi Chen[1], Yunfang Lu[1], Yun Zhong[1,2,3*]

[1] *Institute of Fruit Tree Research, Guangdong Academy of Agricultural Sciences*

[2] *Key Laboratory of South Subtropical Fruit Biology and Genetic Resource Utilization, Ministry of Agriculture and Rural Affairs*

[3] *Guangdong Provincial Key Laboratory of Science and Technology Research on Fruit Tree, Guangzhou, 510640, China*

E-mail: lixiangyang@gdaas.cn; zhongyun@gdaas.cn

## Abstract

Citrus huanglongbing (HLB), primarily caused by the unculturable phloem-limited bacterium *Candidatus* Liberibacter asiaticus (CLas), is one of the most disastrous citrus diseases worldwide. It is current accepted that reactive oxygen species (ROS, mainly $H_2O_2$) play a key role in modulating host resistance/tolerance to pathogen attack. However, the special roles of ROS in HLB pathogenesis and the underlying molecular mechanisms for its homeostatic regulation remain poorly defined. Here, by employing the HLB-susceptible variety Succari orange (*Citrus sinennsis* Osbeck cv. Succari), our results showed that in contrast to the control leaves, the HLB-positive but asymptomatic leaves accumulated higher level of $H_2O_2$, primarily in the leaf veins and petioles, and was accompanied with slightly enhanced cell death. Subsequently, transcriptome analysis and further qRT-PCR verification identified a $H_2O_2$-scavenging catalase (CitCAT1) that is negatively correlated with the progression of HLB in the asymptomatic leaves. Furthermore, *in planta* transformation evidenced that overexpression of CitCAT1 can markedly reduce $H_2O_2$ level in the leaves, alleviate HLB symptoms (including cell death), while enhance the onset of Citrus bacterial canker (CBC). Mechanistically, a cytoplasm-localized glutaredoxin CitGRX9, which is significantly upregulated in response to HLB infection based on the transcriptome analysis, was found to interact with peroxisome-targeted CitCAT1 in peroxisome. And their physiological interaction significantly compromised CitCAT1 activity *in vivo*, thereby regulating $H_2O_2$ homeostasis and the $H_2O_2$-triggered cell death process, which may ultimately participate in the modulation of HLB progression. Collectively, this research suggest how CitGRX9-CitCAT1 interaction modulates $H_2O_2$ homeostasis to function in HLB progression.

# Exogenous 6-BA Induces Female Flower Differentiation in Tung Tree (*Vernicia fordii*)

Lin Zhang[1,2,3], Meilan Liu[1,2,3], Chengbo Yang[1,2,3], Junjie Chen[1,2,3], Jie Cao[1,2,3]

[1] *State Key Laboratory of Woody Oil Resources Utilization, Central South University of Forestry and Technology, Changsha, China*

[2] *Yuelushan Laboratory Non-wood Forests Variety Innovation Center, Central South University of Forestry and Technology, Changsha, China*

[3] *Key Laboratory of Cultivation and Protection for Non-Wood Forest Trees, Ministry of Education, Central South University of Forestry and Technology, Changsha, China*

E-mail: triwoodtim918@126.com

## Abstract

Tung tree (*Vernicia fordii*) is an economically important woody oilseed species with unisexual flowers. Understanding the mechanisms underlying sex determination is crucial for improving fruit yield. We have previously shown that the 6-Benzylaminopurine (6-BA) can induce male-to-female sex conversion in inflorescence buds of tung tree, but the underlying molecular mechanisms remain elusive. Here, we found Cytokinin-Independent 1 (*CKI1)* likely play significant roles in this process. OE-*VfCKI1* also positively promotes the development of female organs. VfCKI1 and VfHP4 interact with Constitutive Photomorphogenesis 9 (*VfCOP9*), respectively. However, OE-*VfCKI1–VfCOP9* largely rescues the stigma development phenotype of OE-*VfCKI1* which implies a competitive relationship between VfHP4 and VfCKI1 proteins. Ubiquitination assays demonstrated that VfCOP9 preferentially interacts with VfHP4 and inhibits its activity, thereby reducing the ubiquitination-mediated degradation of VfCKI1. This stabilization of VfCKI1 protein levels positively regulates the development of female organs. In addition, overexpression of Organ Boundaries Domain 41 (*VfLBD41*) also results in stigma exsertion. The VfLBD41 interacts with Crabs Claw (VfCRC), and *VfCRC* can bind the Histidine phosphotransfer protein 4 (VfHP4) promoter (5'-TCNTCNTYNT-3') to enhance *VfHP4* expression. And the 6-BA signaling can significantly promote *VfLBD41* expression through an ARR motif in its promoter. Collectively, our findings suggest that the application of 6-BA to male inflorescence buds upregulates *VfLBD41* expression, which subsequently activates the VfLBD41-VfCRC signaling module to enhance *VfHP4* expression. This leads to a reconfiguration of ubiquitination dynamics within the VfCKI1-VfCOP9-VfHP4 complex, resulting in the stabilization of VfCKI1 protein and ultimately promoting female floral development.

