

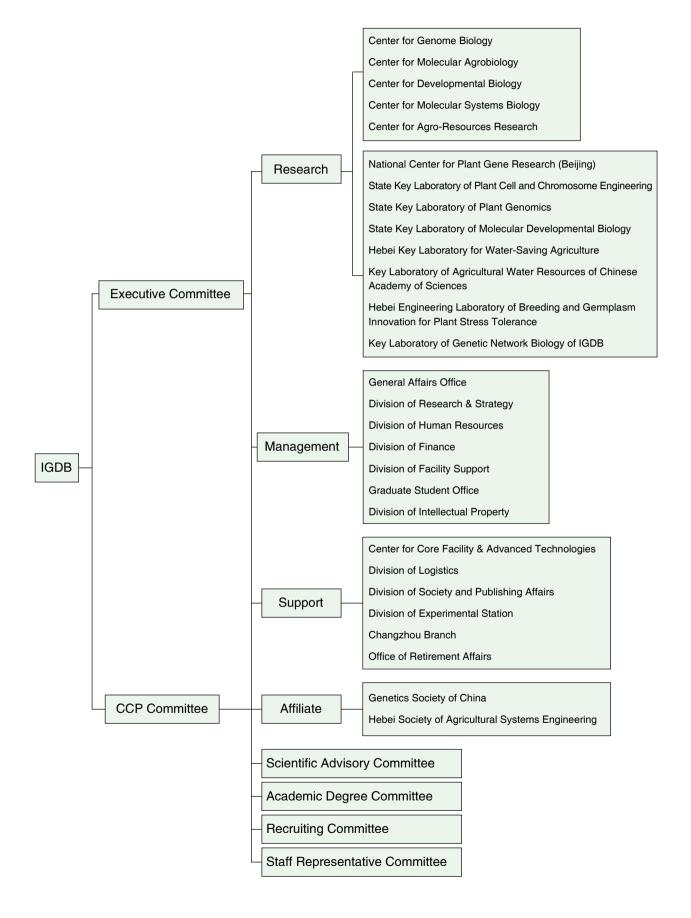
# 2020 Annual Report

Institute of Genetics and Developmental Biology Chinese Academy of Sciences





## Institute Organization



## **Director's Speech**



Year 2020 is an extraordinary year for IGDB. We experienced the inconvenience caused by the COVID-19 pandemic and successfully kept the virus at bay with strong solidarity among our staffs and students.

In 2020, IGDB achieved the targets set by the institute's "13<sup>th</sup> Five-year Plan" as evaluated by CAS, and launched the "Precision seed design and breeding" Strategic Priority Research Program of CAS. In addition, the Innovation Academy for Seed Design (INASEED) completed its preparation period and passed the evaluation by CAS, and also

launched its Hainan branch in Yazhou Bay, Sanya, Hainan Province.

In 2020, the College of Modern Agricultural Sciences, UCAS continued to strengthen its curriculum system and faculty. Currently, 10 CAS Members and 46 winners of the National Fund for Distinguished Young Scholars joined the faculty. 306 graduates were recruited in 2020. The College ranks the 6th in China and the 24th in the world by the 2020 ESI ranking.

In 2020, IGDB advanced in Science too. 404 SCI-indexed papers were published, 87 patents were authorized and 2 software copyrights were obtained. 20 new crop varieties were approved (registered). Dr. Xiangdong Fu and coworkers received the second prize of National Natural Science for their seminar work on coordinated regulation mechanism of high yield and high efficiency of nitrogen use in rice. Dr. Jianmin Zhou and his collaborators received The Outstanding Scientific and Technological Achievement Award of CAS for their contribution on molecular mechanism of plant immunity. Drs. Jiayang Li, Jianmin Zhou, Chengcai Chu, Caixia Gao and Chuanyou Li were recognized by Clarivate Analytics Hightly-Cited Researchers 2020.

Drs. Xiaofeng Cao and John Speakman were named foreign members of the US National Academy of Sciences. Dr. Yuling Jiao won the 16th China Youth Science and Technology Award. Dr. Guanghuo Shui and his collaborators won the innovation team title in key fields of the Innovative Talents Promotion Program of the Ministry of Science and Technology. Drs. Xiang Li, Yanjun Shen, Lei Wang and Shuping Qin were funded by the CAS Pioneer Youth Hundred Talent Program. Drs. Danhua Jiang and Bo Ren were supported by the National High-Level Talent Program, while Drs. Yang Bai and Guanghou Shui were selected as the leading talents of scientific and technological innovation in the "Ten Thousand Talents Program".

The Institute strengthened the application and commercialization of the research outputs, 12 patents/varieties/technologies were transformed in 2020. A joint laboratory was set up between IGDB and Beijing Children's Hospital. A multi-dimensional omics center was jointly established with BGI Life Science Research Institution.

International cooperation is indispensable for IGDB and facing challenging environment in the COVID-19 pandemic period. To strengthen international collaborations, the Center of Excellence for Plant and Microbial Science (CEPAMS) jointly launched by IGDB, the CAS Center of Excellence for Molecular Plant Sciences, and John Innes Center (UK), entered second phase of development. Professor Dale Sanders, Director of JIC, won the 2020 CAS Award for International Science and Technology Cooperation for his seminar contribution to CEPAMS. A virtual symposium between EMPRAPA (Brazil) and IGDB was held in 2020 to promote the establishment of China-Brazil Joint Research Center for Soybean Research. In addition, collaborative projects were successfully solicited by the Centre of Excellence for Plant-Environment Interactions (CEPEI), which was established jointly with Centre for Plant Biotechnology and Genomics (CBGP) in Spain and IGDB.

All these achievements and progresses are not possible without the efforts of IGDB staffs and students. Finally, I would like to express my sincere thanks to our friends, sponsors and funding agencies for their enormous support. I am pleased to present this annual report and wish you a prosperous 2021.

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Weicai Yang, Director, IGDB, CAS January 2021

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The mission of the Center for Genome Biology (the Center, thereafter) is to develop and apply genomic tools to understand how plant growth and development are regulated. In 2020, scientists in the Center have made important advances in multiple fronts, including gene function studies, plant genomic studies, plant hormone biology, plant health, plant biotechnology and molecular breeding.

In the area of gene function, Jiayang Li's group identified key strigolactone-responsive genes, revealed the mechanism through which a transcriptional repressor of hormone signaling can directly recognize DNA and regulate transcription in higher plants (Wang et al., *Nature*, 2020). The Li group and collaborators identified a high-tillering mutant *t20*, found that *T20* encodes ζ-carotene isomerase, demonstrated complex interactions in the biosynthesis of carotenoid, SLs and ABA (Liu et al., *Mol Plant*, 2020). The Li group and collaborators demonstrated that both SLs and KARs could trigger polyubiquitination and degradation of SMXL2, uncovering the convergent pathway in SL- and KAR-regulated gene expression and hypocotyl elongation (Wang et al., *Plant Cell*, 2020). The Li group and collaborators found that SL biosynthesis gene *HTD1* has been widely utilized and co-selected with *SD1* and contributed to the improvement of plant architecture in modern rice varieties (Wang et al., *Mol Plant*, 2020). The Li group and collaborators found that RNA-directed DNA methylation at MITEs positively and negatively regulates *D14* and *OsMIR156* family members respectively, contributing to the regulation of rice tillering (Xu et al., *Mol Plant*, 2020). The Li group engineered five saturated targeted endogenous mutagenesis editors (STEMEs) that can generate de novo mutations and facilitate directed evolution of plant genes (Li et al., *Nat Biotechnol*, 2020). Shanguo Yao's group and Chengcai Chu's group identified the *SLG1* gene, which encodes the cytosolic RCTU2, demonstrated that SLG1 is differentiated between *indica* and *japonica* rice, and the plants with the *indica* allele displayed enhanced thermostolerance (Xu et al., *Nat Commun*, 2020).

In the area of plant genomics, Xiaofeng Cao's group uncovered a novel mechanism by which AtPRMT3 cooperates with RPS2B to facilitate the dynamic assembly/ disassembly of the 90S/SSU processome during ribosome biogenesis and repress nucleolar stress (Hang et al., Mol Plant, 2020). The Cao group revealed the unique RNA structural features of G-rich regions, demonstrated that RG4 formation is sufficient to regulate gene expression and subsequently plant growth (Yang et al., Genome Biol, 2020). Zhukuan Cheng's group revealed that 21-nt phasiRNAs regulated male germ cell development through fine-tuning the expression of a large group of genes that together contribute to the progression of meiosis (Jiang et al., Nat Commun, 2020). The Cheng group found that mutation of OsRDR6 caused alterations in small RNA levels and ultimately led to the downregulation of genes related to DSB formation (Liu et al., Plant Cell, 2020). Yuling Jiao's group found that ATH1 anchors STM to activate STM as well as other axillary meristem regulatory genes, established a self-activation loop that maintains stem cells to enable branching (Cao et al., Curr Biol, 2020). The Jiao group and collaborators reported a genus-level whole-genome sequencing study of wheat, providing novel insights into the genetic driving forces behind wheat adaptation (Zhou et al., Nat Genet, 2020). The Jiao group and collaborators uncovered that leaf shape depends on cortical microtubule mediated cellulose deposition along the adaxial-abaxial axis in internal cell walls, revealed that microtubule-mediated mechanical feedback amplifies an initial asymmetry and maintained directional growth (Zhao et al., Curr Biol, 2020). Chengzhi Liang's group and collaborators de novo assembled 26 soybean genomes and constructed graph-based genome, identified large structural variations and gene fusion events on key agronomic traits (Liu et al., Cell, 2020). The Liang group and collaborators generated a chromosomelevel genome assembly of O. rufipogon using a combination of short-read sequencing, single-molecule sequencing, BioNano and Hi-C platforms (Shen et al., Mol Plant, 2020). Jingsong Zhang's group and collaborators annotated a high-quality sequence of the wild tetraploid peanut genome and performed comprehensive analyses of genomes and gene expression from diploid ancestors to wild and cultivated tetraploids (Yin et al., Adv Sci, 2020). Lihuang Zhu's group resequenced 1143 indica accessions, identified the mostshared SNP and indeled differences between two parents among all superior hybrids for each type of 3- and 2-line systems, and further identified the different loci associated with heterosis in 3- and 2-line hybrids (Lv et al., Nat Commun, 2020).

In the area of plant health, Jian-Min Zhou's group revealed how a plant defense metabolite protects plants by specifically disarming pathogen virulence machinery rather than non-selectively kill microbes. Cruciferous plants use sulforaphane to specifically inhibit a key transcription factor in a bacterial pathogen to dampen virulence activity without negative impact on commensal bacteria (Wang et al., *Cell Host Microbe*, 2020). The Zhou group provided evidence that pathogen proteins trigger resistosome formation *in vivo* and that this is required for disease resistance (Hu et al., *Mol Plant*, 2020). Yang Bai's group and collaborators identified the microorganisms associated with rhizobial nodulation, isolated candidate strains, and explored their roles in rhizobial nodulation, providing the evidence of a role for native microbiota in the adaptation of rhizobia to their environment and in the modulation of the symbiotic efficiency of rhizobia (Han et al., *ISME J*, 2020).

In the area of plant hormone biology, Jingsong Zhang's group and Shouyi Chen's group suggested that MHZ1/OsHK1 mediated the ethylene response partially independently of OsEIN2, and was directly inhibited by ethylene receptors, revealed mechanistic details of ethylene signaling for root growth regulation (Zhao et al., *Nat Commun*, 2020). The Zhang group found that MHZ11 facilitates ethylene signaling through modulating the ethylene receptor-mediated OsCTR2 phosphorylation, revealed a mechanism of MHZ11 participating in ethylene signaling for regulation (Zhao et al., *Plant Cell*, 2020). Yihua Zhou's group found that *MYB61* is regulated by

GRF4 and GRF4 is pivotal in controlling cellulose biogenesis, highlighted a connection of N&C interaction at the molecular level (Gao et al., Nat Commun, 2020). The Zhou group found that the phenylpropanoid pathway was required for the synthesis of both lignin and sporopollenin in vascular plants, discovered that the characteristics of sporopollenin were distinct among the tested land plants (Xue et al., Mol Plant, 2020). Qi Xie's group demonstrate that VPS23A is a critical regulator of the SOS pathway that positively modulates the regulation of salt tolerance, providing a potential strategy for crop salt tolerance engineering (Lou et al., Mol Plant, 2020). The Xie group found that VPS23A is an unstable protein that can be degraded by both the 26S proteasome and the endosome-vacuolemediated degradation pathways, identified that XBAT35 is a candidate protein controlling the stability of VPS23A (Yu et al., Mol Plant, 2020). The Xie group found that UBC27 interacts with the RING E3 ligase AIRP3 and forms an E2-E3 pair to promote the ubiquitination and degradation of ABI1, uncovered a regulatory mechanism of ABA signaling in plants (Pan et al., PNAS, 2020). Chuanyou Li's group demonstrated that MED25 recruits PRP39a and PRP40a to promote the full splicing of JAZ genes, exemplified how a mediator subunit integrates the effects of general splicing factors into a specific signaling pathway to accurately control gene expression (Wu et al., Plant Cell, 2020). The Li group and collaborators uncovered that SEU plays a fundamental role in the cell-fate determination of root stem cell organizers by coordinating the formation of a functional SCR-SEU-SDG4 transcriptional complex (Zhai et al., EMBO J, 2020). Jianru Zuo's group identified a transnitrosylase ROG1, found that ROG1 transnitrosylates GSNOR1 to promote the degradation of GSNOR1, thereby positively regulating nitric oxide (NO) signaling. This study revealed a unique mechanism in regulating NO signaling in plants (Chen et al., Dev Cell, 2020).

In the area of plant biotechnology and molecular breeding, Jiayang Li's group bred and released new rice elite cultivars "Zhongkefa 7", "Zhongkefa 8", "Zhongkefa 176", "Zhonglongjing 107" and "Quan 9 You 220". Shanguo Yao's group bred and released new rice elite cultivar "Zhongke 613". Zhukuan Cheng's group bred and released new rice elite cultivars "Zhongkeyan 3", "Zhongkeyan 5" and "Songkejing 108". Chuanyou Li's group bred and released new tomato cultivars "Zhongqinghong 1" and "Zhongqinggingfen 1".

In recognition of their contributions to related fields, Profs. Jiayang Li, Chengcai Chu, Yuling Jiao, Chuanyou Li, Guodong Wang, Qi Xie, Jian-Min Zhou, and Zhen Zhu were invited to contribute 23 review articles and commentaries for *Cell, Trends Plant Sci, Nat Plants, Mol Plant, New Phytol, Curr Opin Plant Biol, Plant Cell Environ*, and *J Exp Bot*.

#### AWARDS AND RECOGNITIONS

Prof. Xiaofeng Cao was elected as Foreign Member of the US National Academy of Sciences, and was honored with the Pollyanna Chu Outstanding Teacher Award of the Chinese Academy of Sciences, the research team was given the second prize of the State Natural Science Award. The plant immunity research team, led by Prof. Jian-Min Zhou, was awarded the 2020 Outstanding Science and Technology Achievement Prize of the Chinese Academy of Sciences. Prof. Yuling Jiao was honored with the 16th China Youth Science and Technology Award. Prof. Chengzhi Liang received the 2020 Outstanding Mentors Award of Chinese Academy of Sciences. Profs. Jiayang Li, Chengcai Chu, Chuanyou Li and Jian-Min Zhou were recognized as Clarivate Analytics Highly-Cited Researchers 2020.

### **Interaction Between Plant and Root Microbiome**

Yang Bai, Principal Investigator, Ph.D. (2010, University of Cologne, Germany).

High-throughput cultivation and identification of bacteria from the

Cultivating native bacteria from roots of plants grown in a given environment is essential

for dissecting the functions of the root microbiota for plant growth and health with strain-

specific resolution. Here, we established a straightforward protocol for highthroughput

bacterial isolation from fresh root samples using limiting dilution to ensure that the majority

of cultured bacteria originated from only one microorganism. This is followed by strain

characterization using a two-sided barcode PCR system to identify pure and heterogeneous

bacterial cultures. Our approach overcomes multiple difficulties of traditional bacterial

isolation and identification methods, such as obtaining bacteria with diverse growth rates

while greatly increasing throughput. To facilitate data processing, we developed an easy-touse bioinformatic pipeline called "Culturome" (https://github.com/YongxinLiu/Culturome) and a graphic user interface web server (http://bailab.genetics.ac.cn/culturome/). This protocol allows any research group (2–3 lab members without expertise in bioinformatics) to

Research Interests: The role of root microbiome in plant nutrition, disease resistance and yield, and related research methods.

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plant root microbiota



#### **Publications**

- Guo, X., Zhang, X., Qin, Y., Liu, Y.-X., Zhang, J., Zhang, N., Wu, K., Qu, B., He, Z., Wang, X., Zhang, X., Hacquard, S., Fu, X., and Bai, Y. (2020). Host-associated quantitative abundance profiling reveals the microbial load variation of root microbiome. Plant Communications, 1 (1): 100003.
- Han, Q., Ma, Q., Chen, Y., Tian, B., Xu, L., Bai, Y., Chen, W., and Li, X. (2020). Variation in rhizosphere microbial communities and its association with the symbiotic efficiency of rhizobia in soybean. The ISME Journal, 14 (8): 1915-1928.

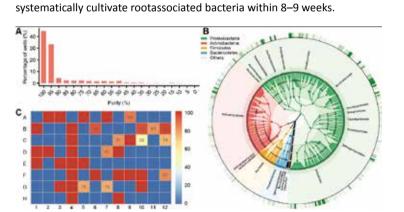
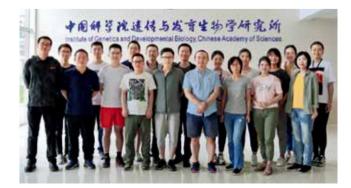


Figure: Anticipated results for bacterial cultivation from the Oryza sativa L. roots.





- Hang, R., Wang, Z., Yang, C., Luo, L., Mo, B., Chen, X., Sun, J., Liu, C., and Cao, X. (2020). Protein arginine methyltransferase 3 fine-tunes the assembly/disassembly of pre-ribosome to repress nucleolar stress by interacting with RPS2B in *Arabidopsis*. Molecular Plant, DOI: 10.1016/ i.molp.2020.10.006.
- Si, F., Cao, X., Song, X., and Deng, X. (2020). Processing of coding and non-coding RNAs in plant development and environmental responses. Essays Biochem. 64 (6): 931.
- Yang, X., Cheema, J., Zhang, Y., Deng, H., Duncan, S., Umar, M.I., Zhao, J., Liu, Q., Cao, X., Kwok, C.K., and Ding, Y. (2020). RNA G-quadruplex structures exist and function in vivo in plants. Genome Biology, 21 (1): 226.
- Xue, Y., Chen, R., Qu, L., and Cao, X. (2020). Noncoding RNA: from dark matter to bright star. Science China Life Sciences, 63 (4): 463.
- Jia, J., Long, Y., Zhang, H., Li, Z., Liu, Z., Zhao, Y., Lu, D., Jin, X., Deng, X., Xia, R., Cao, X., and Zhai, J. (2020). Post-transcriptional splicing of nascent RNA contributes to widespread intron retention in plants. Nature Plants, 6 (7): 780.

### **Epigenetics in Higher Plants**

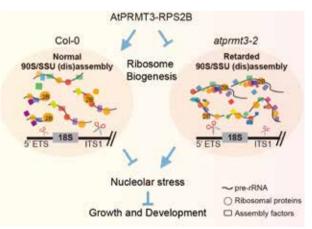
Xiaofeng Cao, Principal Investigator, Member of Chinese Academy of Sciences, Academician of The third World Academy of Sciences. Academician of Eurasian Academy of Sciences; Foreign academicians of the American Academy of Sciences.

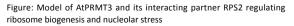
Research Interests: The laboratory mainly focuses on investigating the epigenetic regulation in higher plants, including histone methylation in plant development and genome stability, regulatory mechanism of protein arginine methylation in *Arabidopsis* and RNA metabolism in response to ambient temperature in rice.

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### Protein arginine methyltransferase 3 fine-tunes the assembly/ disassembly of pre-ribosome to repress nucleolar stress by interacting with rps2b in *Arabidopsis*

Ribosome biogenesis is strictly controlled and regulated. The defects of ribosome biogenesis can lead to embryo lethal of animals and plants, high frequency of human genetic diseases, and aberrant responses to environmental stress. The regulation of ribosome biogenesis has always been a hot topic in life science. However, the underlying mechanism of ribosome assembly in plants remains unknown. Our previous work found that Arabidopsis arginine methyltransferase AtPRMT3 is involved in ribosome biogenesis by promoting proper prerRNA processing. To reveal the molecular mechanism by which AtPRMT3 regulates prerRNA processing, we found that AtPRMT3 interacted with ribosomal protein RPS2 and worked together to regulate pre-ribosome assembly. The defects of AtPRMT3 result in unbalanced assembly/disassembly of 90S/SSU processome in nucleus. Further, the retarded dynamic assembly/disassembly pre-ribosome resulted in the imbalance of alternative prerRNA processing in atpmt3 mutant and nucleolar stress. This work elucidated the essential functions and molecular mechanisms of AtPRMT3 and RPS2 in the regulation of the dynamic assembly of nuclear 90S/SSU processome and the process of nucleolar stress. This work was published online in Molecular Plant on October 15, 2020 (DOI: 1 https://doi.org/10.1016/ j.molp.2020.10.006).







### **Plant Comparative Genomics**

Mingsheng Chen, Principal Investigator, Ph.D. (1998, Purdue University, USA).

Research Interests: Comparative genomics and genome evolution of Oryza and grasses.

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miliaceum L.



#### **Publications**

- Tian, P., Zhang, X., Xia, R., Liu, Y., Wang, M., Li, B., Liu, T., Shi, J., Wing, R.A., Meyers, B.C., and Chen, M. (2020). Evolution and diversification of reproductive phased small interfering RNAs in *Oryza* species. New Phytologist, DOI: 10.1111/nph.17035.
- Zou, X., Du, Y., Wang, X., Wang, Q., Zhang, B., Chen, J., Chen, M., Doyle, J.J., and Ge, S. (2020). Genome evolution in *Oryza* allopolyploids of various ages: Insights into the process of diploidization. The Plant Journal, DOI: 10.1111/tpj.15066.

Broomcorn millet (*Panicum miliaceum* L.) is a relatively complex allotetraploid. Since the parental progenitors of broomcorn millet have not been determined yet, the separation of the two subgenome is very challenging. We de novo assembled a highquality chromosome-scale genome sequence of *P. miliaceum*. By analyzing the collinearity of *P. miliaceum* and *Panicum hallii*, more than 80% orthologous genes of *P. hallii* are closer to one subgenome of *P. miliaceum*. Then we combined the homologous genes on each chromosome as a supergene to construct phylogenetic trees, and named the group which is evolutionarily closer to *P. hallii* as A subgenome of *P. miliaceum*, and the other group as B subgenome. Among the 13425 expressed genes in spikelet tissue of broomcorn millet, 9.22% genes are expressed in A subgenome only, 8.67% genes are expressed in B subgenome only, and 82.11% genes are co-expressed in both A and B subgenomes. The results showed that there was no significant expression advantage between A and B subgenomes. By analyzing the ATAC-seq data sets, we discovered that the highly expressed genes have more open chromatin than lowly expressed genes, and open chromatin peaks are mainly located at gene transcriptional start sites.