# Molecular mechanisms underlying cool temperature-induced reduction of fruit quality in *Siraitia grosvenorii* via ethylene/ABA-mediated suppression of glycosylation

Jingjing Yang[1], Huan Peng[2], Dong Xiao[2], Jingjian Li[1], Changming Mo[2]*， Wenjuan Xie[1*]

[1] *Guilin Medical University*

[2] *Guangxi University*

E-mail: uuu171722@yeah.net

## Abstract

This study systematically compared the climatic factors, fruit development, Mogroside content, endogenous hormone dynamics, and gene expression of main varieties from summer and autumn pollination using a major cultivated variety to elucidate the effects of climatic differences between summer and autumn on *Siraitia grosvenorii* fruits quality and the underlying molecular regulatory mechanisms. The results indicated that: (1) The mean temperature and effective cumulative temperature during the late development stage of autumn-pollinated fruits were significantly lower; (2) Fruit maturation in autumn was delayed by 10-30 days, the glycosylation process of Mogroside III, Mogroside IVA, Mogroside SI to Mogroside V and 11-O-mogroside V was postponed by approximately 10 days, and the contents of Mogroside V and 11-O-mogroside V at maturity decreased by 40.66% and 46.07%, respectively; (3) During the rapid accumulation phase of Mogroside V (55-75 days after pollination), the contents of ACC and ABA in autumn fruits were significantly lower than those in summer; (4) The glycosyltransferase genes *SgUGT85-269-1/2/4* and *SgUGT94-289-1/2/3* were rapidly and consistently upregulated in summer fruits but significantly delayed in autumn, with the key gene *SgUGT94-289-3* showing a downregulated trend; (5) The promoter regions of transcription factors such as *AP2/ERF*, *bHLH*, and *MYB*, which were co-expressed with *SgUGT85-269-1*, *SgUGT94-289-1/3*, *SgUGT85-269-1*, and *SgUGT94-289-1/2*, contained binding elements for *AP2/ERF*, *bHLH*, *bZIP*, and *MYB*. In conclusion, habitat cooling inhibits glycosyltransferase gene expression, particularly *SgUGT94-289-3*, by regulating ethylene/ABA signaling and transcription factor networks, resulting in a significant reduction in Mogroside quality in autumn-pollinated fruits. This study provides a theoretical basis for regulating *Siraitia grosvenorii* cultivation quality.

# Processing-Shaped Bioactive Profile in *Illicium verum* 'Thirteen-horn' Revealed by Metabolomics–Network Pharmacology: A Roadmap for Breeding High-Value Cultivars

Yunying Ma[1,2], Lu´an Xu[1].Wenhai Li[1], Rongjun Fan[1], Bo Zhao[2*]

[1] *Guangxi Key Laboratory of Drug Discovery and Optimization, School of Pharmacy, Guilin Medical University, Guilin 541199, China*
[2] *College of Pharmacy, Guilin Medical University, Guilin 541199, China*

E-mail: 2601284677@qq.com; 122017017@glmc.edu.cn

## Abstract

*Illicium verum* 'Thirteen-horn' is a high-quality star anise cultivar bred in China and has been widely promoted in Guangxi Province as a crop with both culinary and medicinal value. Illicium species are rich in essential oils, terpenoids, and flavonoids, which possess diverse bioactivities and have important applications in the food industry, economic development, and pharmacological research. However, standardized postharvest processing and chemical treatment protocols for 'Thirteen-horn' have not yet been established, which limits the full exploitation of its potential.

In this study, we integrated untargeted metabolomics, network pharmacology, and in vitro cell experiments to systematically evaluate the effects of three drying methods-freeze-drying (FD), oven-drying (OD), and sun-drying (SD)-on the bioactive anti-inflammatory constituents and their underlying mechanisms in 'Thirteen-horn'. Metabolomics analysis revealed that essential oils, terpenoids, and flavonoids were the predominant metabolite classes. Among the three drying methods, FD preserved the highest levels of bioactive compounds, whereas SD caused a pronounced loss. Consistently, in vitro anti-inflammatory assays demonstrated that FD samples exhibited the strongest anti-inflammatory activity.

Integrated network pharmacology analysis further elucidated the molecular mechanisms of the key bioactive compounds: anethole inhibited inflammatory responses by downregulating tumor necrosis factor-α (TNF-α) expression; limonene and α-pinene targeted the nuclear factor-κB (NF-κB) signaling pathway to suppress inflammatory signal transduction; quercetin exerted dual anti-inflammatory and antioxidant effects by scavenging reactive oxygen species (ROS) and downregulating cyclooxygenase-2 (COX-2) and inducible nitric oxide synthase (iNOS), thereby reducing nitric oxide (NO) production. Notably, kaempferol, a key anti-inflammatory flavonoid, mitigated inflammation and oxidative stress-induced tissue injury by inhibiting the mitogen-activated protein kinase (MAPK) signaling pathway.

Collectively, this study established a processing–metabolite–bioactivity association model, identified key anti-inflammatory constituents and molecular targets of 'Thirteen-horn', and provides a theoretical basis and molecular markers for optimizing standardized processing, developing functional products, and guiding future breeding programs of high-value star anise cultivars.

**Polyclonal antibody prepared with selected peptides provides high specificity in serological detection of tobamovirus**

Meisheng Zhao[1], Shuai Xu[1], Dejie Cheng[3], Xiao Yin[1], Shanshan Jiang[1], Yanping Tian[2], Chao Geng[2], Zhiyong Yan[2], Xingbo Lu[1], Xiangdong Li[1, 2]

[1] *Institute of Plant Protection, Shandong Academy of Agricultural Sciences, Jinan, 250100, China*

[2] *College of Plant Protection, Shandong Agricultural University, Tai'an, 271018, China*

[3] *Agricultural College, Guangxi University, Nanning, 520004, China*

E-mail: meisheng@saas.ac.cn

## Abstract

Tobamoviruses pose severe threats to crops worldwide. Early diagnosis is crucial for the control of tobamoviruses and antibody-based serological detection is the common method. However, polyclonal antibody (pAb) has a risk of the cross-reaction with similar tobamoviruses, and monoclonal antibody (mAb) has a complex production process, though it can effectively distinguish tobamovirus. Our study synthesized peptide antigens derived from C-terminus of coat protein (CP) of cucumber green mottle mosaic virus (CGMMV) and tomato brown rugose fruit virus (ToBRFV), and used these peptides to prepare high-specific polyclonal antibodies, CGMMV pAb-2 and ToBRFV pAb. CGMMV pAb-2 has no cross-reaction with other cucurbit viruses or tobamoviruses, with a detection limit of 0.075 μg plant tissue and an 87.0% detection rate for 23 CGMMV-infected cucumber cultivars. And it was utilized to devise indirect enzyme-linked immunosorbent assay (ELISA) and dot-ELISA. ToBRFV pAb shows a 100.0% detection rate for three field ToBRFV-infected tomato fruits. Further analysis reveals that the tobamoviral CP protein structure is conserved, and the approximately 15 residues at C-terminus may be a good antigenic epitope, providing a new approach to prepare antibodies for the rapid detection of tobamoviruses.

# Rubber biosynthesis drives the biogenesis and development of rubber particles, the rubber-producing organelles

Tiancheng Xu [a, b], Yongmei Li [a, b], Xing Liu [a, b], Xue Yang [a, b], Zhejun Huang [a, b], Jianfeng Xing [a, b], Cuili Liang [a, b], Junyi Li [a, b], Yingchao Tan [a, b], Shengmin Zhang [a, b], Jiyan Qi [a, b], De Ye [a, b], Zhonghua Li [a, b], Jie Cao [a, b], Chaorong Tang [a, b, *], Kaiye Liu [a, b, *]

[a] *National Key Laboratory for Tropical Crop Breeding, School of Tropical Agriculture and Forestry, School of Breeding and Multiplication (Sanya Institute of Breeding and Multiplication), Hainan University, Sanya, 572025, China*

[b] *Natural Rubber Cooperative Innovation Center of Hainan Province and Ministry of Education of P.R.C., Hainan University, Haikou, 570228, China*