The separation and comparison of two subgenomes of Panicum

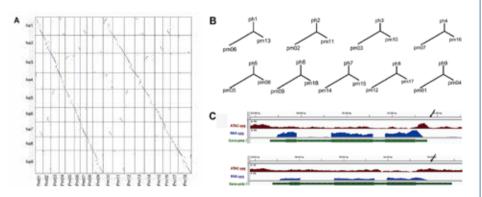


Figure: The genome comparison of *P. miliaceum* and *P. hallii*. (A) The dot plot of orthologous chromosomes between *P. miliaceum* and *P. hallii*. (B) Super-gene trees of *P. miliaceum* and *P. hallii*. (C) Sequencing tracks for the orthologous loci showed the distinct gene-specific ATAC-seq peaks at the promoter region (black arrow).





#### **Publications**

- Zhao, H., Ma, B., Duan, K.-X., Li, X.-K., Lu, X., Yin, C.-C., Tao, J.-J., Wei, W., Zhang, W.-K., Xin, P.-Y., Man Lam, S., Chu, J.-F., Shui, G.-H., Chen, S.-Y., and Zhang, J.-S. (2020). The GDSL lipase MHZ11 modulates ethylene signaling in rice roots. The Plant Cell, 32 1626-1643.
- Zhao, H., Duan, K.-X., Ma, B., Yin, C.-C., Hu, Y., Tao, J.-J., Huang, Y.-H., Cao, W.-Q., Chen, H., Yang, C., Zhang, Z.-G., He, S.-J., Zhang, W.-K., Wan, X.-Y., Lu, T.-G., Chen, S.-Y., and Zhang, J.-S. (2020). Histidine kinase MHZ1/OSHK1 interacts with ethylene receptors to regulate root growth in rice. Nature Communications, 11 (1): 518.
- Bian, X.H., Li, W., Niu, C.F., Wei, W., Hu, Y., Han, J.Q., Lu, X., Tao, J.J., Jin, M., Qin, H., Zhou, B., Zhang, W.K., Ma, B., Wang, G.D., Yu, D.Y., Lai, Y.C., Chen, S.Y., and Zhang, J.S. (2020). A class B heat shock factor selected for during soybean domestication contributes to salt tolerance by promoting flavonoid biosynthesis. New Phytologist, 225 (1): 268-283.

## Molecular Mechanisms of Abiotic Stress Response in Higher Plants and Soybean Seed Trait Regulation

**Shouyi Chen**, Principal Investigator Emeritus. She graduated from the Department of Biology of Peking University in 1963. She worked at the Institute of Biophysics, CAS, from 1963 to 1989, and worked at the Columbia University and Public Health Research Institute of City of New York from 1981 to 1984.

Research Interests: 1) Identification of new genes and new mechanisms related to plant abiotic stress tolerance; 2) Identification of genes for regulating seed trait by QTL mapping and genomic analysis.

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## A seed specific NFYA/TZF regulatory module controls lipid accumulation in soybean seeds

Oil accumulation in soybean seeds is regulated by a unique seed-specific lipid metabolism transcriptional network. However, limited information is known about these regulatory pathways. Here, we report that a tandem CCCH zinc finger protein, GmZF392, is a key regulator of lipid biosynthesis. Overexpression of *GmZF392* increases the seed oil content in transgenic *Arabidopsis* and soybean plants without detrimental effects on plant growth, development and final yield. Consistently, GmZF392 activates the expression of genes involved in glycolytic pathway, de novo synthesis of fatty acids and TAG assembly by directly binding to TPBE motifs present in the promoters of the target genes. Protein-protein interaction experiments demonstrate that GmZF392 and GmZF351 collaboratively activate the expression of fmZF392 and GmZF392 and GmZF391 to activate their mRNA expression directly and indirectly, respectively. Taken together, our study provides mechanistic insights into the modulation of seed oil biosynthesis, and presents an opportunity for improving oil production in soybean and other oilseed crops.



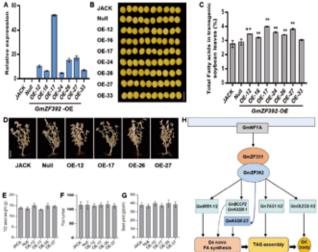


Figure: The NFYA/TZF module regulates lipid accumulation in soybean seeds A, Expression level of *GmZF392* in transgenic soybean plants. B, Morphology of seeds from Jack, Null, and *GmZF392*-transgenic soybean plants. Ten seeds are used for photographing (Bar = 1 cm). C, Total fatty acid contents in *GmZF392*-transgenic soybean seeds. D-G, Morphology, 100 seed weight, pod number and yield per plant of controls and *GmZF392*-transgenic soybean plants at the harvest stage. H, Working model of NFYA/TZF transcriptional regulatory network for oil accumulation.

### **Plant Molecular Cytogenetics**

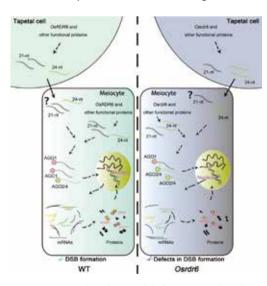
Zhukuan Cheng, Principal Investigator, Ph.D. (1999, Institute of Genetics, CAS, China).

Research Interests: Regulatory mechanism of plant meiosis.

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Meiosis is a specialized cell division that takes place in sexually reproducing eukaryotes and homologous recombination (HR) during meiosis increases diversity. During meiosis, HR is initiated by programmed DNA double-strand break (DSB) formation. RNA-dependent RNA polymerase 6 (RDR6) is a core component of the small RNA biogenesis pathway, but its function in meiosis is unclear. Here, we report a new allele of *OsRDR6 (Osrdr6-mei*), which causes meiosis-specific phenotypes in rice (*Oryza sativa*). In *Osrdr6-mei*, meiotic DSB formation is partially blocked. We created a biallelic mutant with more severe phenotypes, *Osrdr6-bi*, by crossing *Osrdr6-mei* with a knockout mutant, *Osrdr6-edi*. In *Osrdr6-bi* meiocytes, 24 univalents were observed, and no histone H2AX phosphorylation foci were detected. Compared with the wild type, the number of 21-nt small RNAs in *Osrdr6-mei* was dramatically lower, while the number of 24-nt small RNAs was significantly higher. Thousands of differentially methylated regions (DMRs) were discovered in *Osrdr6-mei*, implying that OsRDR6 plays an important role in DNA methylation. There were 457 genes down-regulated in *Osrdr6-mei*, including three



genes, CENTRAL REGION COMPONENT 1 (CRC1), P31<sup>comet</sup> and Oryza sativa SOLO DANCERS (OSSDS), related to DSB formation. Interestingly, the downregulated genes were associated with a high level of 24-nt small RNAs, but less strongly associated with DMRs. Therefore, we speculate that the alteration in expression of small RNAs in Osrdr6 mutants leads to the defects in DSB formation during meiosis, which might not be directly dependent on RNA-directed DNA methylation.



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- Jiang, P.F., Lian, B., Liu, C.Z., Fu, Z.Y., Shen, Y., Cheng, Z.K., and Qi, Y.J. (2020). 21-nt phasiRNAs direct target mRNA cleavage in rice male germ cells. Nature Communications, 11 (1): 1591.

Figure: A proposed working model of OsRDR6-mediated DSB formation in rice meiosis. In *Osrdr6* mutants, increased 24-nt small RNA levels lead to down-regulation of 457 genes through binding to AGO2/4. Three genes associated with meiotic DSB formation, *OsSDS*, *P31<sup>comet</sup>* and *CRC1*, are among the down-regulated genes. As a result, meiotic DSB formation is defective in *Osrdr6* mutants.





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- Hu B. and Chu C. (2020) Nitrogen-phosphorus interplay: old story with molecular tale. New Phytologist 225(4): 1455-1460.
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- Tang J. and Chu C. (2020) Strigolactone signaling: Repressor proteins are transcription factors. Trends in Plant Science 25(10): 960-963.

## **Rice Functional Genomics and Agrobiotechnology**

## Chengcai Chu, Principal Investigator, Ph.D. (1996, Martin-Luther University, Germany).

Research Interests: The laboratory mainly focuses on the dissecting the molecular basis of nutrient use efficiency and source-sink interaction using a combination of genetic, biochemical, physiological, and molecular tools. We are also interested in molecular design of super green rice based on the knowledge, resources and tools obtained from our studies.

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## Genetic dissection of molecular basis of rice geographical adaptation to soil nitrogen

Application of nitrogen fertilizer largely contributes to crop yield improvement in modern agriculture and global food security. Meanwhile, overuse of nitrogen fertilizer caused severe threat to the environment such as air pollution, water eutrophication, and soil acidification. Thus, dissecting the genetic basis of nitrogen use efficiency (NUE) in rice natural population is effective way for high NUE crop breeding. We assessed the nitrogen response of 110 accessions from Rice Mini-Core Collection, which collected from 52 countries/regions worldwide, under three different nitrogen conditions of field. Among all the traits examined, tiller number displayed the most significant response to increased nitrogen supply. We therefore defined the tillering response to nitrogen (TRN) as the relative NUE phenotype and carried out genome-wide association study (GWAS) for NUE. OsTCP19, which encodes a TCP transcription factor, was identified as the casual gene of TRN via candidate gene annotation and transcript nitrogen response detection. Transcript abundance of OsTCP19 was negatively regulated by increasing nitrogen, and as a transcription factor, it could repress the expression of rice tillering-promoting gene, thus leading to the function of regulating TRN. We further found that a 29-bp InDel in OsTCP19 promoter region was the casual variation underlying the TRN divergence among different rice varieties in Rice Mini-Core Collection. OsTCP19 promoter without the 29-bp InDel, which exists in high-TRN varieties, was bound directly by OsLBD proteins, the negative regulators of nitrogen response, and resulting in its transcription activity repression and higher TRN. Conversely, in low-TRN varieties, the 29-bp insertion in OsTCP19 promoter, which is adjacent to the binding sites of OsLBD, partially prevents the repression of OsLBD proteins, resulting in low expression nitrogen response and TRN. We next performed multiple transcriptomics analysis and identified DLT, a tiller-promoting gene, as a vital target gene of OsTCP19. Genetic analysis and biochemical experiment showed that OsTCP19 could directly bind to the promoter of DLT and repress its expression, thereby involving in modulating rice tillering. Therefore, OsTCP19-DLT module underlies the molecular basis of nitrogen-regulated rice tiller development. Genetic analysis showed that high-TRN genotype of OsTCP19 (OsTCP19-H) mainly existed in the varieties derived from poor soil regions while lost in most modern cultivars. Correlation analysis showed that the allele frequency of OsTCP19-H is negatively correlated with soil nitrogen content. Besides,

analysis showed that the allele frequency of *Os* we also found high *OsTCP19*-H allele frequency (> 90%) in wild rice, indicating that *OsTCP19*-H contributes to geographic adaptation to low soil nitrogen and is preserved in rice domestication. Furthermore, introducing *OsTCP19*-H into modern rice cultivars could significantly increase its yield and NUE under reduced nitrogen conditions. Collectively, our work not only unveils the genetic basis underlying the geographical adaptation towards the soil nitrogen content of cultivated rice, but also provides a promising target for large-scale application in high NUE breeding.

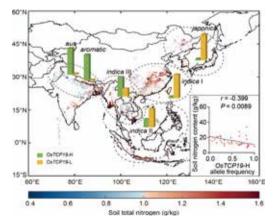




Figure: *OsTCP19*-H frequency is significantly correlated with soil nitrogen content. Main plot, geographical distribution of soil nitrogen content and *OsTCP19* allele frequency among different rice subgroups in Asia. The color key (blue to red) represents soil total nitrogen content (g/kg). Different rice types include *japonica* (n = 874 accessions), *indica* II (n = 205 accessions), *indica* II (n = 217 accessions), *indica* III (n = 891 accessions), and *aus* (n = 811 accessions) and *aus* (n = 201 accessions). Subplot, Pearson correlation coefficient analysis of *OsTCP19*-H allele frequency with soil total nitrogen content of 42 countries/regions worldwide.

## **Plant Chromatin Regulation**

**Danhua Jiang**, Principal Investigator, Ph.D. (2012, National University of Singapore, Singapore).

Research Interests: The function of histone variants and their incorporation mechanisms. Epigenetic mechanisms of plant environmental response.

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## The evolution and functional divergence of the histone H2B family in plants

Chromatin regulation of eukaryotic genomes depends on the formation of nucleosome complexes between histone proteins and DNA. Histone variants, which are diversified by sequence or expression pattern, can profoundly alter chromatin properties. While variants in histone H2A and H3 families are well characterized, the extent of diversification of histone H2B proteins is less understood. Here, we report a systematic analysis of the histone H2B family in plants, which have undergone substantial divergence during the evolution of each major group in the plant kingdom. By characterising *Arabidopsis* H2Bs, we substantiate this diversification and reveal potential functional specialization that parallels the phylogenetic structure of emergent clades in eudicots. In addition, we identify a new class of highly divergent H2B variants, H2B.S that specifically accumulate during chromatin compaction of dry seed embryos in multiple species of flowering plants. Our findings thus identify unsuspected diverse properties among histone H2B proteins in plants that has manifested into potentially novel groups of histone variants.

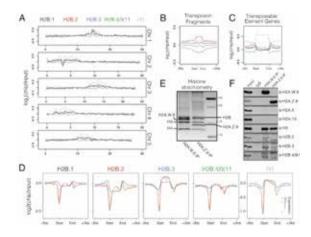


Figure: **Genomic distribution of somatic** *Arabidopsis* **H2Bs.**(A) Chromosomal distribution of somatic H2Bs alongside histone H3 over each of the five *Arabidopsis* chromosomes calculated in 100 kb bins. (B-C) Distribution of somatic H2Bs and histone H3 over transposon fragments (B) and transposable element genes (C). Plotted is the ChIP-seq log<sub>2</sub> enrichment relative to input. (D) Distribution of somatic H2Bs and histone H3 over genes grouped by their level of expression in *Arabidopsis* seedlings. (E) Silver stained gel of immunoprecipitated H2A.W.6 and H2A.Z.9 mono-nucleosomes confirms histone stoichiometry. Identities of protein bands are indicated. (F) Western blotting analysis of samples in panel E with indicated antibodies.





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Jiang, D.H., Borg, M., Lorkovic, Z.J., Montgomery, S.A., Osakabe, A., Yelagandula, R., Axelsson, E., and Berger, F. (2020). The evolution and functional divergence of the histone H2B family in plants. PLoS Genetics, 16 (7): e1008964.



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- Zhou, Y., Zhao, X., Li, Y., Xu, J., Bi, A., Kang, L., Xu, D., Chen, H., Wang, Y., Wang, Y.G., Liu, S., Jiao, C., Lu, H., Wang, J., Yin, C., Jiao, Y., and Lu, F. (2020). Triticum population sequencing provides insights into wheat adaptation. Nature Genetics, 52:1412-1422.
- Du, F. and Jiao, Y. (2020). Mechanical control of plant morphogenesis: concepts and progress. Current Opinion in Plant Biology, 57: 16-23.
- Du, F., Mo, Y., Israeli, A., Wang, Q., Yifhar, T., Ori, N., and Jiao, Y. (2020). Leaflet initiation and blade expansion are separable in compound leaf development. The Plant Journal, 104 (4): 1073-1087.

## Systems Biology of Plant Organogenesis

Yuling Jiao, Principal Investigator, Ph.D. (2006, Yale University, USA).

Research Interests: We combines transcriptome analysis, imaging, molecular genetics, and modeling to understand shoot lateral appendage formation and patterning, and its application in molecular design of wheat spike architecture.

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## Single-nucleus RNA-seq resolves spatiotemporal developmental trajectories in the tomato shoot apex

Single cell transcriptomics is revolutionizing our understanding of development and response to environmental cues. Recent advances in single cell RNA sequencing (scRNA-seq) technology have enabled profiling gene expression pattern of heterogenous tissues and organs at single cellular level and have been widely applied in human and animal research. Nevertheless, the existence of cell walls significantly encumbered its application in plant research. Protoplasts have been applied for scRNA-seq analysis, but mostly restricted to tissues amenable for wall digestion, such as root tips. However, many cell types are resistant to protoplasting, and protoplasting may yield ectopic gene expression and bias proportions of cell types. Here we

demonstrate a method with minimal artifacts for high-throughput singlenucleus RNA sequencing (snRNA-Seg) that we use to profile tomato shoot apex cells. The obtained highresolution expression atlas identifies numerous distinct cell types covering major shoot tissues and developmental stages, delineates developmental trajectories of mesophyll cells, vasculature cells, epidermal cells, and trichome cells. In addition, we identify key developmental regulators and reveal their hierarchy. Collectively, this study demonstrates the power of snRNA-seq to plant research and provides an unprecedented spatiotemporal gene expression atlas of heterogeneous shoot cells.

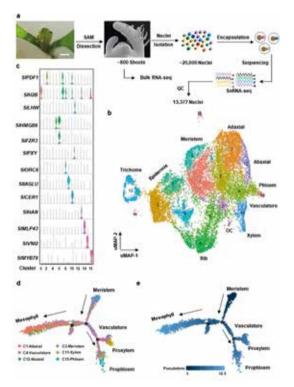




Figure: A cell atlas of tomato shoot apex by snRNA-seq identified the cell heterogeneity and mapped the developmental trajectory of mesophyll and vasculature cells. a. Procedure of snRNA-seq. b. Visualization of tomato shoot apex cell clusters by uMAP algorithm. OC: organization center. c. Marker gene expression pattern in each cluster. d and e. Developmental trajectory of mesophyll and vasculature cells highlighting clusters (d) and pseudotime (e).

## **Regulation of Plant Immunity and Stem Cell Fate**

Chuanyou Li, Principal Investigator, Ph.D. (1999, Institute of Genetics, CAS, China).

Research Interests: Tomato as a model to investigate systemin/jasmonate-signaled systemic plant immunity and the molecular mechanism of tomato quality formation. Tomato and *Arabidopsis* as models to investigate the mechanisms of stem cell-determinant root growth plasticity in plants.

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## SEUSS integrates transcriptional and epigenetic control of root stem cell organizer specification

Higher plants are sessile and respond to environmental challenges by adjusting the process of organogenesis and development. Compared with animals, plants show super plasticity which mainly depends on their stem cells. The WOX5 gene is specifically expressed in root stem cell organizer cells and plays a central role in QC specification. However, the molecular mechanism regulating WOX5 expression and QC specification remains largely unknown. Our recent research have revealed that the SEUSS (SEU) protein, homologous to the animal LIMdomain binding proteins, assembles a functional transcription complex that regulates QC specification. The seu-3 seedlings displayed markedly reduced WOX5 expression compared with the WT. Similar to the wox5-1 mutant, seu-3 seedlings showed supernumerary cells with nonstereotyped shapes in the QC position. Further study showing that SEU acts in the SHR/ SCR pathway to regulate WOX5 expression and QC specification. SEU is physically recruited to the WOX5 promoter by the master transcription factor SCR. And SEU physically recruits SDG4 to promote WOX5 expression by regulating H3K4me3 modification in the WOX5 promoter. In summary, our results suggest that SEU acts as a multi-adaptor protein that integrates the actions of genetic and epigenetic regulators to orchestrate the formation of the SCR-SEU-SDG4 transcriptional complex, which regulates WOX5 expression during QC specification.

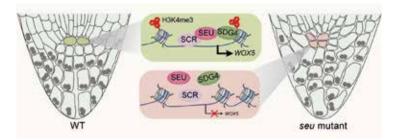


Figure: Schematic representation of the role of SEU in SCR-mediated activation of WOX5 expression.



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- Wang, L., Xu, Q., Yu, H., Ma, H., Li, X., Yang, J., Chu, J., Xie, Q., Wang, Y., Smith, S.M., Li, J., Xiong, G., and Wang, B. (2020). Strigolactone and karrikin signaling pathways elicit ubiquitination and proteolysis of SMXL2 to regulate hypocotyl elongation in *Arabidopsis* thaliana. Plant Cell, 32: 2251-2270.
- Zhao, Y., Yu, H., Zhou, J.M., Smith, S.M., and Li, J. (2020). Malate circulation: linking chloroplast metabolism to mitochondrial ROS. Trends in Plant Sciences, 25 (5): 446-454.
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## Developmental Biology and Metabolic Pathways of Higher Plants

**Jiayang Li**, Principal Investigator, Member of Chinese Academy of Sciences, Fellow of TWAS, Member of the German National Academy of Sciences, Foreign Associate of USA National Academy of Sciences and Foreign Fellow of the Royal Society of London for Improving Natural Knowledge.

Research Interests: Molecular genetics of plant development and metabolism, focusing on the understanding of molecular bases of plant architecture features and key metabolic pathways that define major agronomic traits and devoting on the breeding of high-yield, superiorquality, stress-resistant, and resource-use-efficiency new varieties of rice and natural rubber crops.

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#### Roles of dual-functional repressor proteins in strigolactone signaling

Strigolactones (SLs) are carotenoid-derived plant hormones that have fundamental effects on shoot branching, leaf development, plant height, anthocyanin accumulation, root architecture, and adaptation to drought and phosphate starvation. SLs also function as rhizosphere signals important for interactions between host plants and symbiotic arbuscular mycorrhizal (AM) fungi or rootparasitic plants. Our group has identified key components in SL biosynthesis and signaling, and proposed the SL signaling model featured by the SL-triggered ubiquitination and degradation of D53. The limited knowledge of SL responsive genes has seriously hampered the understanding of SL signaling. We have synthesized SL analogs GR24<sup>400</sup>, GR24<sup>505</sup> and rac-GR24, and compared their effects in stimulating SL signaling. GR244DO specifically stimulates SL signaling in a D14-dependent manner, and the efficiency of GR24<sup>4D0</sup> is higher than that of and rac-GR24. Through RNA-SEQ analysis, we identified 401 SL responsive genes after GR24<sup>±</sup> GR24<sup>400</sup> treatment for 2 h and 4 h. Genetic analyses further indicated thatSLs repress shoot branching through transcriptional activation of BRC1, which further promotes expression of HB40 and ABA accumulation in axillary buds. TCP1 is important for leaf elongation promoted by SL signaling. Furthermore, SLs promote anthocyanin accumulation through transcriptional activation of PAP1 and its homologues PAP2, MYB113 and MYB114. These findings demonstrate that SLs control diverse aspects of plant development through distinct SL responsive genes. We further identified 729 SMXL6-target genes through ChIP-SEQ analysis and found that SMXL6 failed to directly bind the BRC1 promoter in EMSA assays, indicating that SMXL6 may work with unknown TFs to inhibit downstream gene expression. Notably, we found that SMXL6, SMXL7 and SMXL8 directly bind each other's promoters in vitro. The ATAACAA motif in SMXL7 promoter is required for the direct binding and transcriptional

repression of SMXL6 on the *SMXL7* promoter. These findings demonstrate that SMXL6, 7, 8 could directly bind DNA and function as self-regulating transcription factors, revealing a novel mechanism distinguished from the classical model that repressor proteins in IAA, JA, GA, and SL signaling need transcription factors to repress gene expression.

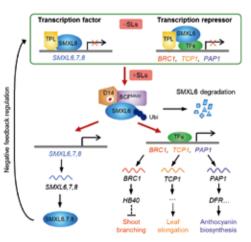


Figure: Model of transcriptional regulation by SMXL6 in SL signaling. SMXL6 directly binds promoters of *SMXL6*,7,8 and functions as an auto-regulated transcription factor (TF). Meanwhile, SMXL6 can also form a complex with unknown TFs that are expected to recognize and bind to the promoters of BRC1, TCP1 and *PAP1*, thus regulating shoot branching, leaf elongation and anthocyanin biosynthesis in *Arabidopsis thaliana*.



### Single Cell Omics and Plant Molecular Genetics

Xiang Li, Principal Investigator, Ph.D. (2017, Huazhong Agricultural University, China).