E-mail: kaiyeliu@hainanu.edu.cn

## Abstract

Rubber particles (RPs) are specialized organelles for the biosynthesis and storage of natural rubber in rubber-producing plants. However, the mechanisms underlying the biogenesis and development of RPs remain unclear. In this study, two latex-specific *cis*-prenyltransferases (CPTs), TkCPT1 and TkCPT2, were identified in *Taraxacum kok-saghyz*, with almost identical orthologues retained across other *Taraxacum* species. For the first time, *Tkcpt1* single and *Tkcpt1/2* double mutants were successfully generated using the CRISPR/Cas9 system. Rubber biosynthesis was significantly depressed in *Tkcpt1* mutants, and completely blocked in *Tkcpt1/2* mutants. The absence of RPs in the *Tkcpt1/2* was confirmed using oil red O and Nile red staining, high-speed centrifugal stratification, cryo-SEM and TEM on fresh latex or laticifer cells. Transcriptomic and proteomic analyses revealed that, in the latex of *Tkcpt1/*2, rubber biosynthesis was blocked at the protein level, while metabolomic profiling indicated an enrichment of lipids and terpenoids. Furthermore, knockout of *TkCPTL1*, a latex-specific *CPT-Like* gene that encodes a rubber transferase activator, resulted in outright disruption of rubber biosynthesis and RP ontogeny, a phenotype similar to that of *Tkcpt1/2* mutants. These findings indicate that rubber biosynthesis is a driving force for the biogenesis and development of RPs, providing new insights into rubber production mechanisms.

**Prediction and quantification of potential banana fruit damage under robotic harvesting and de-handing**

Jie Guo[1,2], Zhou Yang[3], Jieli Duan[3], Xirui Zhang[1,*], Yong He[2,*], Manoj Karkee[4]

[1] *Hainan University, Haikou, China*

[2] *Zhejiang University, Hangzhou, China*

[3] *South China Agricultural University, Guangzhou, China*

[4] *Cornell University, New York, USA*

E-mail: jie.guo@hainanu.edu.cn

## Abstract

Squeezing, collision, friction and other behaviors are widely present in the post-harvest processing operations of bananas, such as field picking, de-handing, cleaning, packaging and transportation, which will inevitably cause a certain degree of mechanical damage to banana fruits. In view of the difficulty in accurately detecting and quantifying fruit damage in the early stage of production, this study adopted dynamic finite element numerical simulation and multi-parameter optimization of response surface to carry out simulation analysis on the collision behavior of banana fruit in the post-harvest operations, and the accurate assessment and quantitative characterization of fruit damage degree and erosion rule were carried out. The three-dimensional solid model of banana was established by reverse engineering method based on 3D scanning technology. The multi-scale finite element model of banana was further established by combining the constitutive parameters of banana bunch obtained from quasi-static mechanical experiments. In this study, a total of 27 collision systems including different contact materials, collision angles and impact speeds were developed to quantitatively characterize the changing rules of contact force, equivalent stress, energy, damage volume and bruise susceptibility of fruit under different experimental scenarios. An empirical prediction model for banana fruit's bruise susceptibility was established using response surface optimization methodology, and the comprehensive effects of contact material, collision angle and impact speed on fruit damage were obtained. On this basis, three dynamic collision systems including banana hand and impact panel were developed, and the generation region and erosion rule of equivalent stress of banana hand and impact panel under different encounter postures were further investigated. Finally, the physical verification experiments of banana impact and collision were carried out based on the characteristics of banana growth distribution and movement in the actual post-harvest operations. These results indicate that the multi-scale dynamic finite element model and the empirical prediction model of collision system for bananas are reasonable and correct, and the study have certain reference value for the numerical simulation and quantitative characterization of damage degree of other fruits and vegetables.

**Identification and Expression Analysis of the Sea Buckthorn *Dof* Gene Family in the Whole Genome**

LU Mei[1], DING Jian[1,2,3,4*], RUAN Chengjiang[1,2,3,4]

[1] *Institute of Plant Resources,Dalian Minzu University,Dalian 116600,Liaoning,China;*

[2] *State Key Laboratory of Utilization of Woody Oil Resource, Central South University of Forestry and Technology, Changsa 410000, Hunan, China*

[3] *Key Laboratory of Biotechnology and Bioresources Utilization, Ministry of Education, Dalian Minzu University, Dalian 116600, Liaoning, China*

[4] *Institute of Plant Resources, Dalian Minzu University, Dalian 116600, Liaoning, China*