Research Interests: 1) Developing single cell omic technology, 2) to answer key questions in plant reproduction, and 3) to investigate the genetic and molecular mechanism of cell fate decision in photosynthesis.

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#### Dissection of haploid induction mechanism

In maize, following pollination with Stock6 mutants, the stock6-derived chromatin of zygotes would be eliminated during embryogenesis, resulting in haploid individuals. The application of Stock6 mutants accelerates the production of inbred lines, and enhances crop breeding. However, the current haploid induction rate is only ~13%. The further improvement may depend on the molecular mechanism dissection of this phenomenon. One gene controlling haploid induction was cloned, encoding a maize spermspecific phospholipase A (ZmPLA1). Here, we applied multiple omic analysis on *zmpla1* pollen and anthers, integratingtranscriptomes, metabolomes, protein abundance-based proteomes as well as modifications. The results suggested that a reactive oxygen species (ROS) burst plays a critical role in haploid induction (Figure a). This was validated by the results from pollens treated with ROS-associated reagents. This study suggests a conserved role of ROS in abnormal reproduction of plant and animal (Figure b), provides a potential route to accelerate crop breeding.

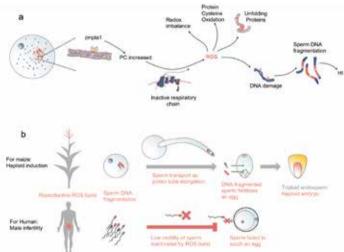
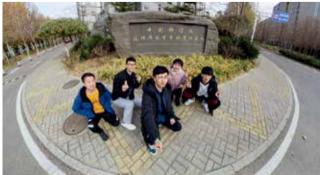


Figure: Hypothesis for the whole HI processing. (a) An entire pathway of *zmpla1*-triggered HI processing is summarized here. (b) The effects of reproductive ROS burst in plants and mammals are described causing HI and male infertility, respectively.





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### Genomic Big Data Analysis

**Chengzhi Liang**, Principal Investigator, Ph.D. (1995, Institute of Genetics, CAS). M.Math. (2001, Computer Science, University of Waterloo, Canada).

Research Interests: (1) Genome Assembly and Annotation. Genome was assembled with data from the lastest sequencing technology and annotated based on gene expression data. (2) Population Genomic Analysis. The favorable alleles and genetic basis of environmental adaptability in rice breeding were analyzed by population genomic analysis on a large collection of cultivated rice. (3) Database Construction. Construct databases for genotypic and phenotypic data management and integration.

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#### Analysis of genetic architecture of agronomic traits, introgression and differentiation in Chinese cultivated rice

Several rice populations have been re-sequenced in the past decade, but less focus on current Chinese rice cultivars which carry many agronomically favorable alleles. In this study, we analyzed large-scale genomic and phenomic datasets for a collection of 1,275 rice accessions. With GWAS, we identified a total of 143 significant association loci, including three newly identified candidate genes or alleles that control heading date or amylose content. Furthermore, we inferred inter-subpopulation introgressions in these rice accessions combined with 3K-RG. We found that alien introgressions are present extensively in the rice population with many of them containing functional sequence variants, quantitative trait loci (QTL) or genes controlling flowering, grain and resistance traits. It was found many of the differentiated

regions between subgroups within a subpopulation contained agronomically important loci, providing new candidates for studying local adaptation or heterosis. Tracing these footprints allows us to better understand the genetic exchange or differentiation underlying agronomic traits in modern Chinese rice cultivars.

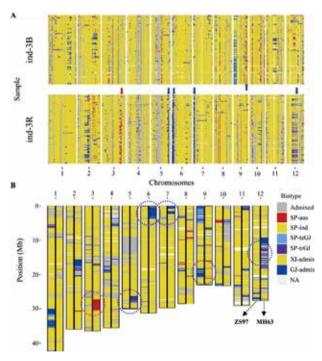




Figure: Differentiated distribution of alien introgressions among ind-3B and ind-3R subgroups. A: Stacked plot of genomewide subpopulation inference results of samples from ind-3B and ind-3R subgroups. B: Comparison of genome-wide subpopulation inference of Zhenshan97B (left) and Minghui63 (right).

### **Rice Genome Design and Molecular Breeding**

Shaoyang Lin, Principal Investigator, Ph.D. (1993, Chiba University, Japan).

Research Interests: We focus on building a new model of genomic updating and design breeding for crops. We replace the 'bug' loci of rice variety Kongyu131 with other favorable alleles by the results of QTL analysis and re-sequencing. Our projects involve rice blast resistance, eating quality, high yield, regional adaptation and other agronomic traits.

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## Molecular breeding for rice single and multi-traits without phenotype selection

Achieving precise and targeted breeding is the primary goal of molecular breeding. Inspired by constant updates of App, our group keeps finding 'bugs' of the genome of elite rice variety Kongyu131 and updating it to a series of SPSLs (Single Point Substitution Lines), which are only replaced by a small chromosome segment carrying a favorable trait allele from donor parents. And new elite varieties can be designed and developed by pyramiding different SPSLs that significantly improve complex agronomic traits.

Recently, we obtain two new SPSLs by replacing favorable alleles at rice quality locus Wx and disease-resistant locus Stvb-i of Kongyu131, and two pyramided lines Ghd7/Stvb-i, GS3/  $Wx^{mp}$  by separately combing SPSL(+Ghd7) and SPSL(+GS3) obtained before. Waxy (Wx) and Stvb-i are the significant genes that control Amylose Content (AC) and rice stripe disease resistance, respectively. The japonica variety Kongyu131 shows 18% amylose content with a  $Wx^{b}$  allele and is highly susceptible to stripe virus, while a donor parent GKMP shows 10% low amylose content with Wx<sup>mp</sup> allele and contains disease-resistant Stvb-i allele derived from variety Modan. Thus, we introduce the favorable allele *Wx<sup>mp</sup>*, *Stvb-i*, from the donor GKMP into recurrent parent Kongyu131 by backcross breeding(Fig. B). Using 5 SNP markers designed for the region within and around the targeted locus, the introgressed chromosome segments in two SPSLs are shortened to nearly 1223 Kb (+ $Wx^{mp}$ ) and 1506 Kb(+Stvb-i), minimizing the linkage drag effectively. Finally, the genotype of all SNP markers suggests that the recovery ratio of the recurrent parent genome(RRPG) of the SPSLs are 99.83% (+Wx<sup>mp</sup>) and 99.74% (+Stvb-i). The results of field trial show that the SPSL $(+Wx^{mp})$  has 11% amylose content as GKMP and exhibits no statistical difference in yield and other agronomic appearance of Kongyu131(Fig. C-D). The SPSL(+Stvb-i) exhibits significant resistance to stripe disease with a lower disease index after virus infection but shows a 4% yield decrease per plant. Moreover, we successfully develop two combined lines Ghd7/Stvb-i,  $GS3/Wx^{mp}$  by separately pyramiding SPSLs, to expand the suitable cultivated area of SPSL(+Stvb-i) to Beijing and increase the grain length of SPSL(+ $Wx^{mp}$ ). The agronomic appearance of the pyramided lines has no significant difference to their SPSLs(Fig. A-D). With our studies above, we realize selecting SPSLs and pyramided lines based only on genotype information from genome-wide SNP markers. It eliminates the breeders' dependence on phenotype selection in traditional breeding and marker-assisted selection strategy,

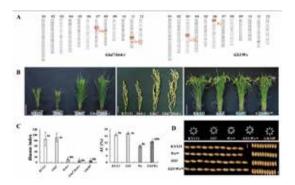


Figure: Graphical genotype and phenotype of the pyramided lines *Ghd7/Stvb-i* and *GS3/Wx<sup>mp</sup>*. (A) Graphical genotype of the lines *Ghd7/Stvb-i*, *GS3/Wx<sup>mp</sup>*. (B) Plant architecture and main panicle. (C) Rice stripe disease resistance performance of line *Ghd7/Stvb-i* and Amylose Content of line *GS3/Wx<sup>mp</sup>*. (D) Milled rice's clarity and grain size of line *GS3/Wx<sup>mp</sup>*.

which is also proved workable by final traits evaluation of the SPSLs

and their pyramided lines.





## The Molecular Mechanism of Symbiotic Nitrogen Fixation in Legume

**Bo Ren**, Principal Investigator, Ph.D. (2010, Institute of Genetics and Developmental Biology).

Research Interests: Nitrogen is one of essential nutrient elements for plant development and crop yields. Our lab mainly focus on dissecting the mechanism of soybean nodulation, aimed to improve biological nitrogen fixation and nitrogen use efficiency in legume.

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#### GmGARP1 regulates soybean nodulation

Nitrogen is one of essential nutrient elements for plant development and crop yields. In agricultural system, the main nitrogen source is chemical fertilizers, which increase both economic and environmental costs. Therefore, it is very important to develop and utilize some other nitrogen sources, such as biological nitrogen fixation (BNF), in agriculture for sustainable development. Cytokinin signaling is essential for legume nodulation, but the molecular mechanism remains elusive. We treated rhizobiainoculated soybean roots with exogenous cytokinin and performed time-course RNA-seg analysis to find key differential expression genes during nodulation. We found that many phytohormone-related transcription factors dramatically changed in the early stage of rhizobia infection. Among them, overexpression one of a plant-specific GARP transcription factor, GmGARP1, causes less nodule number in hairy roots. Marker genes both in nodulation pathway and cytokinin signal pathway significant changed in hairy roots which overexpressed different versions of GmGARP1 transcripts, indicating that GmGARP1 directly effects nodulation and cytokinin pathway. Further analysis of the nucleotide diversity ( $\pi$  and  $F_{st}$ ) in wild (Soja) and improved cultivar (Cultivar) soybeans showed that GmGARP1 is located in a ~400-kb selective sweep region, implying that this locus had been artificially selected during soybean domestication.

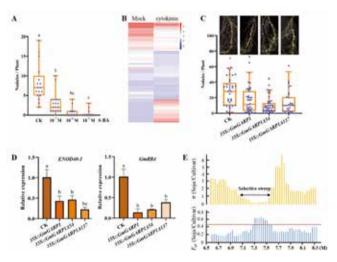




Figure: *GmGARP1* regulates soybean nodulation. (A) Exogenous cytokinin inhibits soybean nodulation; (B) Different genes expression in the absence and presence of exogenous cytokinin during nodulation; (C) Overexpression different versions of *GmGARP1* cause nodule number decrease in transgenic hairy roots; (D) Expression levels of *ENOD40-1* and *GmRR4* in transgenic hairy roots; (E) Nucleotide diversity ( $\pi$  and *F<sub>st</sub>*) in wild (*Soja*) and improved cultivar (Cultivar) soybeans. The red dash line denotes the threshold of the whole-genome level.

### **Plant Functional Metabolomics**

**Guodong Wang**, Principal Investigator, Ph.D. (2003, Shanghai Institute of Plant Physiology and Ecology, CAS, China).

Research Interests: Plants collectively produce huge diverse specialized metabolites, many of which have beneficial effect to human health. The overall interest within my laboratory is to study how plant synthesizes these healthy specialized metabolites, the key enzymes potentially involved in these biosynthesis pathways and the mechanism of the enzymatic reactions.

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#### Evolution and structural elucidation of sester-TPS in plants

Plants synthesize many terpenoids (including phytohormones) with diverse ecological functions to help them survive in their local environments. Sesterterpene synthase (sester-TPS, C25), a branch of class I TPSs, was recently identified in Brassicaceae plants.We previously demonstrated that the structural diversity of plant terpenoids is largely determined by two consecutive, but separate, enzymes: short-chain prenyltransferases (SC-PTs) and terpene synthases (TPSs). Here, we futher identified three nonclustered functional sester-TPSs (AtTPS06, AtTPS22 and AtTPS29) in Arabidopsis thaliana. AtTPS06 utilizes a type-B cyclization mechanism, while most other sester-TPSs produce various sesterterpene backbones via a type-A cyclization mechanism. We then determined the crystal structure of the AtTPS18-FSPP complex to explore the cyclization mechanism of plant sester-TPSs. We used the structural comparisons and site-mutagenesis to further elucidate the mechanism: 1) mainly due to outward shift of helix-G, plant sester-TPSs have a larger catalytic pocket than do mono-, sesqui-, and di-TPSs to accommodate GFPP; 2) more aromatic residues (five or six) in the catalytic pocket of type-A sesterTPSs than that of classic TPSs (two or three) is an unique feature to type-A sesterTPSs, which also determine whehter the type-A or type-B cyclization mechanism is active; 3) the other residues responsible for product fidelity were determined by interconversion of AtTPS18 and its close homologs. Altogether, this study deepens our understanding of the catalytic mechanism of plant sester-TPS, which ultimately enables rational engineering of sesterterpenoids for future applications.



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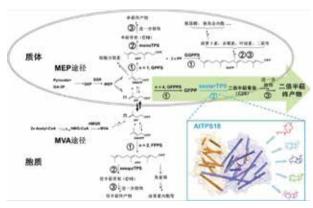


Figure: Terpenoid network in plants and the structural elucidation of sesterterpene synthase (AtTPS18, inset)





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### Molecular Basis of Shoot Branching

**Yonghong Wang**, Principal Investigator, Ph.D. (2004, Institute of Genetics and Developmental Biology CAS, China).

Research Interests: Using *Arabidopsis* and rice as model systems, we mainly focus on dissecting the molecular networks involved in formation and development of the shoot axillary meristems, including the initiation, dormancy and activation of axillary buds and the determination of axillary shoot angle, trying to identify genes applicable to improving crop yields.

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## Functional analysis of *INCREASED GRAIN NUMBER 1* in regulating rice grain number

Grain number per panicle (GNP) is an important agronomic trait that contributes to rice grain yield. Despite its importance in rice breeding, the molecular mechanism underlying GNP regulation remains largely unknown. In this study, we identified a dominant mutant, *increased grain number 1-Dominant (ign1-D)*, which exhibited significantly increased grain number and yield. Genetic variation in the CArG box-containing inverted repeat (IR) sequence of *IGN1* promoter alters its expression level and contributes to GNP variation among rice varieties. The IR sequence regulates *IGN1* expression by affecting the direct binding of an OsMADS to the CArG box within the IR sequence. The divergent *pIGN1*<sup>9311</sup> and *pIGN1*<sup>NBP</sup> alleles originated from different *O.rufipogon* accessions, and were independently inherited into the *japonica* and *indica* subspecies respectively during domestication. Our findings not only provide new insight into the molecular basis of rice grain number, but also provide useful locus for the genetic improvement of rice grain yield.

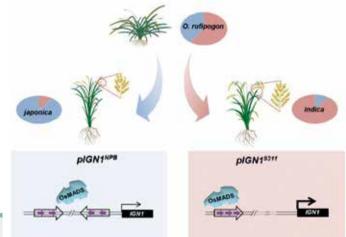


Figure: A proposed working model of *IGN1* in regulation of grain number per panicle and its divergent during domestication. Blue indicates  $p/GN1^{NBP}$  with low expression activity, and red indicates  $p/GN1^{S311}$  with high expression activity. The arrows with black line indicate the IR sequences. The purple arrows indicate the CArG motifs.



### Ubiquitination Modification and Plant Stress Signal Transduction

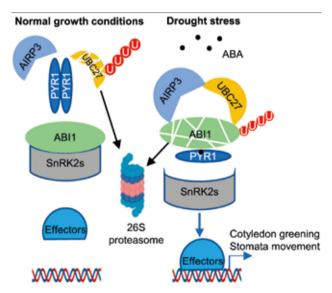
Qi Xie, Principal Investigator, Ph.D. (1994, Universidad de Madrid, Spain).

Research Interests: Uncover the functional mechanism on ubiquitination modification of proteins in response to biotic and abiotic stresses with the biochemical and genetic approaches in the model plant *Arabidopsis* Breed crops (mainly focus on sorghum and maize) with high biomass and stress tolerance through the molecular genetics, bioinformatics and traditional breeding methods.

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#### A novel regulation mechanism of ABA co-receptor ABI1 by the E2-E3 complex UBC27-AIRP3

The phytohormone Abscisic Acid (ABA) is one of the important regulators in plant abiotic stress adaptation. The activity control of the co-receptor PP2C proteins like ABI1 is the central hub of ABA signaling transduction, as ABI1 is competitively bond by ABA-PYR1/ PYL/RCAR receptor complex to release SnRK2s, which are inhibited by ABI1 under normal conditions. A specific E2 enzyme UBC27 was identified, which positively regulates plant drought tolerance and ABA response. By IP/MS assay, ABI1 and a RING-type E3 ligase AIRP3 were found to be interaction proteins of UBC27. Biochemical assays show that UBC27 interacts with and promotes the degradation of ABI1, and activates the E3 activity of AIRP3, which works as the E3 ligase for ABI1. Furthermore, ABI1 acts epistasis of UBC27 and AIRP3 and functions of AIRP3 is UBC27-dependent. In addition, ABA treatment induces the expression of *UBC27*, inhibits the degradation of UBC27, and enhances the interaction between UBC27 and ABI1. The above results uncovered a novel E2-E3 complex in ABI1 degradation. These findings not

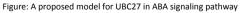


only bring new insights into our understanding of the mechanism of ubiquitination-mediated regulation of ABA signaling, but also are helpful for molecular breeding to improve water stress tolerance in crops.



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## Plant Cell Communications and Biological Robustness

**Cao Xu**, Principal Investigator, Ph.D. (2012, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences), Postdoctoral Fellow (2013-2017, Cold Spring Harbor Laboratory).

Research Interests: Small signaling peptides and plant cell-to-cell communications. Plant robustness and rational design of smart crops

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## Post-translational modification of small peptide and phase separation of transcription factor determine shoot apical stem cell fate

The fate decision of the shoot apical stem cells relies on the balance of cell proliferation and differentiation. We found that this balance is maintained by small peptide signaling regulated stem cell proliferation and phase separation mediated transcriptional control. The arabinosyltransferase FASCIATED INFLORESCENCE (FIN) catalyzes transferring an I-arabinose to small peptide SICLV3, which enables its function in controlling inflorescence architecture and fruit size. TERMINATING FLOWERING (TMF), an *Arabidopsis* LSH1 and *Oryza* G1 (ALOG) family transcription factor, senses developmentally produced  $H_2O_2$  to form transcriptional condensates via protein phase separation. The resulting TMF transcriptional condensates repress floral identity gene expression to time the maturation of shoot apical meristem and flowering transition. Our findings uncover a chemical biology layer in the control of plant stem cell fate decision achieved by integration of small peptide modification with protein phase separation.

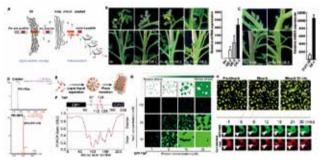


Figure: (A) Post-translational processing and modifications of small peptide. (B-C) Phenotypes of *SICLV3* overexpression transgenics in *fin* mutant background (left panel). qRT-PCR assay showing up-regulation of *SICLV3* in different transgenic lines (right panel). (D) *In vitro* modification assay showing FIN directly catalyzes the arabinosylation of SICLV3. (E) Schematics illustrating the process of protein phase separation. (F) Graphs indicating intrinsic disorder regions and cysteine residues in TMF proteins. (G) Phase diagram showing droplet formation under various concentration combinations of GFP-TMF protein and NaCl. Scale bar, 5 µm. (H) Fluorescence recovery after photobleaching (FRAP) assay showing dynamic property of biomolecular condensates formed by GFP-TMF and Cy-3 labelled DNA fragments from *AN* promoter. White arrowheads indicate bleached area in droplets. Scale bars, 20 µm (upper panel), 1 µm (lower panel).



## **Rice Genetics and Molecular Breeding**

Shanguo Yao, Principal Investigator, Ph.D. (2004, Ehime University, Japan).

Research Interests: Rational design of new rice varieties in northeast China. Molecular mechanism of rice plants in response to extreme temperature stress.

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#### ZK613 was approved for extension in Heilongjiang province

DHX is the most popular variety in China for the good eating quality and better appearance quality. However, this variety also faces some problems, such as weak lodging resistance, poor cold tolerance, and decreasing blast resistance. By whole-genome resequencing, we analyzed in detail the allelic genotypes of agronomically important traits in DHX, and identified favorable alleles of the target traits. Rational combination of these alleles resulted in a new variety of ZK613 with improved comprehensive traits (Figure 2), and the variety was approved for extension in the first accumulative temperature zoon of Heilongjiang province in July 2020.



Figure: Molecular design of the new rice variety ZK613.

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## **Ethylene Signaling and Seed Trait Regulation**

Jinsong Zhang, Principal Investigator, Ph.D. (1991, Peking University, China).

Research Interests: We identified novel components and mechanisms in rice ethylene signaling, and discovered molecular networks involved in regualtion of seed size and oil accumulation in soybean. Genes in stress tolerance were also investigated.

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## Histidine kinase MHZ1/OsHK1 interacts with ethylene receptors to regulate root growth in rice

In semiaquatic monocot rice, while ethylene plays essential roles in its adaptive responses to the hypoxia conditions and regulates multiple agronomic traits, its signaling mechanism is largely unclear. Using an efficient system, we have screened a set of rice ethylene response mutants which were named as mao huzi (mhz). A root-specific ethylene insensitive mutant mhz1 was characterized. MHZ1 encodes a histidine kinase. Through biochemical experiments, we found that MHZ1 has autophosphorylation ability on a conserved histidine residue and transfers the phosphoryl groups to OsRR21 via OsAHPs, which is called the multistep phosphorelay. This MHZ1-mediated phosphorelay is essential for ethylene-regulated root growth. Further, we found that ethylene receptor OsERS2 interacts with MHZ1 to suppress its kinase activity, and subsequently inhibits the MHZ1-mediated phosphorelay. This OsERS2-MHZ1-phosphorelay pathway functions partially independent of the conserved OsEIN2-OsEIL1 pathway to regulate ethylene response in rice roots. This study revealed a novel ethylene signaling mechanism in rice roots, of which MHZ1 interacts with the ethylene receptors to regulate ethylene response (Figure). The discovery suggests that rice may have adopted a more complicated ethylene signaling mechanism than Arabidopsis to adapt to the watersaturated environment.

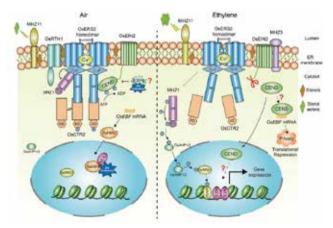




Figure: Histidine kinase MHZ1 participates in rice ethylene signaling. In air, the ethylene receptor OsERS2 interacts with MHZ1 and inhibits MHZ1 kinase activity through the GAF domain. Ethylene binds to the receptor, releasing its inhibition on MHZ1, activating MHZ1-mediated phosphorelay for root ethylene responses. The MHZ1-mediated pathway and the OsEIN2-mediated pathway act downstream of ethylene receptors to collaborately regulate root ethylene responses in rice.

## **Molecular Basis of Plant-Microbe Interaction**

**Jianmin Zhou**, Principal Investigator, Ph.D. (1994, Purdue University, USA), Assistant Professor and Associate Professor (1997-2004, Kansas State University, USA), Associate Investigator and Investigator (2004-2012, National Institute of Biological Sciences, China).

Research Interests: Plants use numerous immune receptors, pattern-recognition receptors (PRRs) and NB-LRR proteins, to detect invading pathogens and resistance against pathogen infection. Our work aims at understanding how plant immune receptors work at the molecular level and how downstream signaling is activated.

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## An *Arabidopsis* secondary metabolite inhibits bacterial virulence for defense

Plant deploy thousands of secondary metabolites that inhibit microbes and protect plants from infection. However, these secondary metabolites likely inhibit both pathogenic bacteria and beneficial microbes, which is not desirable to plants.Many Gramnegative pathogenic bacteria secretes effector proteins into plant cells through type III secretion system (T3SS), a process essential for pathogenicity. T3SS in Pseudomonas *syringae* (*Pst*) is encoded by hrp (*hypersensitive response and pathogenicity*) genes, which are induced by an alternative sigma factor HrpL. Transcription factors HrpR and HrpS form a hetero-hexamer and activate the expression of *hrpL*. In this study, we identified sulforaphane (4-methylsulfinylbutyl isothiocyanate), a natural product derived from aliphatic glucosinolates in *Arabidopsis*, covalently modifies the Cys 209 of bacterial HrpS to decrease the transcription of T3SS genes and attenuate the virulence of pathogenic bacteria. But SFN does not affect *P. syringae* lacking a type III secretion system or leaf-associated bacterial microbiota. This study demonstrated that the plant metabolism SFN directly inhibits the virulence rather than broad killing of bacteria and revealed the mechanism of its action.



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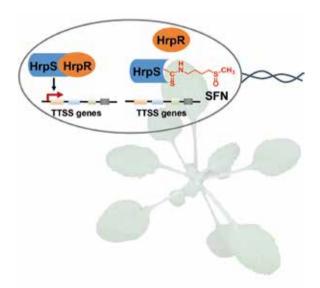


Figure: Model for the SFN-mediated repression of TTSS gene expression.





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### Molecular Genetics and Cell Wall Biology

Yihua Zhou, Principal Investigator, Ph.D. (1998, Institute of Genetics, CAS, China).

Research Interests: Dissecting mechanisms for secondary wall biosynthesis and functions on plant growth, which may be helpful for improving agronomic traits, e.g. lodge resistance in crops.

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## Identify a molecular connection between biomass production and nitrogen utilization efficiency in rice

Nitrogen (N) fertilizer is crucial for plant growth and crop yield, and improving crop N-use efficiency (NUE) is benefit for decreasing fertilizer usage and sustainable agriculture. Plants uptake N to produce nucleotides, proteins, and numerous cellular components through the N metabolism to facilitate carbon dioxide (CO<sub>2</sub>) fixation for carbohydrate generation, referred to as carbon (C) metabolism. Most of the CO<sub>2</sub> assimilated products are converted into cell wall polysaccharides to build the plants themselves, intuitively showing as plant growth; vice versa the robustness in plant growth can promote N uptake and utilization. Therefore, nitrogen-mediated plant growth is highly coordinated with plant cell wall biogenesis. However, the clear-cut molecular links between cell wall production and nitrogen use remain unclear. Here, we report that differences in nitrogen utilization efficiency and cellulose biogenesis between indica and japonica rice subspecies can be explained by a variation at MYB61, which transcription is controlled by GRF4, an integrative regulator of N metabolism. QTL analysis in a set of introgression lines generated from indica variety 9311 and japonica variety Nipponbare (NP) identified a QTL coregulating N-mediated leaf area changes and cellulose content. Gene cloning indicated this locus to transcriptional factor MYB61. The variation at MYB61 promoter, which is in presence of a helitron transposon insertion or not, confers the divergence in NUE and cellulose content between NP and 9311. MYB61 transcription is induced by low N availability and regulated by GRF4, which is pivotal to control cellulose biogenesis. The helitron transposon is only present in all examined japonica varieties, suggesting that MYB61 likely proceeded this selection while indica-japonica divergence. Furthermore, the indica allele of MYB61 confers the japonica introgression lines with robustness in simultaneously governing nitrogen use and biomass production, leading to improved grain yield, especially at limited N supply. This finding in identifying the GRF4-MYB61 regulatory module as a link of C&N metabolism uncovers the mechanism for C&N coordination and provides a solution to

maintenance of grain yield while reducing agricultural N use in crop production.

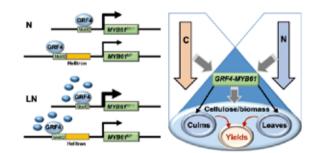


Figure: The working model of rice GRF4-MYB61 regulatory module that coordinates carbon & nitrogen metabolism and improves nitrogen use efficiency and grain yield. Under limited nitrogen availability, accumulative GRF4 can bind the promoter of *MYB61* without the helitron transponson insertion and activate its transcription more efficiently, which promotes cellulosic biomass production and nitrogen utilization, resulting in improved grain yield. N, Normal nitrogen; LN, Low nitrogen.



## Plant Functional Genomics and Biotechnologies

Zhen Zhu, Principal Investigator, Ph.D. (1988, Institute of Genetics, CAS, China).

Research Interests: Study on no-homologous gene inducing post-transcriptional gene silencing; Study the heterosis mechanism by large-scale comparative transcriptome and molecular biology analysis of the super-hybrid rice; Development of transgenic insect resistant rice, including the Lepidoptera pests resistant rice, and the novel anti-Brown planthopper rice.

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## OsJAZ13 negatively regulates jasmonate signaling and activates hypersensitive cell death response in rice

Jasmonate ZIM-domain (JAZ) proteins belong to the subgroup of TIFY family and act as key regulators of jasmonate (JA) responses in plants. To date, only a few JAZ proteins have been characterized in rice. Here, we report the identification and function of rice OsJAZ13 gene. The gene encodes three different splice variants: OsJAZ13a, OsJAZ13b, and OsJAZ13c. The expression of OsJAZ13 was mainly activated in vegetative tissues and transiently responded to JA and ethylene. Subcellular localization analysis indicated OsJAZ13a is a nuclear protein. Yeast two-hybrid assays revealed OsJAZ13a directly interacts with OsMYC2, and also with OsCOl1, in a COR-dependent manner. Furthermore, OsJAZ13a recruited a general co-repressor OsTPL via an adaptor protein OsNINJA. Remarkably, overexpression of OsJAZ13a resulted in the attenuation of root by methyl JA. Furthermore, OsJAZ13a-overexpressing plants developed lesion mimics in the sheath after approximately 30-45 days of growth. Tillers with necrosis died a few days later. Gene-expression analysis suggested the role of OsJAZ13 in modulating the expression of JA/ethylene response-related genes to regulate growth and activate hypersensitive cell death. Taken together, this study indicates a novel regulatory mechanism in rice and provides the basis for elucidating the function of OsJAZ13 in signal transduction and cell death in plants.



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- Feng, X., Zhang, L., Wei, X., Zhou, Y., Dai, Y., and Zhu, Z. (2020). OsJAZ13 negatively regulates jasmonate signaling and activates hypersensitive cell death response in rice. International Journal of Molecular Sciences, 21 (12): 4379.

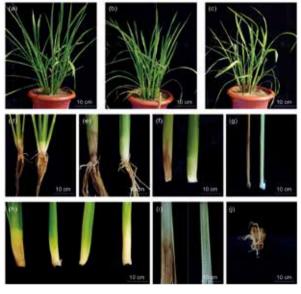
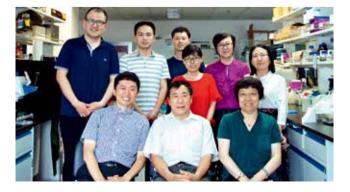


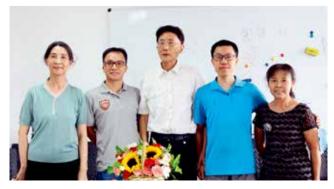
Figure: Phenotype analysis of *OsJAZ13a*-overexpression plants under paddy fields. *OsJAZ13a*-overexpression lines and the wild type were transplanted to paddy fields about 30-45 days till plants developed to the tillering stage. (a) Wild type, (b) One tiller died, (c) Two tiller died, (d)-(g) Comparation of the sheath between dead and normal tillers, (h) Differential level of the watersoaked tiller, (i) the comparison of leaves, (j) the cross-section of a dead tiller.





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- He, M., Su, J., Xu, Y., Chen, J., Chern, M., Lei, M., Qi, T., Wang, Z., Ryder, L.S., Tang, B., Oses-Ruiz, M., Zhu, K., Cao, Y., Yan, X., Eisermann, I., Luo, Y., Li, W., Wang, J., Yin, J., Lam, S.M., Peng, G., Sun, X., Zhu, X., Ma, B., Wang, J., Liu, J., Qing, H., Song, L., Wang, L., Hou, Q., Qin, P., Li, Y., Fan, J., Li, D., Wang, Y., Wang, X., Jiang, L., Shui, G., Xia, Y., Gong, G., Huang, F., Wang, W., Wu, X., Li, P., Zhu, L., Li, S., Talbot, N.J., and Chen, X. (2020). Discovery of broad-spectrum fungicides that block septindependent infection processes of pathogenic fungi. Nature Microbiology, DOI: 10.1038/ s41564-020-00790-y.
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- Jiang, G., Liu, D., Yin, D., Zhou, Z., Shi, Y., Li, C., Zhu, L., and Zhai, W. (2020). A rice NBS-ARC gene conferring quantitative resistance to bacterial blight is regulated by a pathogen effector-inducible miRNA. Molecular Plant, DOI: 10.1016/j.molp.2020.09.015.
- Jiang, G.H., Yin, D.D., Shi, Y., Zhou, Z.Z., Li, C.R., Liu, P.C., Jia, Y.F., Wang, Y.Y., Liu, Z.Z., Yu, M.X., Wu, X.H., Zhai, W.Z., and Zhu, L.H. (2020). OsNPR3.3-dependent salicylic acid signaling is involved in recessive gene xa5-mediated immunity to rice bacterial blight. Scientific Reports, 10 (1).
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### Molecular Mechanisms on Plant Disease Resistance

Lihuang Zhu, Principal Investigator Emeritus.

Research Interests:Molecular mechanisms on rice heterosis. Identification and functional analysis of key molecules in plant disease resistance

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## OsNPR3.3-dependent salicylic acid signaling is involved in recessive gene xa5-mediated immunity to rice bacterial blight

Salicylic acid (SA) is a key natural component that mediates local and systemic resistance to pathogens in many dicotyledonous species. However, its function is controversial in disease resistance in rice plants. Here, we show that the SA signaling is involved in both pathogen-associated-molecular-patterns triggered immunity (PTI) and effector triggered immunity (ETI) to *Xanthomonas oryzae* pv. *Oryzae* (*Xoo*) mediated by the recessive gene xa5, in which OsNPR3.3 plays an important role through interacting with TGAL11. Rice plants containing homozygous xa5 gene respond positively to exogenous SA, and their endogenous SA levels are also especially induced upon infection by the *Xoo* strain, PXO86. Depletion of endogenous SA can significantly attenuate plant resistance to PXO86, even to 86ΔHrpXG (mutant PXO86 with a damaged type III secretion system). These results indicated that SA plays an important role in disease resistance in rice plants, which can be clouded by high levels of endogenous SA and the use of particular rice varieties.

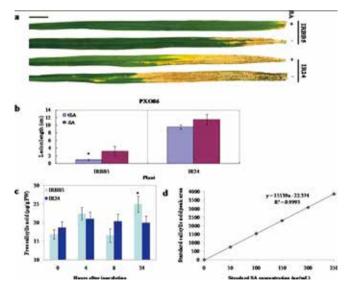


Figure: Influence of salicylic acid on bacterial blight resistance in rice plants to *Xoo* strain PXO86 and contents of SA in rice. (a) Resistance phenotype of rice plants at the tillering stage 14 dai. (b) Lesion length on leaves of rice plants at 14 dai. (c) Free SA levels in IRBB5 and IR24 inoculated with Xoo strain PXO86. (d) Standard curve of SA. The plants used in a and b were pretreated with 2 mM exogenous SA or H<sub>2</sub>O for one day, and then inoculated with PXO86. IRBB5 is the rice variety containing the homozygous recessive resistance gene xa5; IR24 is the susceptible near isogenic line of IRBB5.

## **Plant Molecular Genetics**

**Jianru Zuo**, Principal Investigator. Ph.D. (1994, Miami University, USA), Postdoctoral fellow (1995-2001, Rockefeller University, USA).

Research Interests: Nitric Oxide Signaling and Nitrogen Nutrition in Plants.

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#### Molecular mechanism of nitric oxide signaling in plants

Nitric oxide (NO) is an important signaling molecule. The major form of bioactive NO species is *S*-nitrosoglutathione (GSNO), which is degraded by GSNO reductase (GSNOR). NO executes its physiological roles mainly through *S*-nitrosylation, a redox-based protein posttranslational modification by covalently adding an NO group to the thiol group of a cysteine residue. We found that the *Arabidopsis* catalase ROG1/CAT3 is a functional transnitrosylase that mediates the *S*-nitrosylation of GSNOR1 via transnitrosylation and its transnitrosylase activity is dependent on the highly conserved Cys-343 residue. The ROG1-mediated transnitrosylation of GSNOR1 promotes its autophagic degradation to regulate its stability, thereby modulating plant development and stress responses. Collectively, this study revealed a novel molecular mechanism of NO signaling in plants.

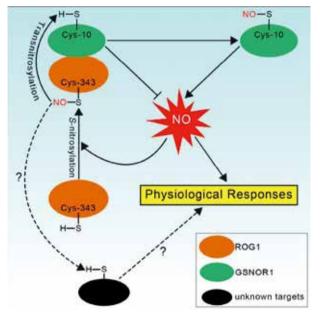


Figure: A proposed model of ROG1/CAT3-regulated NO signaling transduction.

#### Publication

Chen, L.C., Wu, R., Feng, J., Feng, T.P., Wang, C., Hu, J.L., Zhan, N., Li, Y.S., Ma, X.H., Ren, B., Zhang, J., Song, C.P., Li, J.Y., Zhou, J.M., and Zuo, J.R. (2020). Transnitrosylation mediated by the non-canonical catalase ROG1 regulates nitric oxide signaling in plants. Developmental Cell, 53 (4): 444-457.



The main mission of the Center for Molecular Agrobiology (CMA) is oriented towards the national strategic needs of food security and sustainable agricultural production, and is geared to international frontier of plant science and focused on the key scientific issues of the genetics and breeding research in the major crops, to conduct basic and applied basic research.

In 2020, CMA scientists have published 78 papers in peer-reviewed academic journals such as *Science, Cell, Nature Genetics, Nature Biotechnology*, etc. CMA scientists made important contributions to analysis of wheat genome, molecular mechanism underlying the complex agronomic traits, as well as advancing technologies of genome editing, chromosomal engineering, and crop plant breeding.

**Integrative Analysis of Complex Genomics:** Zhixi Tian's lab completed *de novo* genome assemblies and comparative genomic analysis for 26 most representative soybeans, and firstly realized the construction of a graph-based genome in crop plants (Liu et al., *Cell*, 2020). Fei Lu's lab performed whole-genome sequencing of 414 accessions from 25 subspecies of wheat relatives in the genus *Triticum* and *Aegilops*, and constructed a genus-level genetic variation map of wheat (VMap 1.0), which is the first time to construct a genus-level whole-genome genetic variation map in crops and revealed the genetic mechanisms of the successful adaptation of wheat to both natural environments and human selections (Zhou et al., *Nature Genetics*, 2020). Yongbiao Xue's lab defined an epigenomic atlas of subgenome-divergent regulation in haploid wheat, and provide new clues for dissecting the genetic and epigenetic interplay in determining polyploid wheat heterosis.

Coordinated Regulation of Grain Yield and Nutrient Use Efficiency: Xiangdong Fu's lab made an important breakthrough in the improvement of nitrogen use efficiency in rice, they identified a new component of GA signalling, an AP2 transcription factor NGR5, which enhanced grain yield and nitrogen use efficiency through chromatin modification, and provides a novel strategy for improving food security and future agricultural sustainability (Wu et al., Science, 2020). Zhixi Tian's lab revealed the feedback regulatory network of Pd1-Ps-P1 in controlling the pubescence development (Liu et al., Molecular Plant, 2020), and the molecular mechanism underlying GmSWEET10a and GmSWEET10b coordinated regulation of yield and quality in soybean (Wang et al., National Science Review, 2020). In addition, they also showed that the PHYA (E3E4)-Tof11/Tof12-LHY-E1-FT molecular module was involved in photoperiodic control in soybean (Lu et al., Nature Genetics, 2020). Yunhai Li's lab identified the KIX-PPD-MYC-GIF1 and GSK2-LARGE1/OML4 signal transduction pathways in determining seed size (Liu et al., Nature Communications, 2020; Lyu et al., The Plant Cell, 2020), and they found that CUC2/CUC3-DA1-UBP15 regulatory module builds up a novel molecular link between organ size and lateral branch formation (Li et al., The Plant Cell, 2020). Aimin Zhang's lab identified a novel bZIP family transcription factor from Triticum urartu, and its homologue from Triticum aestivum, which could increase starch content and yield, but had no negative effects on other agronomic traits (Song et al., New Phytologist, 2020). lab, elite could the improvements of Hong-Qing Ling's lab found that GLU1, a gene encoding ferredoxindependent glutamate synthase, was involved in iron transportation (Cui et al., Journal of Integrative Plant Biology, 2020); They finely mapped the awn suppressor B1 (Niu et al., Crop Journal, 2020). Yiping Tong's lab demonstrated that TaLAMP1, a gene encoding a putative carboxypeptidase, is critical for wheat adaptation to N availability, and in shaping plant architecture by regulating plant height, spike number per plant and grain number per spike (Fang et al., Plants, 2020).

**The Disease-Resistance Mechanism**: Zhiyong Liu's lab have identified the powdery mildew resistance genes *Pm5e* and *Pm24* from Chinese landraces, and *Pm41* from wild emmer wheat. *Pm24* encodes a tandem kinase protein WTK3, and a rare 6-bp natural deletion confers the broad spectrum powdery mildew resistance (Lu et al., *Nature Communications*, 2020). Both *Pm5e* and *Pm41* encode NLR receptor proteins that are valuable for wheat breeding via molecular marker assisted selection and genome editing (Xie et al., *New Phytologist*, 2020; Li et al., *New Phytologist*, 2020). Daowen Wang's lab discovered a viral protein that interferes with the normal growth of cereal crops. (Jin et al., *Science Advances*, 2020). Meanwhile, they showed that efficient expression and function of a receptor-like kinase in wheat powdery mildew defense required an intron located MYB binding site (Xia et al., *Plant Biotechnology Journal*, 2020).

Qian-Hua Shen's lab identified a new regulatory mechanism of immune depression that SnRK1 could enhance barley immunity against powdery mildew by phosphorylating and destabilizing the WRKY3 (Han et al., *Plant Communications*, 2020). Wenxue Zhai's lab showed that the *OsNBS8R* gene conferred quantitative resistance to bacterial blight in rice, which is regulated by a pathogen effector-inducible miRNA (Jiang et al., *Molecular Plant*, 2020).

**Gene Editing Technology:** Caixia Gao's Lab reviewed applications of CRISPR–Cas in agriculture and plant biotechnology (Zhu et al., *Nature Reviews Molecular Cell Biology*, 2020); developed saturated targeted endogenous mutagenesis editors for directed evolution of plant genes (Li et al., *Nature Biotechnology*, 2020); established plant prime editor systems enabling point mutations, small insertions and deletions (Lin et al., *Nature Biotechnology*, 2020); developed a suite of APOBEC-Cas9 complexes for predictable and targeted deletions in plant genomes (Wang et al., *Nature Biotechnology*, 2020); developed a suite of APOBEC-Cas9 complexes for predictable and targeted deletions in plant genomes (Wang et al., *Nature Biotechnology*, 2020); developed new APOBEC3B-based cytosine base editors to eliminate sgRNA-independent DNA off-target edits (Jin et al., *Molecular Cell*, 2020); developed a multiplexed orthogonal genome editing in plants with a Cas9 nickase and engineered CRISPR RNA scaffolds to simultaneously generate C-to-T, A-to-G and knock out mutations (Li et al., *Genome Biology*, 2020); and achieved fine-tuning quantitative traits in asexually reproducing strawberry and obtained novel

mutants containing a continuum of sugar content using base editing (Xing et al., Genome Biology, 2020).

**Chromosome Engineering and Crop Breeding**: Fangpu Han's lab identified multiple circular RNAs generated via trans-splicing from specific centromeric retrotransposons in maize, which could bind to the centromere region through R-loop structure and affects the centromeric chromatin organization (Liu et al., *PLoS Biology*, 2020). They also found that, in addition to sister chromatid cohesion, *ZmSMC3* participates in early meiotic centromere pairing (Zhang et al., *The Plant Cell*, 2020). Huabang Chen's lab revealed the molecular mechanism underlying maize S-type cytoplasmic male sterility gene *orf355* and transcription factor *ZmDREB1.7* in the synergistic regulation of pollen fertility (Xiao et al., *Molecular Plant*, 2020). Zanmin Hu' lab found the SSR sequence expansion occurred predominately in the centromeric and pericentromeric regions of B genome chromosomes accompanied by little expansion and elimination on other chromosomes during wheat speciation . In addition,

three rice varieties "Zhongheyou 3", "Zhongheyou 5", "Zhongheyou 6" cultivated by Xiangdong Fu's lab, and a soybean variety "Kedou 10" cultivated by Baoge Zhu's lab, were approved by the national variety committee. A wheat variety "Kenuo 2" cultivated by Aimin Zhang's lab was approved by local variety committee of Shandong Province. Junming Li's lab bred a new high-yield water-saving wheat variety Kenong 8162 through a distant hybridization between wheat and *Aegilops*.



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- Zang, J., Huo, Y., Liu, J., Zhang, H., Liu, J., and Chen, H. (2020). Maize YSL2 is required for iron distribution and development in kernels. Journal of Experimental Botany, 71 (19): 5896-5910.
- Huo, Y., Pei, Y., Tian, Y., Zhang, Z., Li, K., Liu, J., Xiao, S., Chen, H., and Liu, J. (2020). *IRREGULAR POLLEN EXINE2* encodes a GDSL lipase essential for male fertility in maize. Plant Physiology, 184 (3): 1438-1454.

## Maize Genetics and Breeding

Huabang Chen, Principal Investigator, Ph.D. (1999, Purdue University, USA).

Research Interests: My lab is focused on corn genetics and breeding. Three areas are of particular interests: 1) Maize germplasm enhancement. 2) Mapping, cloning, functional molecular marker development, and utilization of genes of agronomic importance. 3) Integration of corn genomics, proteomics, and bioinformatics into corn breeding program to maximize corn improvement efficiency and efficacy.

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#### Molecular mechanism of ZmYSL2 in regulating maize kernel size

Maize is one of the most important crops. Studies on kernel development would add value to maize improvement. Here, we characterized a novel small kernel mutant *yellow stripe like 2 (ysl2)* in maize (*Zea mays*). The mutant kernel has light color, delayed development of embryo and endosperm, abnormal protein accumulation and starch deposition. *ZmYSL2* was map-based cloned. It was expressed in endosperm and encode a metal–nicotianamine (NA) transporter. Analysis of transporter activity revealed ZmYSL2-mediated Fe transport from endosperm to embryo during kernel development. Significant changes of nitric oxide accumulation, mitochondrial Fe–S cluster content, and mitochondrial morphology indicated that the proper function of mitochondria also was affected in *ysl2*. Collectively, our study demonstrated that ZmYSL2 had a pivotal role in mediating Fe distribution within kernel and kernel development in maize.

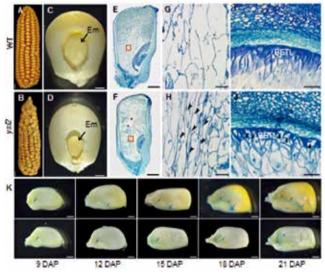




Figure: Phenotype of wild-type and ysl2 mutant and the Fe transport activity of ZmYSL2

### Plant Molecular and Genetic Biology

Xiangdong Fu, Principal Investigator, Ph.D. (2001, Zhejiang University, China & John Innes Centre, UK).