E-mail: dingjian@dlnu.edu.cn

## Abstract

Identification of *Dof* (DNA binding with one finger) gene family members in *Hippophae rhamnoides* subsp. *mongolica* and exploration of key *Dof* genes involved in oil synthesis and accumulation in the fruit flesh bioinformatics approaches were used to identify the *Dof* gene family members in the sea buckthorn genome, followed by a systematic analysis of their protein domains, conserved motifs, cis-regulatory elements, and phylogenetic relationships. Absolute quantification transcriptome sequencing (Unique Molecular Identifier RNA-seq, UMI RNA-seq) was performed on the fruit flesh of 12 sea buckthorn germplasm lines. Candidate key *Dof* genes were identified based on the expression levels of *Dof* family members and the oil content in the fruit flesh. A total of 47 *Dof* members were identified in the sea buckthorn genome, all of which contained the typical *Dof* domain and were distributed across 12 chromosomes. Ten motifs were identified, with cis-regulatory elements related to plant oil synthesis and development. The *Dof* gene family was classified into three subfamilies, and phylogenetic analysis revealed that some sea buckthorn Dofs were closely related to the Dofs regulating oil synthesis in soybean (NP001236530.2, NP001315363.1). UMI RNA-seq analysis of the fruit flesh during different developmental stages of the sea buckthorn germplasms showed that the expression of *HrDof1.2* (Hic asm 12.3128)gene during the orange fruit stage was significantly higher than in the green fruit stage, and was closely correlated with the variation in oil content in the corresponding germplasms. Members of the sea buckthorn *Dof* gene family are involved in oil synthesis and development. The expression of the *HrDof1.2* gene is closely related to the oil content in the fruit flesh, providing a scientific basis for further elucidation of the regulatory mechanism of oil synthesis and accumulation by *Dof* genes in sea buckthorn.

# The FvMYB17 targets *FvDHQS* to promote ellagic acid biosynthesis in strawberry

Yingying Lei[1], Yuxin Nie[1], Jing Wang[1], Shuo Zhao[1], Jinxiang Yao[1], Junxiang Zhang[1]*, HongyanDai[1]* and ZhihongZhang[1]*

[1] *Shenyang Agricultural University*

E-mail:13804984105@163.com; zhangz@syau.edu.cn

## Abstract

Subgroup 4 R2R3-MYB factors are known to act as MYB repressors of phenylpropanoid metabolism in most plant species. However, whether these MYB repressors regulate other plant secondary metabolisms, such as ellagic acid, remains unclear. Additionally, the mechanism by which some MYB repressors function as activators remains unknown. MYB repressors typically contain conserved repression motifs, including the C2 motif (pdLNL$^{D}/_{E}$Lxi$^{G}/_{S}$) and the C5 motif (TLLLFR), which recruit corepressors via the $L_XL_XL_X$ core motif. In this study, we identified FvMYB17, a subgroup 4 R2R3-MYB repressor in woodland strawberry (*Fragaria vesca*). The stable overexpression of *FvMYB17* in strawberry fruits led to a significant increase in ellagic acid content. Amino acid sequence analysis revealed a single residue substitution within the C2 motif of FvMYB17, which impaired its interaction with the corepressor FvTPR2, potentially explaining its conversion into a transcriptional activator. Furthermore, mutating the C5 motif restored the lack of interaction between FvMYB17 and FvTPR2, even when the histidine in the C2 motif was mutated. Additionally, FvMYB17 is directly bound to the promoter of 3-dehydroquinate synthase (*FvDHQS*), a key gene in the gallic acid biosynthesis pathway, and activated its expression. Gallic acid serves as a precursor for ellagic acid biosynthesis, and stable silencing of *FvDHQS* in transgenic strawberry plants significantly reduced ellagic acid accumulation. These findings expand our understanding of MYB17 functionality in plants and provide insights for improving strawberry fruit quality through metabolic engineering.

**Root-to-Shoot Mobile mRNA CmoKARI1 Promotes JA-Ile Biosynthesis to Confer Chilling Tolerance in Grafted Cucumbers**

Miao Zhang[1†], Wenqian Liu[1†], Cuicui Wang[1†], Shujin Lin[2†], Ying Chen[1], Heran Cui[1], Chenggang Xiang[3], Yujia Ma[1], Xiaojun Li[1], Yongzhen Lu[1], Xiao Han[2], Yihan Dong[4@], Lihong Gao[1] and Wenna Zhang[1*, #]

[1] *Beijing Key Laboratory of Growth and Developmental Regulation for Protected Vegetable Crops, China Agricultural University, Beijing, 100193, China*

[2] *College of Biology Science and Engineering, Fuzhou University, Fuzhou, 350116, China.*

[3] *College of Life Science and Technology, Honghe University, Mengzi, Yunnan, 661100, China.*

[4] *Institut de Biologie Moléculaire des Plantes, Centre National de la Recherche Scientifique, UPR 2357, Université de Strasbourg, 67000, Strasbourg, France.*