Research Interests: The research interests include determining GA action in the coordinated regulation of plant growth and metabolism, and investigating the genetic basis of the improvement of panicle branching and grain yield in crops.

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## Enhanced sustainable green revolution yield via nitrogen-responsive chromatin modulation in rice

Because environmentally degrading inorganic fertilizer use underlies current worldwide cereal yields, future agricultural sustainability demands enhanced nitrogen use efficiency. We found that genome-wide promotion of histone H3 lysine 27 trimethylation (H3K27me3) enables nitrogen-induced stimulation of rice tillering: APETALA2-domain transcription factor NGR5 (NITROGEN-MEDIATED TILLER GROWTH RESPONSE 5) facilitates nitrogen-dependent recruitment of polycomb repressive complex 2 to repress branching-inhibitory genes via H3K27me3 modification. NGR5 is a target of gibberellin receptor GIBBERELLIN INSENSITIVE DWARF1 (GID1)–promoted proteasomal destruction. DELLA proteins (characterized by the presence of a conserved aspartate-glutamate-leucine-leucine-alanine motif) competitively inhibit the GID1-NGR5 interaction and explain increased tillering of green revolution varieties. Increased NGR5 activity consequently uncouples tillering from nitrogen regulation, boosting rice yield at low nitrogen fertilization levels. NGR5 thus enables enhanced nitrogen use efficiency for improved future agricultural sustainability and food security.

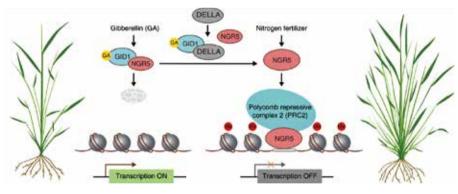


Figure: Nitrogen-responsive chromatin modulation enhances rice tillering



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- Han, X., Wu, K., Fu, X., and Liu, Q. (2020). Improving coordination of plant growth and nitrogen metabolism for sustainable agriculture. aBIOTECH, 1:255–275.
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- Li, C., Zhang, R., Meng, X., Chen, S., Zong, Y., Lu, C., Qiu, J.L., Chen, Y.H., Li, J., and Gao, C. (2020). Targeted, random mutagenesis of plant genes with dual cytosine and adenine base editors. Nature Biotechnology, 38 (7): 875-882.
- Lin, Q., Zong, Y., Xue, C., Wang, S., Jin, S., Zhu, Z., Wang, Y., Anzalone, A.V., Raguram, A., Doman, J.L., Liu, D.R., and Gao, C. (2020). Prime genome editing in rice and wheat. Nature Biotechnology, 38 (5): 582-585.
- Wang, S., Zong, Y., Lin, Q., Zhang, H., Chai, Z., Zhang, D., Chen, K., Qiu, J.L., and Gao, C. (2020). Precise, predictable multinucleotide deletions in rice and wheat using APOBEC-Cas9. Nature Biotechnology, 38(12):1460–1465.
- Jin, S., Fei, H., Zhu, Z., Luo, Y., Liu, J., Gao, S., Zhang, F., Chen, Y.H., Wang, Y., and Gao, C. (2020). Rationally designed APOBEC3B cytosine base editors with improved specificity. Molecular Cell, 79 (5): 728-740.

## **Plant Genome Editing**

Caixia Gao, Principal Investigator, Ph.D. (1997, China Agricultural University, China).

Research Interests: Developing novel technologies to achieve efficient and specific genome engineering, and applying them to study the function of genes and modify plant traits for high-quality, disease resistance and stress tolerance in crop species.

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## Targeted, random mutagenesis of plant genes with dual cytosine and adenine base editors

Targeted saturation mutagenesis of crop genes could be applied to produce genetic variants with improved agronomic performance. However, tools for directed evolution of plant genes, such as error-prone PCR or DNA shuffling, are limited. We engineered five saturated targeted endogenous mutagenesis editors (STEMEs) that can generate de novo mutations and facilitate directed evolution of plant genes. In rice protoplasts, STEME-1 edited cytosine and adenine at the same target site with C > T efficiency up to 61.61% and simultaneous C > T and A > G efficiency up to 15.10%. STEME-NG, which incorporates the nickase Cas9-NG protospaceradjacent motif variant, was used with 20 individual single guide RNAs in rice protoplasts to produce near-saturated mutagenesis (73.21%) for a 56-amino-acid portion of the rice acetyl-coenzyme A carboxylase (OsACC). We also applied STEME-1 and STEME-NG for directed evolution of the *OsACC* gene in rice and obtained herbicide resistance mutations. This set of two STEMEs will accelerate trait development and should work in any plants amenable to CRISPR-based editing.

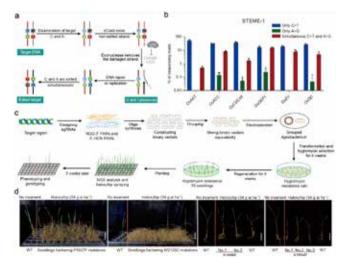


Figure: The development of dual-base editors and its application on directed evolution of endogenous gene. (a) The STEME-mediated C:G>T:A and A:T>G:C base editing strategy. (b) Analysis of editing products by STEME-1. The Y-axis was log-scaled by base 10. All values represent means  $\pm$  s.e.m. (n = 3). (c) Schematic of the procedure for mutating the OsACC CT domain via STEMEs using groups of individual sgRNAs. (d) Mutants of STEMEs-induced amino acid substitutions in OsACC confer resistance to herbicide.



### Plant Chromosome Biology

Fangpu Han, Principal Investigator, Ph.D. (1998, Northeast Normal University, China).

Research Interests: The laboratory is mainly focused on wheat chromosome engineering and genome evolution of polyploidy, structure and function of plant centromere, molecular mechanism of plant meiosis, plant artificial chromosome and specific genome modification.

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## Back-spliced RNA from retrotransposon binds to centromere and regulates centromeric chromatin loops in maize

In most plants, centromeric DNA contains highly repetitive sequences, including tandem repeats and retrotransposons, however, the roles of these sequences in the structure and function of the centromere are unclear. Here, we found that multiple RNA sequences from centromeric retrotransposons (CRMs) were enriched in maize (Zea mays) centromeres, and circular RNAs were generated from CRM1. These circular RNAs bound to the centromere through R-loops. The R-loop formation can promote chromatin loop structure in centromeric regions, which may be important for centromeric H3 variant (CENH3) localization. Our work reveals centromeric chromatin organization by circular CRM1 RNAs via R-loops and chromatin loops, which suggested that CRM1 elements might help build a suitable chromatin environment during centromere evolution. These results highlight that Rloops are integral components of centromeric chromatin and proper centromere structure is essential for CENH3 localization.

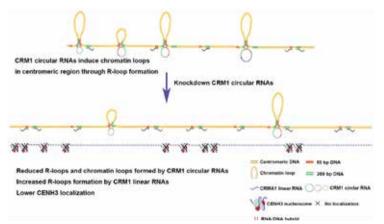


Figure: The model for roles of circular CRM1 RNAs in centromere structure and function.



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### **Plant Biotechnology**

Zanmin Hu, Principal Investigator, Ph.D. (1995, Peking University, China).

Research Interests: Improment of oil seed rape in yield and quality; The theoretical and applied research on biosynthesis of polypeptides and lipopeptid using Chlorella and Bacillus subtilis as bioreactors; Research on plant chromosome repetitive sequences and chromosome engineering.

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## Genome evolution during bread wheat formation unveiled by the distribution dynamics of SSR sequences on chromosomes using FISH

During the bread wheat speciation by polyploidization, a series of genome rearrangement and sequence recombination occurred. Simple sequence repeat (SSR) sequences, predominately located in heterochromatic regions of chromosomes, are the effective marker for tracing the genomic DNA sequence variations. However, to date the distribution dynamics of SSRs on chromosomes of bread wheat and its donors, including diploid and tetraploid Triticum urartu, Aegilops speltoides, Ae. tauschii, T. turgidum ssp. dicocoides, reflecting the genome evolution events during bread wheat formation had not been comprehensively investigated. The genome evolution was studied by comprehensively comparing the distribution patterns of (AAC), (AAG), (AGC), and (AG), in bread wheat Triticum aestivum var. Chinese Spring and its progenitors T. urartu, A. speltoides, Ae. tauschii, wild tetroploid emmer wheat T. dicocoides, and cultivated emmer wheat T. dicoccum. Results indicated that there are specific distribution patterns in different chromosomes from different species for each SSRs. They provided efficient visible markers for identification of some individual chromosomes and SSR sequence evolution tracing from the diploid progenitors to hexaploid wheat. During wheat speciation, the SSR sequence expansion occurred predominately in the centromeric and pericentromeric regions of B genome chromosomes accompanied by little expansion and elimination on other chromosomes. This result indicated that the B genome might be more sensitive to the "genome shock" and more changeable during wheat polyplodization.

During the bread wheat evolution, SSRs including  $(AAC)_n$ ,  $(AAG)_n$ ,  $(AGC)_n$  and  $(AG)_n$  in B genome displayed the greatest changes (sequence expansion) especially in centromeric and pericentromeric regions during the polyploidization from *Ae. speltoides* S genome, the most likely donor of B genome. This work would enable a better understanding of the wheat genome formation and evolution and reinforce the viewpoint that B genome was originated from S genome.

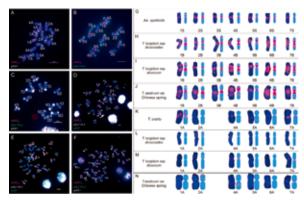


Figure: (AAC), distribution on chromosomes of wheat and its progenitors. A: T. urartu; B: Ae. speltoides; C: Ae. tauschii; D: T. turgidum ssp. dicoccoides; E: T. turgidum ssp. dicoccum; F: T. aestivum var. Chinese Spring; G: Images and mode patterns of genome S individual chromosomes taken from Ae. speltoides (B); H: Images and mode patterns of genome B individual chromosomes taken from T. turgidum ssp. dicoccoides (D); I: Images and mode patterns of genome B individual chromosomes taken from T. turgidum ssp. dicoccum (E): J: Images and mode patterns of genome B individual chromosomes taken from T. aestivum var. Chinese Spring (F); K: Images and mode patterns of genome A individual chromosomes taken from T. urartu (A): L: Images and mode patterns of genome A individual chromosomes taken from T. turgidum ssp. dicoccoides (D); M: Images and mode patterns of genome A individual chromosomes taken from T. turgidum ssp. dicoccum (E): N: Images and mode patterns of genome A individual chromosomes taken from T. aestivum var. Chinese Spring (F). To highlight the red signals, othercoloursof signals were removed artificially in G - J. Red signals, (AAC),. Bar = 10 um.



### Control of Seed and Organ Size in Plants

**Yunhai Li**, Principal Investigator, Ph.D. (2003, Institute of Genetics and Developmental Biology, China). Postdoc fellow (2003-2008, John Inners Center, UK).

Research Interests: To elucidate the molecular mechanisms that control the final size of seeds and organs, and to identify ways of rationally engineering seed and biomass yield in key crops.

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## Transcriptional repression of *GIF1* by the KIX-PPD-MYC repressor complex controls seed size in *Arabidopsis*

Seed size is a key agronomic trait that greatly determines plant yield. Elucidating the molecular mechanism underlying seed size regulation is also an important question in developmental biology. Here, we show that the KIX-PPD-MYC-GIF1 pathway plays a crucial role in seed size control in *Arabidopsis thaliana*. Disruption of KIX8/9 and PPD1/2 causes large seeds due to increased cell proliferation and cell elongation in the integuments. KIX8/9 and PPD1/2 interact with transcription factors MYC3/4 to form the KIXPPD-MYC complex in *Arabidopsis*. The KIX-PPD-MYC complex associates with the typical G-box sequence in the promoter of *GRF-INTERACTING FACTOR 1* (*GIF1*), which promotes seed growth, and repressesits expression. Genetic analyses support that KIX8/9, PPD1/2, MYC3/4, and GIF1 function in a common pathway to control seed size. Thus, our results reveal a genetic and molecular mechanism by which the transcription factors MYC3/4 recruit KIX8/9 and PPD1/2 to the promoter of *GIF1* and repress its expression, thereby determining seed size in *Arabidopsis*.



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- Liu, Z., Li, N., Zhang, Y., and Li, Y. (2020). Transcriptional repression of *GIF1* by the KIX-PPD-MYC repressor complex controls seed size in *Arabidopsis*. Nature Communications, 11 (1): 1846.
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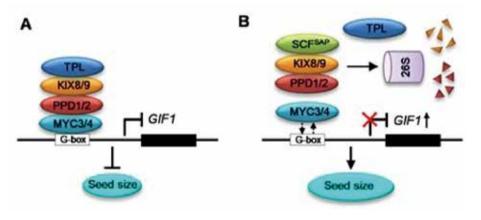


Figure: GIF1 acts genetically with the KIX-PPD-MYC complex to control seed size





#### Publication

Ying, Y., Liu, F.F., Li, G.P., Zheng, Q., Li, B., Li, Z.S., Cheng, J.F., and Li, H.W. (2020). Silencing of the receptor-like cytoplasmic kinase gene *TaRKL1* reduces photosynthetic capacity in wheat. Photosynthetica, 58 (5): 1188-1199.

### Wheat Genetics and Breeding

**Zhensheng Li**, Academician, Chinese Academy of Sciences. Principal investigator. Winner of National Supreme Award of Science and Technology of China in 2006, Award of Outstanding Contribution to China Agriculture in 2005.

Research Interests: The laboratory is mainly interested in: (1) development of new wheat germplasms via wheat distant hybridization, chromosome engineering, and molecular markers; (2) breeding new wheat varieties with tolerance to saline land and improved photosynthetic efficiency.

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## *TaVTE3* regulates photosynthetic efficiency and its alleles may be used for wheat radiation use efficiency improvement

Identification of genes/favorable alleles regulating photosynthetic efficiency can provide molecular elements for wheat radiation use efficiency (RUE) improvement. VTE3 encodes a 2-methyl-6-phytyl-1,4-benzoguinone/2-methyl-6-solanyl-1,4-benzoguinone methyltransferase which catalyzes a key methylation step in both tocopherol and plastoquinone (PQ) synthesis. In this study, TaVTE3 was silenced in wheat by using barley stripe mosaic virus (BSMV)induced gene silencing system to explore its role in regulation of photosynthetic efficiency in wheat. The results showed that when TaVTE3 was silenced, the photosynthetic rate, stomatal conductance, maximal quantum efficiency of photosystem II (Fv/Fm), actual photochemical efficiency ( $\Phi_{psyl}$ ), and electron transport rate (ETR) declined while the dissipated energy flux and non-photochemical quenching (NPQ) elevated significantly in the BSMV:TaVTE3plants. Moreover, the antioxidants contents decreased, which resulted in enhanced susceptibility to photo-oxidative stress in the BSMV:TaVTE3plants. Further, a C/T SNP variation was detected in TaVTE3 on chromosome 6B, which leads to a stop codon. Consistent with the BSMV-VIGS results, the T allele resulted in decline of the photosynthetic rate, Fv/Fm, photosynthetic pigments content, protein quantity and activity of Rubisco, and expression of TaRbcS compared with the C allele wheat. In addition, the T allele wheat was more sensitive to photooxidation stress than the C allele wheat. Moreover, the plant height, spike length, root length and yield reduced significantly in the T allele wheat. Totally, 74 of the 149 winter wheat varieties from Chinese major wheat growth regions carry the T allele, suggesting that this locus was not efficiently selected during traditional wheat breeding processes. Therefore, TaVTE3 and its favorable allele may be a potential target gene for wheat RUE improvement in the future.



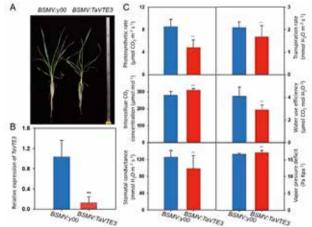


Figure: BSMV-induced gene silencing of *TaVTE3* in wheat reduced photosynthetic efficiency.

### Plant Nutrition and Wheat Genomics

**Hongqing Ling**, Principal Investigator, Director of the State Key Laboratory of Plant Cell and Chromosome Engineering, Ph.D. (1993, Christian-Albrechts University of Kiel, Germany), Postdoctoral fellow (1993-1998, Leibniz Institute of Plant Genetics and Crop Plant Research, Germany), Senior Scientist (1998-2001, Institute of Plant Biology, University of Zurich, Switzerland).

Research Interests: The research directions of our group are molecular biology of plant nutrition and wheat genomics. In the molecular biology of plant nutrition, our research mainly focuses on the molecular regulation mechanism of phosphorus and iron uptake with *Arabidopsis* and tomato; In wheat genomics, the studies focus on illustrating genome structures of Triticum urartu and common wheat by genome sequencing and their comparative genomic analysis.

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## Glutamate synthase 1 is involved in iron-deficiency response and long-distance transportation in *Arabidopsis*

Iron is an essential microelement for plant growth. After uptake from the soil, iron is chelated by ligands and translocated from roots to shoots for subsequent utilization. However, the number of ligands involved in iron chelation is unclear. In this study, we identified and demonstrated that *GLU1*, which encodes a ferredoxin-dependent glutamate synthase, was involved in iron homeostasis. First, the expression of GLU1 was strongly induced by iron deficiency condition. Second, lesion of GLU1 results in reduced transcription of many irondeficiency-responsive genes in roots and shoots. The mutant plants revealed a decreased iron concentration in the shoots, and displayed severe leaf chlorosis under the condition of Fe limitation, compared to wild-type. Third, the product of GLU1, glutamate, could chelate ironin vivoand promote iron transportation. Last, we also found that supplementation of glutamate in the medium can alleviate cadmium toxicity in plants. Overall, our results provide evidence that GLU1 is involved in iron homeostasis through affecting glutamate synthesis under iron deficiency conditions in *Arabidopsis*.



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- Niu, J. Q., Zheng, S. S., Shi, X. L., Si, Y. Q., Tian, S. Q., He, Y. L., & Ling, H. Q. (2020). Fine mapping and characterization of the awn inhibitor *B1* locus in common wheat (*Triticum aestivum* L.). Crop Journal, 8(4), 613-622.

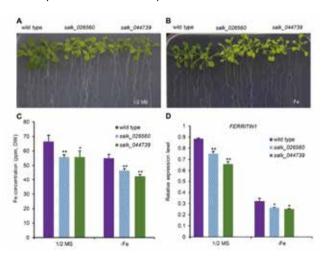
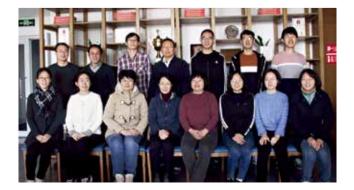


Figure: Phenotypic comparison among the wild-type and SALK\_026560 and SALK\_044739 mutants





#### **Publication**

Lu, P., Guo, L., Wang, Z., Li, B., Li, J., Li, Y., Qiu, D., Shi, W., Yang, L., Wang, N., Guo, G., Xie, J., Wu, Q., Chen, Y., Li, M., Zhang, H., Dong, L., Zhang, P., Zhu, K., Yu, D., Zhang, Y., Deal, K.R., Huo, N., Liu, C., Luo, M.C., Dvorak, J., Gu, Y.Q., Li, H., and Liu, Z. (2020). A rare gain of function mutation in a wheat tandem kinase confers resistance to powdery mildew. Nature Communications, 11 (1): 680.

## Structure and Function of Photosynthetic Complexes

**Cuimin Liu**, Principal Investigator, Ph.D. (2006, University of Freiburg, Germany), Postdoc fellow (2006-2010, Max Planck Institute of Biochemistry, Germany).

Research Interests:Study on the function and activity of enzymes in Calvin-Benson Cycle, try to reveal the mechanisms and synergistic effects of these enzymes which lead to the improvement of plant photosynthetic efficiency. Investigation of structure, function and biogenesis of Rubisco, try to elucidate the mechanisms of Rubisco folding and assembly. Study of composition and function regulation of chloroplast chaperonin Cpn60, try to decipher its function as a housekeeping protein and its regulation roles on stress response.

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#### Folding and assembly of Arabidopsis Rubisco in vitro

Photosynthesis is the material foundation of crop growth, biomass formation and crop production. As the most critical rate-limiting enzyme in the Calvin-Benson cycle, Rubisco directly catalyzes CO<sub>2</sub> fixation, and its activity directly determines plant photosynthetic efficiency and crop yield. Therefore, Rubisco has long been one of the mostprominent targets for improving and increasing crop productivity. In plant, Rubisco is a complex composed of subunits, and its holo-enzyme synthesis is divided into folding and assembly. The folding of large subunits strictly depends on the chaperonin system, and the assembly of holoenzymes strictly requires a number of assembly factors. This complex biogenesis mechanism restricts Rubisco's recombinant expression and large scale mutagenesis, and has been a key factor for hindering the improvement of Rubisco. In the study, we constructed a reconstitution system

of AtRubisco in vitro, achieved the first synthesis of this enzyme and revealed the function mechanism of assembly factors. Our results will provide a theoretical basis for screening efficient Rubisco.

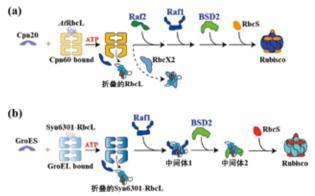




Figure: The biogenesis model of AtRubisco and Syn6301-Rubsico. (A) The biogenesis model of Arabidopsis thaliana Rubisco. The chloroplast chaperonin Cpn60-Cpn20 system mediated the folding of AtRbcL, and the assembly factor promoted the assembly of the folded AtRbcL in the order of RbcX2/Raf2 $\rightarrow$ Raf1 $\rightarrow$ BSD2. (B) The biogenesis model of Syn6301-Rubisco. The cyanobacterial chaperonin GroEL-GroES system mediated the folding of Syn6301-RbcL, and the assembly factors promoted the assembly of the folded Syn6301-RbcL in the order of Raf1 $\rightarrow$ BSD2, producing the Syn6301-RbcL intermediates II.

### Wheat Genomics, Genetics and Breeding

**Zhiyong Liu**, Principal Investigator, Ph.D. (1999, China Agricultural University, China). Postdoctural researcher, (1999-2001, ETH Zurich, Swiss; 2001-2004, Hawaii Agriculture Research Center, USA), Professor (2004-2015, China Agricultural University, China).

Research Interests: My lab is doing wheat genetics & genomics research focused on identification, mapping and map-based cloning of important genes related to disease resistance (powdery mildew, stripe rust, leaf rust, spot blotch, etc.) and agronomic traits (yield, plant architecture, etc.). We are trying to use the genes identified for developing wheat breeder friendly germplasms and wide adapted high-yielding new cultivars.

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## A rare single nucleotide variant in *Pm5e* confers powdery mildew resistance in common wheat

Powdery mildew poses severe threats to wheat production. We report the map-based cloning of the powdery mildew resistance allele Pm5e from a Chinese wheat landrace. We applied a two-step bulked segregant RNA sequencing (BSR-Seq) approach in developing tightly linked or co-segregating markers to *Pm5e*. The function of *Pm5e* was validated by transgenic assay, loss-offunction mutants and haplotype association analysis. *Pm5e* encodes a nucleotide-binding domain leucine-rich-repeat containing (NLR) protein. A rare nonsynonymous single nucleotide variant (SNV) within the C-terminal leucine rich repeat (LRR) domain is responsible for the gain of powdery mildew resistance function of Pm5e, an allele endemic to wheat landraces of Shaanxi province of China. Results from this study demonstrate the value of landraces in discovering useful genes for modern wheat breeding. The key SNV associated with powdery mildew resistance will be useful for marker-assisted selection of Pm5e in wheat breeding programs.

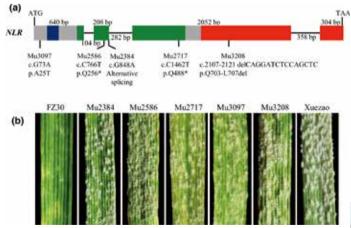


Figure: Validation of the wheat powdery mildew resistance gene *Pm5e* candidate gene nucleotide-binding leucine-rich-repeat receptor (*NLR*) using ethyl methane sulfonate (EMS) mutants.



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Zhou, Y., Zhao, X., Li, Y., Xu, J., Bi, A., Kang, L., Xu, D., Chen, H., Wang, Y., Wang, Y.-g., Liu, S., Jiao, C., Lu, H., Wang, J., Yin, C., Jiao, Y., and Lu, F. (2020). Triticum population sequencing provides insights into wheat adaptation. Nature Genetics, DOI: 10.1038/ s41588-020-00722-w.

### Genomics and Quantitative Genetics in Plant

**Fei Lu**, Principal Investigator, Ph.D. (2010, Institute of Genetics and Developmental Biology, CAS, China), Postdoc Fellow and Research Associate (2010-2017, Institute for Genomic Diversity, Cornell University, USA). He is also a group leader at CAS-JIC Centre of Excellence for Plant and Microbial Science.

Research Interests: To speed up breeding cycle and further improve crop varieties, we are trying to understand the genetics of agronomic traits, especially the complex traits (e.g. yield, heterosis, drought tolerance, resistance, etc.) by using quantitative genetic and statistical genomic approaches. We use wheat and maize as research models. Our research goal is to enhance the genomic selection and/or develop a new system for breeding by efficiently using genomic editing technologies.

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## *Triticum* population sequencing provides insights into wheat adaptation

Bread wheat expanded its habitat from a core area of the Fertile Crescent to global environments within ~10,000 years. The genetic mechanisms of this remarkable evolutionary success are not well understood. By whole-genome sequencing of populations from 25 subspecies within the genera *Triticum* and *Aegilops*, we identified composite introgression from wild populations contributing to a substantial portion (4–32%) of the bread wheat genome, which increased the genetic diversity of bread wheat and allowed its divergent adaptation. Meanwhile, convergent adaptation to human selection showed 2- to 16-fold enrichment relative to random expectation—a certain set of genes were repeatedly selected in Triticum species despite their drastic differences in ploidy levels and growing zones, indicating the important role of evolutionary constraints in shaping the adaptive landscape of bread wheat. These results showed the genetic necessities of wheat as a global crop and provided new perspectives on transferring adaptive success across species for crop improvement.

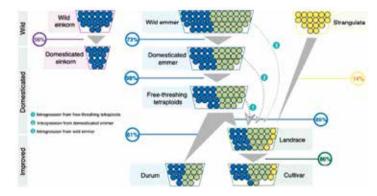




Figure: The shift of genetic diversity of Triticum species and model of diversity recovery in bread wheat.

### **Molecular Plant and Microbe Interactions**

Qianhua Shen, Principal Investigator, Ph.D. (2004, Max Planck Institute for Plant Breeding Research, Germany).

Research Interests: Mechanism of NLR receptor-mediated disease resistance in plants Disease resistance genetics and gene cloning from barley and wheat Cross-kindom signaling and regulation in plant and fungal pathogen interactions.

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## SnRK1 phosphorylates and destabilizes WRKY3 to enhance barley immunity to powdery mildew

Plants recognize pathogens and activate immune responses which usually involve massive transcription reprogramming. The evolutionarily conserved kinase, Sucrose non-fermentingrelated kinase 1 (SnRK1), functions as a metabolic regulator that is essential for plant growth and stress responses. We identity the barley SnRK1 and WRKY3 transcription factor by screening a cDNA library. SnRK1 interacts with WRKY3 in yeast, as confirmed by pull-down and luciferase complementation assays. Forster resonance energy transfer combined with noninvasive fluorescence lifetime imaging analysis indicates that the interaction occurs in the barley nucleus. Transient expression and virus-induced gene silencing analyses indicate that WRKY3 acts as a repressor of disease resistance to the Bgh fungus. Barley plants overexpressing WRKY3 have enhanced fungal microcolony formation and sporulation. Phosphorylation assays show that SnRK1 phosphorylates WRKY3 mainly at Ser83 and Ser112 to destabilize the repressor, and WRKY3 non-phosphorylation-null mutants at these two sites are more stable than the wild-type protein. SnRK1- overexpressing barley plants display enhanced disease resistance to Bah. Transient expression of SnRK1 reduces fungal haustorium formation in barley cells, which probably requires SnRK1 nuclear localization and kinase activity. Together, these findings suggest that SnRK1 is directly involved in plant immunity through phosphorylation and destabilization of the WRKY3 repressor, revealing a new regulatory mechanism of immune derepression in plants.



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- Han, X., Zhang, L., Zhao, L., Xue, P., Qi, T., Zhang, C., Yuan, H., Zhou, L., Wang, D., Qiu, J., and Shen, Q.-H. (2020). SnRK1 phosphorylates and destabilizes WRKY3 to enhance barley immunity to powdery mildew. Plant Communications, 1 (4): 100083.

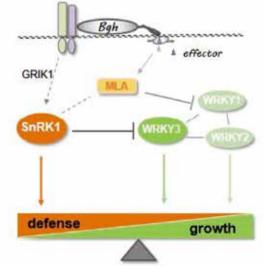
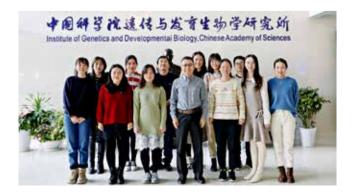


Figure: Molecular model of SnRK1-WRKY3 module regulating plant growth and disease resistance balance





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- Liu, Y., Du, H., Li, P., Shen, Y., Peng, H., Liu, S., Zhou, G.A., Zhang, H., Liu, Z., Shi, M., Huang, X., Li, Y., Zhang, M., Wang, Z., Zhu, B., Han, B., Liang, C., and Tian, Z. (2020). Pan-genome of wild and cultivated soybeans. Cell, 182 (1): 162-176.
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- Liu, S., Zhang, M., Feng, F., and Tian, Z. (2020). Toward a "Green Revolution" for soybean. Molecular Plant, 13 (5): 688-697.

### Functional Genomics and Genetics of Soybean

**Zhixi Tian**, Principal Investigator, Ph.D. (2007, Institute of Genetics and Developmental Biology, CAS, China), Postdoc Fellow and Rsearch Geneticist (2007-2010, Purdue University, USA).

Research Interests: Our group will combine association mapping, QTL mapping, map-based cloning along with comparative genomic approach, as well as construction and utilization of mutant database to identify genes controlling important agronomical traits and to apply them into the soybean breeding, which will facilitate soybean germplasm enhancement and benefit soybean production.

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#### Pan-genome of wild and cultivated soybeans

Soybean is one of the most important vegetable oil and protein feed crops. To capture the entire genomic diversity, it is needed to constructa complete high-quality pan-genome from diverse soybean accessions. In this study, we performed individual de novo genome assemblies for 26 representative soybeans that were selected from 2,898 deeply sequenced accessions. By the result of gene annotation and gene family cluster, we estimated the core/pan gene family size of soybean. Using these assembled genomes together with three previously reported genomes, we constructed graph-based genome and performed pangenome analysis, whichidentified numerous genetic variations that cannot be detected by

direct mapping of short sequence reads onto a single reference genome. The structural variations from the 2,898 accessions that were genotyped based on thegraph-based genome and the RNA-seq data from the representative 26 accessions helped to link genetic variations to candidate genes that are responsible for important traits. Large scale structural variations may affect genome and trait by inducing gene PAV (seed lustre), gene fusion (E3), chromosome rearrangement (I locus), gene expression (Fe efficiency). This pan-genome resource will promote evolutionary and functional genomics studies in soybean.

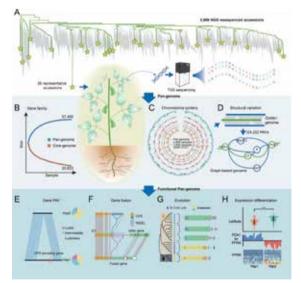


Figure: Pan-genome of wild and cultivated soybeans. (A) Phylogeny of 2,898 soybean accessions and selection of pan-genome samples. (B). Estimation of core/pan-genome size of gene families. (C) Variation of pan-genome and 2,898 soybean accessions. (D) Detecting large scale SV (>50bp) by chromosome synteny and constructing graph-based genome. (E) SV caused gene PAV related to seed lustre. (F) SV induced gene fusion of E3. (G) SV led to complex evolution history of chromosome rearrangement and seed coat color pattern on *I* locus. (H) SV formed haplotypes related to expression differentiation of a gene in an Fe efficiency QTL.



## **Genetics of Plant Nutrition**

Yiping Tong, Principal Investigator, Ph.D. (1999, Institute of Genetics, CAS, China).

Research Interests: 1) Identifying QTL/genes regulating nitrogen and phosphorus use in wheat; (2) Molecular breeding of wheat varieties with improved nitrogen and phosphorus use efficiency.

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## TaLAMP1 plays key roles in plant architecture and yield response to nitrogen fertilizer in wheat

Understanding the molecular mechanisms in wheat response to nitrogen (N) fertilizer will help us to breed wheat varieties with improved yield and N use efficiency. Here we cloned *TaLAMP1-3A*, *-3B* and *-3D* encoding a putativecarboxypeptidase. TaLAMP1s were up-regulated in roots and shoots of wheat plants by low N availability. Compared with wild type, the *TaLAMP1* overexpression lines had a lower spike number per plant but a higher grain number per spike under normal N conditions; while the knockdown lines had a higher plant height but a lower grain number per spike under normal- and low-N conditions (Figure). Comparing the agronomic traits under normal- and low-N conditions revealed that both overexpression and knock-down of *TaLAMP1* inhibited yield response to N fertilizer. Overexpressing *TaLAMP1* greatly increased grain N concentration with no significantly detrimental effect on grain yield under low N conditions, *TaLAMP1* is therefore valuable in engineering wheat for low input agriculture. These results suggested that *TaLAMP1* is critical for wheat adaptation to N availability, and in shaping plant architecture by regulating plant height, spike number per plant and grain number per spike. Optimizing *TaLAMP1* expression may facilitate wheat breeding with improved yield response to N fertilizer.

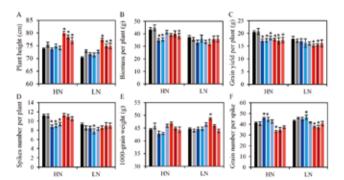


Figure: Agronomic traits of the wide type KN199 and TaLAMP1 transgenic plants grown under low N and normal N conditions in the field experiment. (A) Plant height. (B) Biomass per plant. (C) Grain yield per plant. (D) Spikes number per plant. (E) 1000-grain weight. (F) Grain number per spike. KN199 and NC are wild type and negative transgenic lines, respectively. OE1, OE2 and OE3 are *TaLAMP1-3B* overexpression transgenic lines, R1, R2 and R3 are *TaLAMP1* knock-down transgenic lines. HN represents the normal N condition, and LN represents low N treatment. Data are the mean ± SE of four replications. "\*"indicates statistically significant differences between KN199 and the transgenic lines at the P < 0.05 level.





#### **Publication**

Fang, J.J., Zhu, W.Q., and Tong, Y.P. (2020). Knock-down the expression of brassinosteroid receptor *TaBR11* reduces photosynthesis, tolerance to high light and high temperature stresses and grain yield in wheat. Plants-Basel, 9 (7): 840.



### Molecular Studies of Agronomic Traits and Genetic Improvement of Wheat

**Daowen Wang**, Principal Investigator, Ph.D. (1993, University of East Anglia and John Innes Center, UK).

Research Interests: Research projects include understanding and improving the quality, yield, and disease and stress tolerance traits of wheat through comparative studies of the genes functioning in important biological processes in common wheat and model plant species.

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## A distinct class of plant and animal viral proteins that disrupt mitosis by directly interrupting the mitotic entry switch Wee1-Cdc25-Cdk1

Many animal viral proteins, e.g., Vpr of HIV-1, disrupt host mitosis by directly interrupting the mitotic entry switch Wee1-Cdc25- Cdk1. However, it is unknown whether plant viruses may use this mechanism in their pathogenesis. Here, we report that the 17K protein, encoded by

barley yellow dwarf viruses and related poleroviruses, delays G 2 /M transition and disrupts mitosis in both host (barley) and nonhost (fission yeast, *Arabidopsis* thaliana, and tobacco) cells through interrupting the function of Wee1-Cdc25-CDKA/Cdc2 via direct protein-protein interactions and alteration of CDKA/ Cdc2 phosphorylation. When ectopically expressed, 17K

disrupts the mitosis of cultured human cells, and HIV-1 Vpr inhibits plant cell growth. Furthermore, 17K and Vpr share similar secondary structural feature and common amino acid residues required for interacting with plant CDKA. Thus, our work reveals a distinct class of mitosis regulators that are conserved between plant and animal viruses and play active roles in viral pathogenesis.

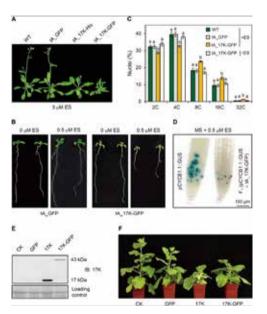


Figure: Inhibition of nonhost plant growth and mitosis by 17K. (A) Effects of 17K expression on Arabidopsis growth analyzed using transgenic lines (tA\_17K-His and tA\_17K-GFP) with inducible expression of 17K by ES and a control line expressing free GFP. (B) Reduction of primary root growth by inducible expression of 17K in transgenic Arabidopsis. (C) Measurement of nuclear DNA contents. DNA contents in the root tip cells of tA\_GFP, tA\_17K-GFP, or WT control cultured with  $0.5\mu M$ ES (+ES) or tA 17K-GFP without ES (-ES) were measured by flow cytometry. The means ( $\pm$  SE) were each calculated from three separate experiments, with significantly different values labeled by nonidentical letters (P < 0.05, ANOVA and LSD for multiple comparisons). (D) Compared to the parental marker line pCYCB1;1::GUS (28), CYCB1,1::GUS signals (represented by the blue precipitates) were substantially decreased in root tips of the F 1 seedlings derived from crossing pCYCB1.1::GUS with tA 17K-GFP, with 17K expression induced by ES. (E) Validation of PEBV vector-mediated expression of 17K and 17K-GFP in N. benthamiana using immunoblotting (IB). (F) Inhibition of N. benthamiana growth by 17K or 17K-GFP. CK, mock control (infiltrated with water).



### **Epigenetic Regulation of Plant Development and Adaptation to Changeable Environments**

Jun Xiao, Principal Investigator, Ph.D. (2012, Institute of Botany, CAS, China).

Research Interests: Construction of the time-course 'Regulome' of wheat spike and seed during distinct developmental stages, and identification as well as functional study of key factors Identification of key factors involved in regulation of agronomic related water use efficiency (WUE) in wheat and functional study Dissecting the epigenetic regulation mechanism of the induction, establishment, maintenance and resetting of vernalization in hexaploid winter wheat

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## Construction the 'Regulome' of wheat spike development and identification of key regulators

Wheat is a major crop in China and worldwide. The yield of wheat is determined by three factors: the spike numbers per unit area, grain number per spike and thousand grain weights. It is of great significance for understanding the molecular mechanism and application of wheat yield improvement to do in-depth study of wheat spike development. Our work integrates multi-dimensional omic data, including transcriptome, histone modification and chromatin accessibility to construct developmental and tissuespecific whole genome-wide transcription regulation network. Based on this, we identified potential key factors involved in regulation of wheat spike development in a systematic and high efficient manner. In combination with DNA variation information from re-sequencing the natural collectives, we have identified elite allele within the regulatory regions of key factors. Through CRISPR-CAS genome editing technique, we are now studying the molecular mechanism of those factors in regulation of the spike development and estimating the potential application value in improving wheat yield.

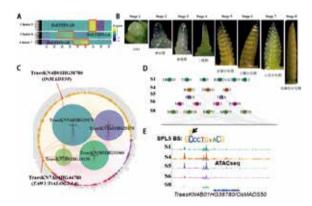


Figure: Construction the dynamic transcriptional regulation network during wheat spike development and preliminary functional analysis of the key factor SPL5-MADS50 module



#### Publication

LEAFY is a pioneer transcription factor and licenses cell reprogramming to floral fate. Nature Communications,In press.





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- Members, C.-N. and Partners (2020). Database resources of the national genomics data center, China national center for bioinformation in 2021. Nucleic Acids Research, DOI: 10.1093/nar/gkaa1022.
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### **Plant Molecular Genetics**

**Yongbiao Xue**, Principal Investigator, Ph.D. (1989, University of East Anglia and John Innes Centre, Norwich, UK).

Research Interests: The laboratory is mainly interested in the molecular control of reproductive barriers in flowering plant, focusing on self-incompatibility (SI) in *Antirrhinum* and *Petunia*. We are also investigating the molecular genetic mechanism of rice response to environmental temperature.

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## Epigenomic atlas of divergent subgenome development and stress responses in wheat

The widely cultivated wheat has a large allohexaploid genome. However, the specificity encoded in wheat genome determining the subgenome-divergent spatio-temporal regulation has been largely unexplored. In this study, we compared the epigenomes (Figure 1) and transcriptomes from a large spectrum of samples under diverse developmental and environmental conditions. A total of 223,976 distal epigenetic regulatory elements (epiREs) were specifically linked to their target promoters with coordinated epigenomic changes. We detected distinct epigenetic architectures of epiREs representing different levels of subgenome divergence. Subgenome-specific epiREs are preferentially marked by H3K27me3 and

displayed high tissue-specific activity. Furthermore, through employing quantitative epigenomic approaches, we detected key responsive cis- and trans-acting factors validated by DNA Affinity Purification and sequencing (DAP-seq), and demonstrated the coordinated interplay between regulatory element (RE) sequence contexts, epigenetic factors, and transcription factors in regulating subgenome divergence. Altogether, our results define an epigenomic atlas of subgenome-divergent regulation in hexaploid wheat and provide new clues for dissecting the genetic and epigenetic interplay in determining polyploid wheat heterosis.

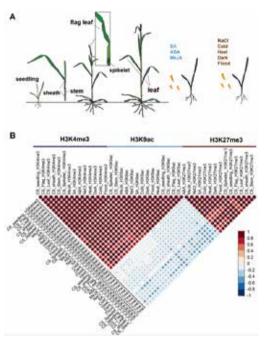




Figure: Profiles of epigenomic modifications across typical developmental stages and in response to various external stimuli

### **Plant Molecular Genetics and Breeding**

Wenxue Zhai, Principal Investigator, Ph.D. (1999, Institute of Genetics, CAS, China).

Research Interests: The laboratory mainly focuses on molecular cloning and molecular breeding of rice functional genes, particularly those with important agronomic traits. The present work includes positional cloning of rice bacterial blight resistance genes, breeding bacterial blight resistant hybrid rice with the cloned genes, as well as rice and maize molecular breeding.

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## A rice NBS-ARC gene conferring quantitative resistance to bacterial blight is regulated by a pathogen effectorinduced miRNA.

The bacterium *Xanthomonasoryzaepv. Oryzae(Xoo)* causes blight in rice worldwide, resulting in significant crop loss. However, no gene underlying quantitative trait loci (QTLs) for resistance against *Xoo* has been cloned yet. Here, we report the map-based cloning of a QTL, in which the *NBS8R* gene confers quantitative resistance to *Xoo. NBS8R* encodes an NB-ARC protein, which is involved in the pathogen/microbe-associated molecular pattern triggered immunity (PTI) and whose expression is regulated by the non-TAL effector XopQ-triggered Osa-miR1876 through DNA methylation.Sequence analysis of *NBS8R* in wild rice species and rice cultivars suggested that the Osa-miR1876 binding sites in the 5'UTR of *NBS8R* are inserted by chance and have undergone variations with Osa-miR1876 throughout the course of evolution. The interaction between *NBS8R* and XopQ-triggered OsamiR1876 ispartially in keeping with the zigzag model (Figure 2), revealing that quantitative genes may also follow this model to control the innate immune response or basal disease resistance, and may prove valuable in utilizing the existing landraces that harbor the *NBS8R* gene but with no Osa-miR1876 binding site in rice breeding for bacterial blight resistance.

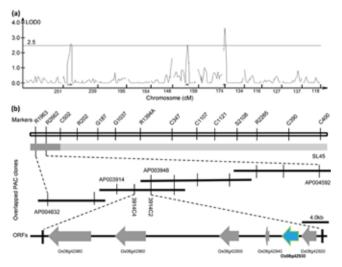
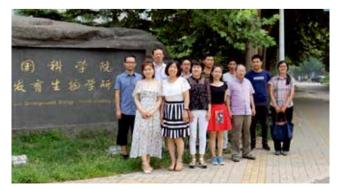


Figure: Map-based cloning of *OsNBS8R* gene conferring rice quantitative resistance to bacterial blight. (a) Three majoreffect QTLs (qBBLL2, qBBLL6 and qBBLL8) that affect lesion length were detected in 98 BIL lines with a LOD threshold of 2.5. Their LOD scores are 2.6, 2.5, and 3.7. (b) Fine mapping of qBBLL8 narrowed it down to a region of ~47.4 kb, which contains six predicted genes. Among them, *NBS8R(Os08g42930)* encoding an NB-ARC protein is the target gene.





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- Jiang, G., Yin, D., Shi, Y., Zhou, Z., Li, C., Liu, P., Jia, Y., Wang, Y., Liu, Z., Yu, M., Wu, X., Zhai, W., and Zhu, L. (2020). OsNPR3.3-dependent salicylic acid signaling is involved in recessive gene xa5-mediated immunity to rice bacterial blight. Scientific Reports, 10 (1): 6313.
- Jia, Y., Li, C., Li, Q., Liu, P., Liu, D., Liu, Z., Wang, Y., Jiang, G., and Zhai, W. (2020). Characteristic dissection of *Xanthomonas oryzae* pv. *Oryzae* responsive MicroRNAs in rice. International Journal of Molecular Sciences, 21 (3):785.
- Han, J., Xia, Z., Liu, P., Li, C., Wang, Y., Guo, L., Jiang, G., and Zhai, W. (2020). TALEN-based editing of *TFIIAy5* changes rice response to *Xanthomonas oryzae* pv. *Oryzae*. Scientific Reports, 10 (1): 2036.



### Wheat Molecular Breeding

Aimin Zhang, Principal Investigator, Ph.D. (1989, Beijing Agricutural University, China).

Research Interests: Marker-assisted selection of important agronomic traits in wheat and molecular design breeding of wheat varieties.

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#### **Publications**

- Wang, D.Z., Yu, K., Jin, D., Sun, L.H., Chu, J.F., Wu, W.Y., Xin, P.Y., Gregova, E., Li, X., Sun, J.Z., Yang, W.L., Zhan, K.H., Zhang, A.M., and Liu, D.C. (2020). Natural variations in the promoter of *Awn Length Inhibitor 1* (*ALI-1*) are associated with awn elongation and grain length in common wheat. Plant Journal, 101 (5): 1075-1090.
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- Liu, H., Mullan, D., Zhang, C., Zhao, S.C., Li, X., Zhang, A.M., Lu, Z.Y., Wang, Y., and Yan, G.J. (2020). Major genomic regions responsible for wheat yield and its components as revealed by meta-QTL and genotypephenotype association analyses. Planta, 252 (4): 65.