E-mail：zm130125@163.com

## Abstract

Pumpkin rootstock is commonly used to graft cucurbit crops, improving their ability to withstand stress. While the significance of systemic signals from rootstocks to scions is recognized, the role of root-to-shoot transported mRNAs remains understudied. Cucumber plants often face growth and productivity limitations due to low temperatures. To shed light on the enhancement of chilling tolerance in grafted cucumber scions by pumpkin-derived mRNAs, we revisited the metabolomic and transcriptomic dataset from the cucumber/pumpkin heterograft under chilling condition. We identified pumpkin Ketol-acid reductoisomerase 1 (CmoKARI1) as the key mobile mRNA that specifically travels from pumpkin rootstock to cucumber scion upon early chilling stress. Overexpressing CmoKARI1 resulted in increased isoleucine level and chilling tolerance in both cucumber and Arabidopsis. The increased isoleucine was further used to synthesize JA-Ile conjugates, activating JA-Ile signaling and enabling heterografts to weather low temperatures. This study represents the first instance of a unidirectional mobile mRNA triggered by specific environmental cues.

**Title: CabHLH035 interacts with CaJAZ7 to regulate CaAOC2-mediated OPDA synthesis and $Na^+/K^+$ homeostasis under salt stress in pepper**

Rugang Chen[1], Huafeng Zhang[1]

[1] *College of Horticulture, Northwest A&F University, Yangling 712100, China*

E-mail: rugangchen@126.com; huafengzhang@nwafu.edu.cn

## Abstract

Members of the bHLH family of transcription factors play important roles in salt stress responses. Salt stress poses a significant challenge to plant growth and agricultural productivity. In this study, Our results show that the expression of *CabHLH035* increased following salt treatment. Transient expression of *CabHLH035* (*CabHLH035*-To) in pepper enhanced salt tolerance, whereas knocking down the expression of *CabHLH035* in pepper plants resulted in decreased salt tolerance. We found that transgenic Arabidopsis plants, ectopic expression of *CabHLH035* and pepper plants transiently overexpressing *CabHLH035* (*CabHLH035*-To) showed lower $Na^+$ content in response to salt treatment, while *CabHLH035*-silenced plants had higher $Na^+$ concentration. Although the exogenous application of methyl jasmonate (MeJA) has been shown to enhance salt tolerance in pepper (*Capsicum annuum*) seedlings, the underlying molecular mechanisms remain largely unclear. In this study, we identify and characterize a regulatory module comprising *CabHLH035*, *CaJAZ7*, and *CaMIEL1* that mediates JA-dependent salt stress responses. Genetic and physiological analyses revealed that CabHLH035 interacts with CaJAZ7 to regulate sodium ($Na^+$) homeostasis under salt stress. Silencing of *CaJAZ7* resulted in enhanced $Na^+$ efflux from roots and increased activity of both vacuolar and plasma membrane $H^+$-ATPases, thereby disrupting $Na^+$ compartmentalization and export. Moreover, the salt tolerance conferred by *CabHLH035* was abolished in *CaJAZ7* knockdown plants, indicating a critical dependence on *CaJAZ7* function. Mechanistically, *CaMIEL1* facilitates the ubiquitination and 26S proteasome-mediated degradation of CaJAZ7. Silencing of *CaMIEL1* enhanced $Na^+$ translocation to roots and exacerbated salt sensitivity. Additionally, CabHLH035 directly binds to the promoter region of *CaAOC2*, a key gene involved in JA biosynthesis, and activates its expression, suggesting a positive feedback loop that amplifies JA synthesis during stress. Our findings clarify the *CabHLH035*-*CaJAZ7* regulatory module that coordinates $Na^+$ homeostasis through $H^+$-ATPase regulation, *CaMIEL1*-mediated ubiquitination, and *CaAOC2*-driven JA biosynthesis, offering new insights into the mechanisms of JA-mediated salt tolerance in plants.

# Analysis of the Effects of Different Processing Methods on the Metabolite Composition and Anti-inflammatory Activity of Illicium verum on Metabolomics and Network Pharmacology

Luan Xu[1], Wenhai Li[1], Bo Zhao[*]

[1] *Guilin Medical University*

## Abstract

*Illicium verum*, a plant within the Schisandraceae family, is widely cultivated in southern China. Its fruit serves as both a traditional Chinese medicine and a common spice, known for its warm and pungent properties with effects such as dispelling cold, regulating qi, and alleviating pain. Modern studies indicate that various parts of the plant contain volatile oils, phenylpropanoids, sesquiterpene lactones, and flavonoids, which exhibit anti-inflammatory, analgesic, and anti-atherosclerotic activities. As Guangxi produces 85% of the global and 90% of China ' s star anise output, its international trade potential is expanding. This study investigated the anti-inflammatory components and mechanisms of *I. verum* under different processing methods (freeze-drying, oven-drying, sun-drying) using non-targeted metabolomics, network pharmacology, and in vitro cell experiments. Non-volatile metabolomics revealed that lipids and terpenoids were the most abundant classes. Freeze-drying best preserved metabolic content, while sun-drying caused significant loss. In vitro anti-inflammatory assays showed that the freeze-dried extract exhibited the strongest activity. Integrative analysis identified kaempferol and luteolin as key anti-inflammatory flavonoids, targeting SRC and STAT3, and involving pathways such as cancer and hepatitis B. These findings support the development of star anise-based natural anti-inflammatory agents and provide insights into its processing-dependent bioactivity.

# Molecular basis and functional analysis of abiotic resistance genes in tomatoes based on multi-omics

Jun Yang[1], Peng Cao[1], Yingchen Hao[1], Qi Qiang, Shouchuang Wang[1]

[1] *State Key Laboratory of Tropical Crop Breeding, School of Breeding and Multiplication (Sanya Institute of Breeding and Multiplication), Hainan University, Sanya 572025, Hainan, China*

## Abstract

Plant metabolites are crucial for the growth, development, environmental adaptation, and nutritional quality of plants. Plant metabolomics, a key branch of systems biology, involves the comprehensive analysis and interpretation of the composition, variation, and functions of these metabolites. Advances in technology have transformed plant metabolomics into a sophisticated process involving sample collection, metabolite extraction, high-throughput analysis, data processing, and multidimensional statistical analysis. Multiple omics strategies based on the metabolomics can be integrated to reveal interactions and regulatory networks across different molecular levels, deepening our understanding of plant biological processes. Several key regulators of abiotic stress have been characterization in tomato in the pathway of steroidal glycoalkaloids and phenolamides. SlERF.H6 repressed the expression of SGA biosynthetic glycoalkaloid metabolism (GAME) genes and caused a subsequent decrease in the abundance of bitter SGAs. SlMYB13 positively regulates the expression of two phenolamide biosynthetic gene clusters BGC7 and BGC11 to enhance drought tolerance by enhancing scavenging of reactive oxygen species and increasing abscisic acid content in tomato. It is the important mechanism of metabolites biosynthesis and regulation in tomato and reveals that crop domestication and improvement shapes metabolic diversity to affect plant environmental adaptation.

**Sorbitol enhances tomato resistance to Botrytis cinerea via the JA pathway**

Yue Yang, ZiYing Kong, Yang Li

*College of Plant Protection, Shandong Agricultural University*

E-mail: yangyuezi2021@163.com

## Abstract

During tomatoes (*Solanum lycopersicum* L.) growth, gray mold disease caused by *Botrytis cinerea* infection seriously inhibits plant growth and development, often leading to a significant decrease in tomato yield and deterioration of fruit quality, seriously threatening the economic benefits of the tomato industry. In this study, metabolomics techniques were employed to explore the metabolic interaction mechanism between *Botrytis cinerea* and its host tomato, and sorbitol, galactitol and mannitol were identified. The results showed that although three metabolites could not directly inhibit *Botrytis cinerea* mycelial growth, exogenous spraying significantly enhanced tomato plants' resistance to gray mold disease, with sorbitol exhibiting the best effect. Further experiments revealed that sorbitol induced the burst of reactive oxygen species (ROS) in tomato leaves, and thereby activated the expression of genes related to the jasmonic acid (JA) pathway. In tomato fruits, sorbitol treatment could maintain the storage quality of the fruits, and significantly reduce the affected area of tomato gray mold disease. In conclusion, sorbitol, as an immune inducer, has demonstrated potential application value in the green control of tomato gray mold disease. This finding lays an important theoretical and practical foundation for the development of environmentally friendly plant-based antifungal preparations and the establishment of a green control technology system for tomato gray mold disease.