## Mapping and genetic basis analysis of *awn Length Inhibitor 1 (ALI-1)* in wheat

Wheat awn plays a vital role in photosynthesis, grain production, and drought tolerance. We conducted a GWAS with 364 wheat accessions and identified 26 loci involved in awn length development, including *B1*, *B2*, *Hd*, and several rice homologs. The dominant awn suppressor *B1* was fine mapped to a 125-kb physical interval, and a C2H2 zinc finger protein *Awn Length Inhibitor 1 (ALI-1)* was confirmed to be the underlying gene of the *B1* locus through the functional complimentary test with native awnless allele. *ALI-1* expresses predominantly in the developing spike of awnless individuals, transcriptionally suppressing downstream genes.

ALI-1 reduces cytokinin content and simultaneously restrains cytokinin signal transduction, leading to a stagnation of cell proliferation and reduction of cell numbers during awn development. Polymorphisms of four SNPs located in ALI-1 promoter region are diagnostic for B1/b1 genotypes, and these SNPs are associated with awn length, grain length and thousand-grain weight. Moreover, ali-1 was observed to increase grain length in wheat, which is a valuable attribute of awn on grain weight, aside from photosynthesis. Therefore, ALI-1 pleiotropically regulates awn and grain development, providing an alternative for grain yield improvement and addressing future climate changes.

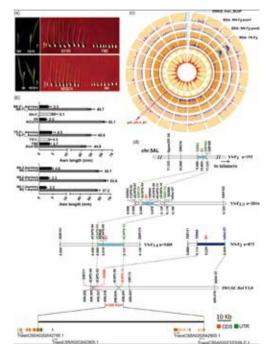


Figure: The ALI-1 locus was fine mapped to a 125 kb interval(a-b) Awn performance of YS-F2, NN-F2, and NILs. (c) A circus plot indicating bulk separating analysis (BSA) enrichment peaks overlapping with the qAL.5A.3\_B1 locus. (d) Fine mapping of the B1 locus using bi-parental mapping populations.



The Center for Developmental Biology (CDB) focuses on addressing fundamental questions of developmental biology using model organisms such as *C. elegans, Drosophila, Xenopus,* zebrafish, mouse, monkey, *Arabidopsis* and rice. The mission is to develop innovative technology to meet our national needs in agriculture and human health. Currently CDB has 21 research groups. Among them, 8 principal investigators were awarded the National Science Fund for Distinguished Young Scholars and 16 Pls were funded by the Hundreds of Talents Program of CAS. During 2020, CDB has made substantial progress in the following areas.

Cell Differentiation and Organ Formation: Successful cloning by somatic cell nuclear transfer (SCNT) requires overcoming significant epigenetic barriers. Dr. Falong Lu with collaborators reports significant enhancement of SCNT efficiency by deriving somatic donor cells carrying simultaneous monoallelic deletion of four H3K27me3-imprinted genes. Quadruple monoallelic deletion of 4 genes increased fibroblast cloning efficiency to 14% compared with a 0% birth rate from wild-type fibroblasts while preventing the placental and body overgrowth defects frequently observed in cloned animals. These results show that lack of H3K27me3 imprinting in somatic cells is an epigenetic barrier that impedes post-implantation development of SCNT embryos and can be overcome by monoallelic imprinting gene deletions in donor cells (Cell Stem Cell, 2020). Dr. Wenxiang Meng's group finds CAMSAP1 plays a decisive role in the neuronal axon identification process by regulating the number of MTs. Neurons lacking CAMSAP1 form a multiple axon phenotype in vitro, while the multipolar-bipolar transition and radial migration are blocked in vivo. They demonstrate that the polarity regulator MARK2 kinase phosphorylates CAMSAP1 and affects its ability to bind to MTs, which in turn changes the protection of MT minus-ends and also triggers asymmetric distribution of MTs. The results indicate that the polarized MT network in neurons is a decisive factor in establishing axon/dendritic polarity and is initially triggered by polarized signals (PNAS, 2020). Dr. Weicai Yang's lab show that MLO5, MLO9 and MLO15 are required for pollen tube responses to ovular signals in Arabidopsis thaliana. This study identifies members of the conserved seven transmembrane MLO family (expressed in the pollen tube) as tethering factors for Ca<sup>2+</sup> channels, reveals a novel mechanism of molecular integration of extracellular ovular cues and selective exocytosis, and sheds light on the general regulation of MLO proteins in cell responses to environmental stimuli (Nature Plants, 2020).

Neurodevelopment and Regeneration: Calcium-dependent proteolytic calpains are implicated in a variety of physiological processes, as well as pathologies associated with calcium overload. Dr. Yongqing Zhang's lab discover calpain activity was inhibited upon knockdown of Ttm50. Their findings reveal the regulation of calpain activation by Ttm50, and shed new light on calpain-associated pathologies (Cell Research, 2020). Zika virus (ZIKV) has evolved into a global health threat because of its causal link to congenital Zika syndrome. Dr. Zhiheng Xu's lab find although mouse brains infected later after birth do not have apparent abnormal brain structure, those mice still show significant impairments of visual cortical functions, circuit organization, and experience-dependent plasticity. The finding suggests that special attention should be paid to all children born to ZIKV infected mothers for screening of abnormal behaviors and sensory function during childhood (Advanced Science, 2020). Neurogenesis in the developing neocortex relies on extensive mitosis of radial glial cells (RGCs) in the apical surface. Dr. Qingfeng Wu's lab find Talpid3, related to Joubert syndrome, is localized to the mother centriole of RGCs and is required for their apical mitosis. Further analysis revealed genetic ablation of Talpid3 results in synchronized, ectopic mitosis of neural progenitors and dysregulated neurogenesis. These findings provide an intriguing perspective for the nonciliogenic role of centriolar proteins in mediating cortical neurogenesis (Cell Reports, 2020). Dr. Jianwu Dai's group have developed a biocompatible collagen scaffold that can be injected directly into the heart, which can provide stable extracellular matrix support for cells and effectively limit cell proliferation from the site of injury. The randomized, single-center clinical trial's result proved collagen gel may be a feasible and safe method to promote cell therapy and set the grounds for adequately powered efficacy studies (JAMA Network Open, 2020).

**Tissue Metabolism and Aging:** The coronavirus disease 2019 (COVID-19) pandemic presents an unprecedented threat to global public health. Dr. Guanghou Shui's lab with collaborator utilize a combination of targeted and untargeted tandem mass spectrometry to analyse the plasma lipidome and metabolome in mild, moderate and severe COVID-19 patients and healthy controls. Their finding suggests that gangliosides (GM3) -enriched exosomes may partake in pathological processes related to COVID-19 pathogenesis, and presents the largest repository on the plasma lipidome and metabolome distinct to COVID-19 (*Cell Metabolism*, 2020). Mild mitochondrial stress experienced early in life can have beneficial effects on the life span of organisms through epigenetic regulations. Dr. Ye Tian's lab find that acetyl-coenzyme A (CoA) represents a critical mitochondrial signal to regulate aging through the chromatin remodeling and histone deacetylase complex (NuRD) in *Caenorhabditis elegans*. The finding reveals the molecular mechanism of the metabolite-mediated epigenome for the regulation of organismal aging (*Science Advances*, 2020). The appearance of Lipid droplets in neurons is not usually observed under physiological conditions, but is associated with neural diseases. Dr. Xun Huang's lab discover that mutations of two key lipolysis genes *atgl-1* and *lid-1* lead to LD appearance in neurons of Caenorhabditis elegans. This neuronal lipid accumulation protects neurons from hyperactivation-triggered neurodegeneration, with a mild decrease in touch sensation. These results suggest the crucial role of neuronal lipolysis in cell-autonomous regulation of neural functions and neurodegeneration (*EMBO Reports*, 2020).



## The Polarity of Golgi Apparatus and Neurodegenerative Diseases

Shilai Bao, Principal Investigator, Ph.D. (1999, The University of Science and Technology of China); Visiting scholar (1999, Purdue University, USA); Postdoctoral fellow (1999-2003, MD Anderson Cancer Center, USA).

Research Interests: Disruption of the polarity of Golgi Apparatus is widely observed in Neurodegenerative Diseases. The investigation into the causal relationship between them and the mechanisms is very important for the understanding and treatment of Neurodegenerative Diseases.

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## Exploring the molecular mechanism to regulate the polarity of Golgi apparatus in neurodegenerative diseases

Abstract: Golgi apparatus is an important component of eukaryotic intracellular membrane system and plays an important role in the posttranslational modification and transport of proteins and lipids. The polar distribution and advanced banded structure of Golgi apparatus is an important structural feature of Golgi apparatus in mammalian cells, but its physiological function is not clear. In previous studies, GM130 knockout mice were found to destroy both the Golgi apparatus Ribbon of Purkinje cells and their polar distribution, leading to progressive Purkinje cells death, the occurrence of severe cerebellar Ataxia. It is the first evidence that a structural disorder of the Golgi apparatus is an important cause of neurodegeneration. Therefore, it is necessary to explore the specific mechanism of the polar distribution of Golgi apparatus. We found severe progressive motor deficits in mice conditioned to knock out LKB1 in the cerebellar Purkinje cells. LKB1 knock-out causes progressive Purkinje cells death, disrupts the Golgi apparatus and affects the polarity distribution, resulting in a more concentrated distribution of Golgi apparatus around the nucleus on the dendrite side. These results will contribute to a comprehensive understanding of the molecular mechanisms regulating Golgi apparatus polarity changes and provide new therapeutic ideas for neurodegeneration.

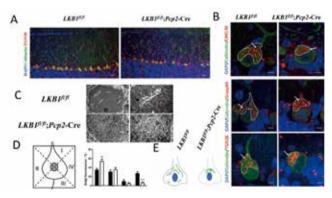




Figure: Altered Golgi morphology and positioning in LKB1fl/fl;Pcp2-Cre mice.(A and B) TGN38 (A) or GM130、Grasp65 and TGN38 (B) immunohistochemical staining (red) of cis- or trans-Golgi and calbindin-D28K (green) of Purkinje cells in mice at 16 wk. White arrows in B indicate Golgi elements (Scale bar, A 20  $\mu$ m and B 5  $\mu$ m.) (C) Electron micrographs of the Golgi apparatus in Purkinje cells of LKB1fl/fl and LKB1fl/fl;Pcp2-Cre mice at 24 wk. (Scale bar, Left 1  $\mu$ m and Right 0.5  $\mu$ m.) (D) Quantitation of Golgi positioning in Purkinje cells of LKB1fl/fl and LKB1fl/fl;Pcp2-Cre mice at 16 wk. The scheme used to determine Golgi distribution in Purkinje cells is shown at Left with quantitation shown at Right. The Golgi distribution was assessed using GM130 as the marker. *n* = 10; \*\*P < 0.01. Data are presented as mean ± SEM.

### Plant Molecular Response and Control Mechanism

Fan Chen, Principal Investigator, Ph.D. (1997, Ehime University, Japan).

Research Interests: The laboratory mainly focuses on plant molecular response and control mechanism. The signal transduction during plant development are concerned. The high-through omics and genetics analysis were used to construct the network of gene expression and regulation in higher plant.

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## Mechanism analysis of FZP 3'UTR involved in the regulation of panicle development in Oryza sativa

The molecular mechanism underlying panicle development regulation has been fascinating, since grain number is a critical factor for grain yield in rice. *FZP* is an important QTL locus for regulating grain number, and its 3'UTR has broad-spectrum inhibitory effect. Overexpression of *FZP* 3'UTR resulted in the decreased tiller number, branches and grain numbers per panicle. Further research showed the CU rich element, i.e. CURE was the functional element in *FZP* 3'UTR, and the copy number of CURE elements in *FZP* 3'UTR were different among varieties, as well as in wild rice. In addition, the copy number of CURE was positively correlated with the inhibition of 3'UTR and the grain number per panicle. OsPTBP1/5, polypyrimidine binding proteins, were trans-acting elements that directly binding with CURE of *FZP* 3'UTR, which regulated the expression of *FZP* by post-transcriptional regulation, and participated in the formation of rice panicle architecture. A new regulation pathway was designed for rice molecular breeding.

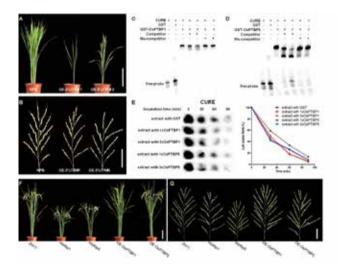
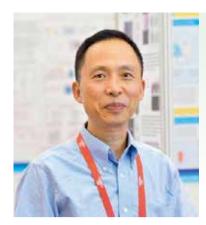


Figure: *FZP* 3'UTR is involved in the regulation of panicle development in *Oryza* sativa. (A-B) *FZP* 3'UTR is a negative regulator of rice grain number per panicle. (A) Bar=15cm. (B) Bar=5cm. (C-E) OsPTBP1/5 bind to the CU-rich element (CURE) of *FZP* 3'UTR and promote the degradation of *FZP* mRNA. (F-G) *OsPTBP1/5* positively regulate grain number per panicle of rice. (F) Bar=15cm. (G) Bar=5cm.





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### **Regenerative Medicine and Tissue/Organ Fabrication**

Jianwu Dai, Principal Investigator, Ph.D. (1998, Duke University, USA).

Research Interests: 1) Study of tissue or organ regenerative microenvironment; 2) Development of products to promote tissue and organ regeneration or repair; 3) Tissue and organ fabrication. Email: jwdai@genetics.ac.cn

# Effect of intramyocardial grafting collagen scaffold with mesenchymal stromal cells in patients with chronic ischemic heart disease---a randomized clinical trial

We developed an injectable porous collagen scaffold hydrogel. Accordingly, a randomized, single-center clinical trial was conducted to evaluate the safety and feasibility of the intramyocardial delivery of collagen hydrogel with human umbilical cord-derived mesenchymal stromal cells (hUC-MSCs) in patients with chronic ischemic heart disease (CIHD) immediately after undergoing coronary artery bypass grafting (CABG). Forty-four participants completed 12 months of follow-up. For the primary end point, no significant differences in serious adverse events, myocardial damage markers, and renal or liver function were observed among all groups after treatment. At 12 months after treatment, the mean infarct size percentage was decreased, life quality and New York Heart Association heart function class were improved in collagen/cell group. When focused on patients with baseline LVEF of 40% of less, mean LVEF in the collagen/cell group patients increased by 9.35% (95%CI, 1.96% to 16.75%; P =0.02) at 12 months, while that in the cell group and the control group increased by 6.59% (95%CI, 2.61% to 10.56%; P =0.004) and 3.62% (95%CI, -3.25% to 10.50%; P =0.25), respectively.

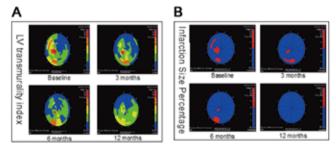


Figure: Representative cardiac magnetic resonance images showed a representative patient with significant reduction in myocardial transmurality (A) and infarct size in the collagen/cell group (B). In panel A, segment colors from blue to red means transmurality from small to large scar size. In panel B, segments with red color means more than half of the segments was infarct tissue.



### The Molecular Mechanisms of Neural Development

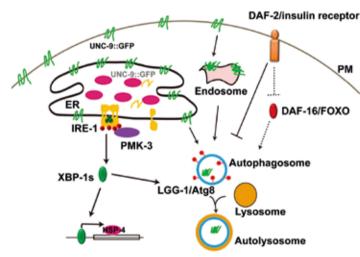
Mei Ding, Principal Investigator, Ph.D. (2004, University of California, Santa Cruz, USA).

Research Interests: The simple nervous system of *C. elegans* is well suited for studying neural development. We systematically explore the molecular mechanisms of neural development by labeling neuronal processes and synapses in living animals. The main research contents include: neurite outgrowth, neuron recognition, synapse formation and steady-state regulation, electrical synapse regulation, neuron-specific ER stress regulation and so on.

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### Alleviating chronic ER stress by p38-Ire1-Xbp1 pathway and insulinassociated autophagy in *C. elegans* neurons

The accumulation of unfolded proteins triggers the ER stress response (UPR), which allows cells to fight against fluctuations in protein expression under both physiological and pathological conditions. Severe acute ER stress responses can be induced by drug treatment. However, such intense ER stress rarely occurs ubiquitously in every cell type in vivo. Here, we designed a genetic system in the nematode *C. elegans*, which allows us to induce ER stress in specific cells, without drug treatment or any other external stimuli, and then to monitor the stress response. The p38 MAPK directly acts on the phosphorylation of IRE-1 to promote the stress response. Meanwhile, the insulin receptor function through autophagy activation to counteract the p38-IRE-1-XBP-1 pathway. Together, these results reveal an intricate cellular regulatory network in response to chronic stress in multicellular organism.





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Figure: The PMK-3-IRE-1-XBP-mediated UPR acts in parallel with insulininhibited autophagy to alleviate chronic stress induced by excess UNC-9::GFP protein.





### Neural Stem Cells and Neurogenesis

Weixiang Guo, Principal Investigator, Ph.D. (2008, Institute of Zoology, CAS, China).

Research Interests: The research in our laboratory focuses on understanding the cellular and molecular mechanisms that regulate neural stem cells and neural development, with the goal to develop better treatment for human neurological disorders.

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## Rab7 effector wdr91 promotes autophagy-lysosome degradation in neurons by regulating lysosome fusion

The lysosomes act as the key organelle for intracellular degradation, and undergo constant fusion-fission cycles. When lysosomal biogenesis or fusion-fission cycles is impaired, autophagic cargo remains trapped in the autophagosomes and is inefficiently degraded, leading to various pathological conditions such as neurodegeneration and lysosomal storage disorders. However, the mechanisms underlying lysosome homotypic fusion and their implications in neurological disorders remain largely unknown. Here, we found that brain-specific inactivation of WDR91 leads to massive neuronal loss and progressive motor and behavioral deficits in mice. Furthermore, WDR91 competes with VPS41 for binding to Rab7 to regulate HOPS complex assembly. However, reducing HOPS activity by knock-down of VPS41 expression partially rescues lysosomal dysfunction and defective autophagy. Thus, these data unravel the essential role of WDR91 in neurons by regulating lysosome fusion and autophagy-lysosomal pathway.

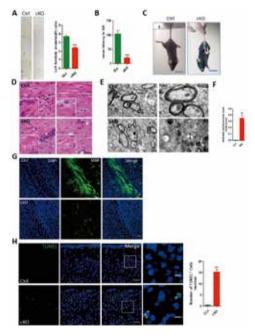


Figure: Loss of WDR91 leads to neuron degeneration. (A) Represenative footprint images of wild type (WT) and Wdr91-/- (cKO) mice at postnatal day 21 (P21). Bars, mm. (B) Quantification the ratio of stride to width of footprint in (A). (C) Duration of falling from a rotating rod of WT and cKO mice at P21. (D) Represenative images of WT and cKO mice at P21 in tail-suspending assays. (E) Haematoxylin and eosin (H&E) staining in WT and cKO mice at P21. Bars, mm. TEM images of myelin (F) and lysosomes (G) of WT and cKO mice at P21. Bars, 200 nm.(H)TUNEL staining of cerebral cortex of WT and cKO mice at P21. Bars, mm. Error bars represent SEM. \*\*, P < 0.01; \*\*\*, P < 0.001.

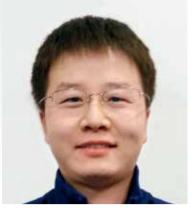


## Single-Molecule Live-Cell Imaging and Lipid Conversion

Kangmin He, Principal Investigator, Ph.D. (2013, Academy for Advanced Interdisciplinary Studies, Peking University); Postdoctoral fellow/Instructor/ Research Associate (2013-2019, Harvard Medical School/Boston Children's Hospital, USA)

Research Interests: Using live-cell single-molecule quantitative imaging, genome editing, sensor design and other tools to study the molecular mechanism of endocytosis and intracellular trafficking, and the dynamic signaling transduction and phospholipid trafficking/conversion during membrane trafficking.

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## Single-molecule live-cell imaging of signaling transduction and lipid conversion during membrane trafficking

By utilizing a varieties of techniques such as live-cell single-molecule imaging, genome-editing and sensor design, we study endocytosis and the accompanying signaling transduction and phospholipid trafficking/conversion in living cells. By creating a new generic PI(3,4,5)P<sub>3</sub> sensor, and a set of clathrin-specific PI(3,4,5)P<sub>3</sub>, PS, PA and cholesterol sensors, we have uncovered the distinct localization and dynamics of different lipids in clathrin-mediated endocytosis. By creating and imaging genome-edited cell lines expressing fluorescent protein tags, we are studying systematically the dynamic signal activation process of EGFR and the mechanism of early endosome formation by single molecule imaging. By using a novel clathrin-specific phosphoinositide sensor, we have uncovered the existence of a novel recycling pathway in cells. We are currently working on solving the structure, dynamics and regulation of the new pathway.

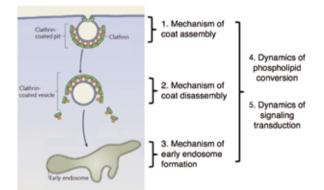


Figure: Research topics in the lab: we use live-cell single-molecule quantitative imaging, genome editing, sensor design and other tools to investigate the molecular mechanisms of endocytosis and intracellular trafficking, the dynamic phospholipid trafficking/conversion as well as the dynamic signaling transduction and regulation during membrane trafficking.





#### **Publications**

Yang, L.L., Liang, J.J., Lam, S.M., Yavuz, A., Shui, G.H., Ding, M., and Huang, X. (2020). Neuronal lipolysis participates in PUFA-mediated neural function and neurodegeneration. EMBO Reports, DOI: ARTN e5021410.15252/embr.202050214. Yao, Y., Ding, L., and Huang, X. (2020). Diverse

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### Lipid Metabolism, Development and Disease

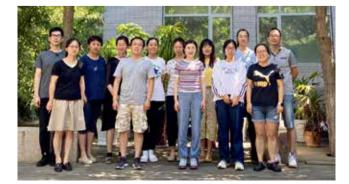
Xun Huang, Principal Investigator, Ph.D. (2003, University of California, Santa Cruz, USA).

Research Interests: The long-term goal of our laboratory is to understand the regulating mechanisms of lipid metabolism and disease. Currently the main interest of our lab is to understand the cell biology and developmental biology of lipid metabolism in adipose tissue, neuron and reproduction system under normal physiological conditions and under disease conditions.

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#### Lipid droplet dynamics in neurons regulates neurodegeneration

Fat is stored in lipid droplets (LDs) which are widespread but unique hydrophobic organelles in cytosol. The abnormality of lipid droplet dynamics could cause many metabolic diseases, such as obesity and fatty liver. The nervous system has lots of lipids but few lipid droplets exist in neurons. However, some neuronal diseases (e.g. hereditary spastic paraplegia, Parkinson's disease) are reported to have relationship with lipid droplet dysfunctions. Studying lipid droplet dynamics in neurons and its effect on neurons is important for treating neural diseases. Our study published in EMBO Reports reveals that lipolysis and lipogenesis regulate lipid droplet dynamics and lipid droplet accumulation reduces the available PUFAs which regulate the neurodegeneration through participating into phospholipids. In this study, we found that disturbing lipolysis or enhancing lipogenesis in neurons leads to lipid droplet accumulation in neurons of C. elegans, which is the first time to confirm lipid droplet regulation in neurons in vivo. The ectopic lipid droplets in neurons reduce the normal functions such as touch sensation but increase the ability to defend from hyperactivation-triggered neurodegeneration. Further investigation showed that PUFA synthesis pathway synergistically with lipolysis regulates neurodegeneration. Blocking PUFA incorporation into phospholipids further increase the protection from neurodegeneration. Thus, PUFAs from lipolysis hydrolyzing and *de novo* synthesis participate in hyperactivation-triggered neurodegeneration through incorporation into phospholipids. This is a promising result since maybe one day we could alleviate neurodegeneration through adjusting the content of polyunsaturated fatty acids in our food.