# Editing the CsRIN4 Gene Confers Resistance to Citrus Canker in Citrus sinensis

Ting Wang[1,2] , Yun Zhong[1]*, Yuanda Lv[1], Bo Jiang[1], Xiangyang Li[1,3]

[1] *Institute of Fruit Tree Research, Guangdong Academy of Agricultural Sciences, Guangzhou, China*

[2] *College of Horticulture, South China Agricultural University, Guangzhou, China*

[3] *Key Laboratory of South Subtropical Fruit Biology and Genetic Resource Utilization, Ministry of Agriculture and Rural Affairs; Guangdong Provincial Key Laboratory of Science and Technology Research on Fruit Tree, Guangzhou, China*

E-mail: zhongyun99cn@163.com

## Abstract

Citrus canker, caused by *Xanthomonas citri ssp. citri* (*Xcc*), is a devastating disease that threatens most citrus species and cultivars, especially sweet oranges (*Citrus sinensis*). Enhancing resistance to citrus canker disease has been a primary objective in citrus breeding. And the exploitation of disease-susceptible genes is high suitable for gene-editing breeding aimed at improving resistance. RIN4 (RPM1-interacting protein 4) is a widely studied conserved plant immunomodulatory protein. It plays a pivotal role in both PTI (pathogen-associated molecular pattern-triggered immunity) and ETI (effector-triggered immunity). And previous researches have demonstrated that RIN4 functions as a negative regulator. So, RIN4 may be involved in citrus susceptibility to canker. In this study, we demonstrate that editing *CsRIN4* in two *Citrus sinensis* cultivars, 'Succari' (designated as AJTC) and 'Valencia' (designated as XC), results in strong resistance to citrus canker. Two guide RNAs [R4-T1 (ACTGAGTCTGCTCATC AACGAGG), and R4-T2 (GAGTCTGCTCATCAACGAGGAGG)] were designed to target CsRIN4 (*Cs5g27740* in the *Citrus sinensis* reference genome v1.0). Both gRNAs shared 100% identity with the two *RIN4* alleles and were separately cloned into a knockout vector backbone containing an *NPTII* (kanamycin resistance)-*GFP* (green fluorescence protein) gene fusion expression element and a *SpCas9* gene expression cassette. While both constructs were introduced into *Agrobacterium tumefaciens* EHA105, only the R4-T2 gRNA construct was successfully transferred into citrus through Agrobacterium-mediated transformation. Co-cultivation of 1500 'AJTC' and 1200 'XC' epicotyl segments with Agrobacterium infection, followed by kanamycin selection and GFP screening, yielded 3 and 2 GFP-positive shoots for 'AJTC' and 'XC', respectively, corresponding to transformation efficiencies of 0.13% and 0.25%. All the mutants were analysed for their possible resistance to *Xcc* by an in vitro pinprick inoculation. In R4-T2-2, around 50% of the reads carried the same 5-bp deletion. In R4-T2-3, approximately 50% reads contained a 3-bp deletion while the remaining half harboured a 1-bpdeletion. All the reads from R4-T2-4 shared an identical 1-bp insertion, implying that both alleles were mutated at the same site. For R4-T2-5, a 1-bp insertion and a 1-bp deletion were detected in reads in a nearly 1:1 ratio, showing that both alleles were mutated differently. Compared to wildtype 'AJTC', both mutant lines showed a significant reduction in canker lesion size at 10 days post inoculation (dpi), with 12.9% and 74.2% decreases for R4-T2-2 and R4-T2-3, respectively. Correspondingly, the *Xcc* cell titers in R4-T2-3 leaves were significantly lower at 5 and 10 dpi. Compared to wild type 'XC' sweet orange, the canker lesion sizes on the leaves of R4-T2-4 and R4-T2-5 were reduced by 72.1% and 84.9%, respectively, at 10 dpi. Agreeably, the *Xcc* titers in leaves of R4-T2-4 and R4-T2-5 were significantly lowered at both 5 and 10 dpi. In summary, mutations in *CsRIN4* induced by SpCas9 under the direction of the R4-T2 guide RNA were enhanced resistance to *Xanthomonas citri* subsp. citri.

## ORGANIZERS:

Xinjiang Uygur Autonomous Region Academy of Agricultural Sciences
Nanjing Agricultural University
*Horticulture Research*
Fujian Agriculture and Forestry University
Huazhong Agricultural University
Northeast Normal University
Northwest A&F University
Agricultural Genomics Institute at Shenzhen, CAAS
Shandong Agricultural University
Southern University of Science and Technology
Sichuan University
Wuhan Botanical Garden, CAS
Zhejiang University
State Key Laboratory of Crop Genetics & Germplasm Enhancement and Utilization, College of Horticulture, Nanjing Agricultural University
State Key Laboratory of Tropical Crop Breeding
Biological Breeding Laboratory, Xinjiang Uygur Autonomous Region Academy of Agricultural Sciences