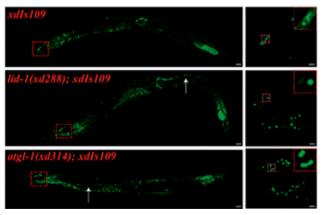


Figure: *lid-1(xd288)* and *atgl-1(xd314*) display ectopic LD accumulation in neurons.

### Bioimaging; Micro/Nano Optics; Nano-biology

Yuqiang Jiang, Principal Investigator, Ph.D. (2004, Shanxi University, China).

Research Interests: 1) Optical micro-manipulation and its application in dynamics of biomacromolecules; 2) Synthesis, characterization, and biological application of nano-bioprobes. 3) R&D of crop phenotypic facility based on CT and optical imaging.

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## Spheroidal trap shell beyond diffraction limit induced by nonlinear effects in femtosecond laser trapping

Beyond diffraction limit multi-trapping of nanoparticles is important in numerous scientific fields, including biophysics, materials science and quantum optics. Here, we demonstrate the 3-dimensionad (3D) shell-like structure of optical trapping well induced by nonlinear optical effects in the femtosecond Gaussian beam trapping for the first time. Under the joint action of gradient force, scattering force and nonlinear trapping force, the gold nanoparticles can be stably trapped in some special positions, or hop between the trap positions along a route within the 3D shell. The separation between the trap positions can be adjusted by laser power and numerical aperture (NA) of the trapping objective lens. With a high NA lens, we achieved dual traps with less than 100 nm separation without utilizing complicated optical systems or any on-chip nanostructures. These curious findings will greatly extend and deepen our understanding of optical trapping based on nonlinear interaction, and generate novel applications in various fields, such as micro/nano-fabrication, sensing, novel micromanipulations.

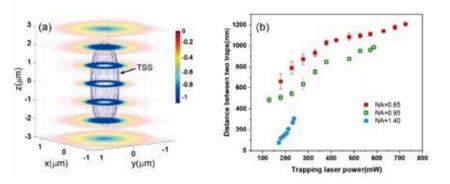


Figure: (a) The three-dimensional (3D) distribution of potential energy of trapped GNPs in the focal volume; (b) The trap separations beyond the diffraction limit as the trapping power decreased. Data of trapping objectives with different NA also showed.



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#### Publication

Dong, X., Zhou, J., Qin, H.B., Xin, B., Huang, Z.L., Li, Y.Y., Xu, X.M., Zhao, F., Zhao, C.J., Liu, J.J., Luo, M.H., and Zeng, W.B. (2020). Anterograde viral tracer herpes simplex virus 1 strain H129 transports primarily as capsids in cortical neuron axons. Journal of Virology, 94 (8): e01957-19.

### Molecular Mechanisms of Vesicular Transport

Jia-Jia Liu, Principal Investigator, Ph.D. (2000, University of Chicago, USA).

Research Interests: The laboratory is mainly interested in membrane trafficking and membrane dynamics in neurons, focusing on regulatory mechanisms for molecular motor-driven vesicular transport in mammalian cells and role(s) of vesicular transport and cytoskeleton dynamics in synaptic development and plasticity.

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#### Endophilin A1 initiates structural plasticity of dendritic spines

Dendritic spines of excitatory neurons undergo activity-dependent structural and functional plasticity, which are cellular correlates of learning and memory. However, mechanisms underlying the rapid morphological changes immediately after NMDAR-mediated Ca<sup>2+</sup> influx into spines remain poorly defined. Our study demonstrates that endophilin A1, a neuronal N-BAR protein, functions in the initial phase of long-term potentiation (LTP). Ca<sup>2+</sup>/calmodulin enhance its binding to its effector p140Cap and the plasma membrane. Upon LTP induction, endophilin A1 rapidly localizes to plasma membrane and promotes actin polymerization via p140Cap. Moreover, both the p140Cap-binding and membrane-binding capacities of endophilin A1 are required for LTP and longterm memory. These data indicate that endophilin A1 functions immediately downstream of calmodulin to promote spine enlargement in response to Ca<sup>2+</sup> influx in the initial phase of structural plasticity.

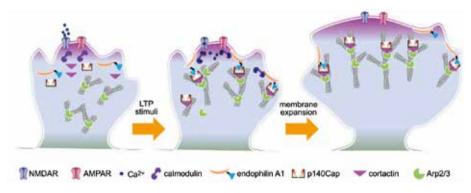


Figure: Model for endophilin A1-mediated initial expansion of spine head upon LTP induction.



### **Epigenetics and Cell Fate Determination**

**Falong Lu**, Principal Investigator, Ph.D. (2011, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences); Postdoctoral fellow (2012-2017, UNC-Chapel Hill / Howard Hughes Medical Institute and Boston Children's Hospital/Harvard Medical School / Howard Hughes Medical Institute, USA).

Research Interests: 1) Epigenetic regulation mechanism not encoded by DNA sequence; 2) Epigenetic regulation of mammalian reproduction and the mechanism of zygote genome activation; 3) Epigenetic mechanism of cell fate determination.

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#### MeCP2 regulates heterochromatin through phase separation

MeCP2 is a well-known methyl-DNA binding protein, which can bind to DNA and CpG methylated DNA through its MBD domain. MeCP2 complete deficiency in human is lethal, while heterozygous MeCP2 mutation in young girls cause Rett syndrome. However, the mechanism of MeCP2 mutation in leading to the serious developmental disease is not very clear. In addition, the role of MeCP2 in regulating heterochromatin structure and function awaits further investigation. We found that MeCP2 can form liquid-liquid phase separation in the presence of DNA or CpG methylated DNA. This phase separation leads to high concentration of MeCP2 in the phase separated compartment. In addition, CpG methylation on DNA can enhance the phase separation of MeCP2-DNA complex. Very interestingly, the phase separation ability of MeCP2 can be impaired in the presence of Rett mutations, indicating that the defects of MeCP2 phase separation ability can be an important mechanism leading to Rett syndrome.

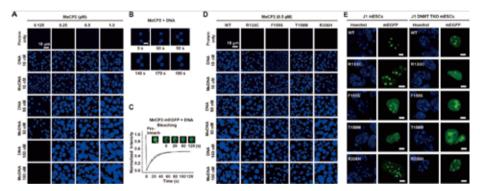


Figure: MeCP2 regulates heterochromatin through phase separation (Cell Discovery, 2020)



- Liu, Y., Nie, H., and Lu, F. (2020). Dynamic RNA 3' uridylation and guanylation during mitosis. iScience, 23 (8): 101402.
- Fan, C., Zhang, H., Fu, L., Li, Y., Du, Y., Qiu, Z., and Lu, F. (2020). Rett mutations attenuate phase separation of MeCP2. Cell Discovery, 6: 38.
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### Molecular Mechanism of Interaction between Microsporidia and Host

**Runlin Z. Ma**, Principal Investigator. Ph.D. (1991, the Kansas State University, USA); Postdoctoral fellow (1992-1996, University of Illinois, USA); Research Assistant Professor (1997-2001, University of Illinois, USA).

Research Interests: Genome structure of Antonospora locustae and transcriptional interactions with its host locust.

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## Complete genome of a unicellular parasite (*Antonospora locustae*) and transcriptional interactions with its host locust

Microsporidia are a large group of unicellular parasites that infect insects and mammals. The simpler life cycle of microsporidia in insects provides a model system for understanding their evolution and molecular interactions with their hosts. However, no complete genome

is available for insect-parasitic microsporidian species. We report a full and complete genome sequence of A. locustae based on second- and third-generation genome-sequencing technologies. The genome of the parasite consists of 17 chromosomes on which a total of 1857 coding genes. An ultra-low GC region of approximately 25 kb on 16 of the 17 chromosomes. Phylogenetic study based on genomes suggested that microsporidia are a special evolutionary group. Transcriptomic analysis showed that A. locustae can systematically inhibit the locust phenoloxidase PPO, TCA and glyoxylate cycles, and PPAR pathways to escape melanization, and can activate host energy transfer pathways to support its reproduction in the fat body. Our study provides a platform and model for studies of the molecular mechanisms of microsporidia-host interactions, and for understanding the evolution of microsporidia.

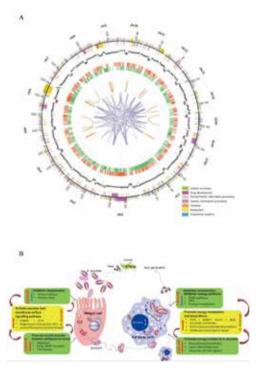


Figure: (A) A circular representation of the complete genome of A. locustae. The outermost circle shows chromosome size (kb) and the distribution of KEGG pathways, as indicated with colour-coded bars for each chromosome. The second circle from the outside shows the variation in GC content for each of the 17 chromosomes, characterized by a sharp decrease in GC content near the centre of each chromosome. The third circle from the outside represents the distribution of coding genes on the positive strand (red) and negative strand (green) of DNA, respectively. The non-coding RNA (ncRNA) detected is shown in the fourth circle. Information about long-fragment repeat sequences is represented in the fifth circle, and genomic longfragment repeat sequences are indicated on the innermost circle. (B) Simplified life cycle of infection by A. locustae and critical interactions with its locust host at the level of gene transcription.



## The Dynamic Coordination of Cytoskeleton Network

**Wenxiang Meng**, Principal Investigator, Ph.D. (2002, Osaka University, Japan), Postdoctoral fellow (2002-2004, Mie University, Japan). Research Scientist (2004-2010, Riken Center for Developmental Biology, Japan).

Research Interests: The functions and regulation mechanisms of dynamic coordinated microtubule- actin filaments network.

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## CAMSAP1 breaks the homeostatic microtubule network to instruct neuronal polarity

The establishment of axon/dendrite polarity is fundamental for neurons to integrate into functional circuits, and this process is critically dependent on microtubules (MTs). In the early stages of the establishment process, MTs in axons change dramatically with the morphological building of neurons; however, how the MT network changes are triggered is unclear. Here we show that CAMSAP1 plays a decisive role in the neuronal axon identification process by regulating the number of MTs. Neurons lacking CAMSAP1 form a multiple axon phenotype *in vitro*, while the multipolar-bipolar transition and radial migration are blocked *in vivo*. We demonstrate that the polarity regulator MARK2 kinase phosphorylates CAMSAP1 and affects its ability to bind to MTs, which in turn changes the protection of MT minus-ends and also triggers asymmetric distribution of MTs. Our results indicate that the polarized MT network in neurons is a decisive factor in establishing axon/dendritic polarity and is initially triggered by polarized signals.

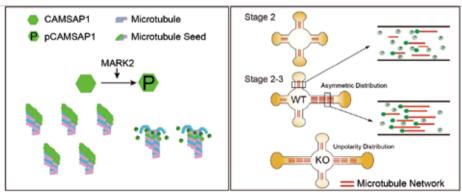


Figure: Hypothetical model of the regulatory mechanisms and functions of CAMSAP1 in establishing neuronal polarization. CAMSAP1 is phosphorylated by MARK2 to modify its MTs binding ability, which alters its protection of MT minus-ends as well as triggers differential accumulation of MTs between individual neuronal processes. The asymmetric distribution of MT networks will instruct the proper formation of the axon during neuronal polarization.



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## Lipidomics, Metabolic Disorders

**Guanghou Shui**, Principal Investigator, Ph.D. (2004, National University of Singapore, Singapore).

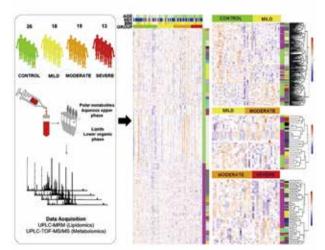
Research Interests: Dr Shui's laboratory is mainly interested in lipidomics and lipid metabolism, with particular focus on the role of dysregulated lipid metabolism in development and/or diseases using model organisms such as Caenorhabditis elegans; and employing lipidomic approaches to elucidate potential biomarkers for major diseases.

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## Omics-driven systems interrogation of metabolic dysregulation in covid-19 pathogenesis

The coronavirus disease 2019 (COVID-19) pandemic presents an unprecedented threat to global public health. Herein, we utilized a combination of targeted and untargeted tandem mass spectrometry to analyze the plasma lipidome and metabolome in mild, moderate, and severe COVID-19 patients and healthy controls. A panel of 10 plasma metabolites effectively distinguished COVID-19 patients from healthy controls (AUC = 0.975). Plasma lipidome of COVID-19 resembled that of monosialodihexosyl ganglioside (GM3)-enriched exosomes, with enhanced levels of sphingomyelins (SMs) and GM3s, and reduced diacylglycerols (DAGs). Systems evaluation of metabolic dysregulation in COVID-19 was performed using multiscale embedded differential correlation network analyses. Using exosomes isolated from the same

cohort, we demonstrated that exosomes of COVID-19 patients with elevating disease severity were increasingly enriched in GM3s. Our work suggests that GM3-enriched exosomes may partake in pathological processes related to COVID-19 pathogenesis and presents the largest repository on the plasma lipidome and metabolome distinct to COVID-19.



1002 identified metabolites 598 lipids & 404 polar metabolites

Figure: 1002 identified metabolites, 598 lipids & 404 polar metabolites



### **Molecular Energetics**

John R. Speakman, Principal Investigator, Ph.D. (1984, University of Stirling, UK); Fellow of the Royal Society of Edinburgh (2004); Fellow of the Academy of Medical Sciences (2008); Fellow of the Academy of Europe (2012). Fellow of the Royal Society (2018); CAS foreign member (2019). Fellow of US National Academy (2020).

Research Interests: The molecular basis of the regulation of food intake, energy expenditure and body composition. In particular we aim to understand the causes and consequences of the phenomenon of obesity. Our work includes studies of humans, model animals in captivity and wild animals.

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## Effects of dietary macronutrients and body composition on glucose homeostasis in mice

As a major health issue, obesity is linked with elevated risk of type 2 diabetes. However, whether disrupted glucose homeostasis is due to altered body composition alone, or dietary macronutrients play an additional role, independent of their impact on body composition, remains unclear. We investigated the associations between macronutrients, body composition, blood hormones and glucose homeostasis. We fed C57BL/6N mice 29 different diets with variable macronutrients for 12 weeks. After 10 weeks, intraperitoneal glucose tolerance tests (ipGTT) were performed. Generalized linear models (GLMs) were generated to evaluate the impacts of macronutrients, body composition and blood hormones on glucose homeostasis. The area under the glucose curve (AUC) was strongly associated with body fat mass, but not dietary macronutrients. AUC was significantly associated with fasting insulin levels. Six genes from transcriptomic analysis of eWAT (epididymal white adipose tissue) and sWAT (subcutaneous white adipose tissue), were significantly associated with AUC. These genes may encode secreted proteins that play important previously unanticipated roles in glucose homeostasis.

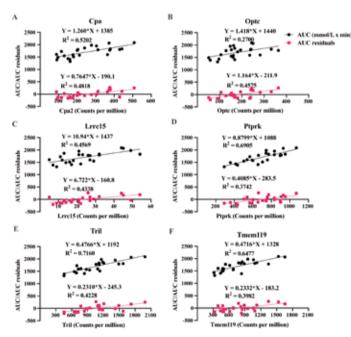
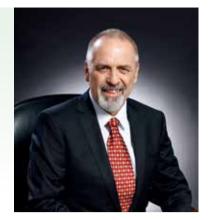


Figure: Linear regression between area under the glucose curve (AUC) and AUC residuals and the expression levels of 6 genes. Values were mean values for each diet group. (A) *Cpa2*. (B) *Optc*. (C) *Lrrc15*. (D) *Ptprk*. (E) *Tril*. (F) *Tmem119*.





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### Early Embryonic Development on Health

**Fangzhen Sun**, Principal Investigator, Ph.D. (1990, Cambridge University, UK); Postdoctoral Fellow (1989-1990, Cambridge University, UK); Research Scientist and Fellow (1990-1994, Cambridge University, UK)

Research Interests: Fertilization, epigenetic reprogramming on major adult diseases of early embryo-origin.

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### The nucleosome domain in human sperm affects male fertility

Infertility in couples of childbearing age has become a common problem affecting over 10-15% of couples worldwide and has been increasing year by year. Male infertility contribute about 50% of overall and the dysregulation of sperm function is the main cause. The histones in human sperm play an important role in regulating sperm normal physiological functions and the embryonic development. Our research has found that the histones in sperm can form a higher structure named nucleosome domain (ND). Combined with epidemiological survey data analysis, we found that this structure are significantly correlated with fertile sperm; by using super-resolution structured illumination microscopy (SIM) and biochemical analysis, it was demonstrated that the nucleosome domain can serve as a biomarker for male sperm fertility, and constitute a new class of epigenetic factors that affect the male reproductive capacity.

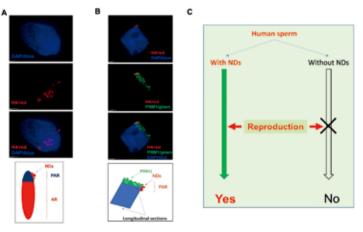


Figure: The nucleosome domains (NDs) are occurred in human normal sperm. (A) NDs localized in the middle-posterior region of sperm. (B) After slicing, NDs were shown to residue only on the surface of sperm nuclei. (C) The working model for NDs in affecting male reproduction capacity.



## Mitochondrial Stress Signaling and Aging

**Ye Tian**, Principal Investigator, Ph.D. (2010, National Institute of Biological Sciences, China); Postdoctoral Fellow, (2010-2016, Salk Institute; University of California, Berkeley).

Research Interests: Dr. Tian's laboratory is mainly interested in molecular mechanism of mitochondrial stress response, focusing on the mitochondrial unfolded protein response (UPR<sup>mt</sup>), epigenetic regulation of mitochondrial stress and transgeneration inheritance.

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## NuRD mediates mitochondrial stress-induced longevity via chromatin remodeling in response to acetyl-CoA level

Mild mitochondrial stress experienced early in life can have beneficial effects on the life span of organisms through epigenetic regulations. Here, we report that acetyl-coenzyme A (CoA) represents a critical mitochondrial signal to regulate aging through the chromatin remodeling and histone deacetylase complex (NuRD) in *Caenorhabditis elegans*. Upon mitochondrial stress, the impaired tricarboxylic acid cycle results in a decreased level of citrate, which accounts for reduced production of acetyl-CoA and consequently induces nuclear accumulation of the NuRD and a homeodomain-containing transcription factor DVE-1, thereby enabling decreased histone acetylation and chromatin reorganization. The metabolic stress response is thus established during early life and propagated into adulthood to allow transcriptional regulation for life-span extension. Furthermore, adding nutrients to restore acetyl-CoA production is sufficient to counteract the chromatin changes and diminish the longevity upon mitochondrial stress. Our findings uncover the molecular mechanism of the metabolite-mediated epigenome for the regulation of organismal aging.



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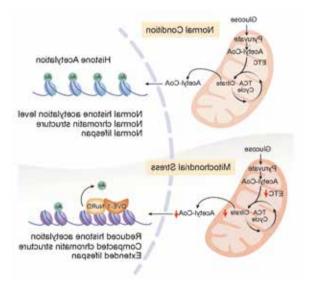


Figure: Model of acetyl-CoA links mitochondrial stress to longevity via NuRDmediated chromatin remodeling





## The Genetic Network of Germline Development

Zhaohui Wang, Principal Investigator, Ph.D. (1998, University of Chicago, USA).

Research Interests: Using fruit fly and mouse models to study: 1) the microenvironment of germline stem cells; 2) the regulation of mitosis-to-meiosis switch

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## An RNA kinase clp1/cbc regulates meiosis initiation in spermatogenesis

CLP1, TSEN complex, and VCP are evolutionarily conserved proteins whose mutations are associated with neurodegenerative diseases. In this study, we have found that they are also involved in germline differentiation. To balance quantity and quality in gametes production, germ cells expand themselves through limited mitotic cycles prior to meiosis. Stemming from our previous findings on the correlation between mRNA 3'-processing and meiosis entry, here we identify that the RNA kinase Cbc, the Drosophila member of the highly conserved CLP1 family, is a component of the program regulating the transition from mitosis to meiosis. Using genetic manipulations in Drosophila testis, we demonstrate that nuclear Cbc is required to promote meiosis entry. Combining biochemical and genetic methods, we reveal that Cbc physically and/or genetically intersects with Tsen54 and TER94 (VCP ortholog) in this process. The C-terminal half of Tsen54 is both necessary and sufficient for its binding with Cbc. Further, we illustrate the functional conservation between Cbc and mammalian CLP1 in the assays of subcellular localization and Drosophila fertility. As CLP1, TSEN complex, and VCP have also been identified in neurodegenerations of animal models, a mechanism involving these factors seems to be shared in gametogenesis and neurogenesis.



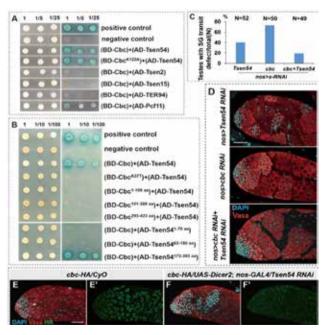


Figure: Cbc interacted with Tsen54 to regulate the transition to meiosis.

## Stem Cell Lineage Tracing and Neural Regeneration

**Qingfeng Wu**, Principal Investigator, Ph.D. (2015, Institute of Neuroscience, Chinese Academy of Sciences, Shanghai, China); Postdoctoral Fellow, (2012-2016, Johns Hopkins University, USA).

Research Interests: 1) Development of nuclear brain structure. 2) Neural control of metabolic disorders. 3) Homeostatic regulation of neural stem cells.

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# Talpid3-mediated centrosome integrity restrains neural progenitor delamination to sustain neurogenesis by stabilizing adherens junctions

The cerebral cortex of mammalian brain is organized into layers of specialized neuronal subtypes by orchestrated stem cell maintenance, expansion, fate commitment and differentiation. Dysfunction of Centrosomal protein Talpid3 in human predisposes the patients to Joubert syndrome, which is an autosomal recessive ciliopathy condition characterized by cerebellum and hindbrain malformation, hypotonia in infancy with laterataxia and intellectual disability. However, the molecular mechanisms underpinning the cognitive impairment in patients with Joubert syndrome and the consequences of Talpid3 dysfunction outside of cilia remain unclear. In this study, the researchers found that Talpid3 dysfunction in RGCs predisposes them to pathological delamination and disrupts basal-to-apical nuclear migration in both cellautonomous and cell non-autonomous manners during cortical neurogenesis. These results were confirmed in conditional Talpid3 knockout mice. Further investigation showed that loss of Talpid3 disrupts adherens junction belt, leading to pathological delamination and premature differentiation of RGCs in the embryonic brains and reduced neuronal number in the postnatal brains. Taken together, Talpid3-mediated centrosome integrity maintains the anchoring of RGCs to VZ and regulate their delamination via stabilizing adherens junctions during cortical neurogenesis.



### **Publication**

Wang, J., Li, T., Wang, J.-L., Xu, Z., Meng, W., and Wu, Q.-F. (2020). Talpid3-mediated centrosome integrity restrains neural progenitor delamination to sustain neurogenesis by stabilizing adherens junctions. Cell Reports 33(11): 108495-108495.

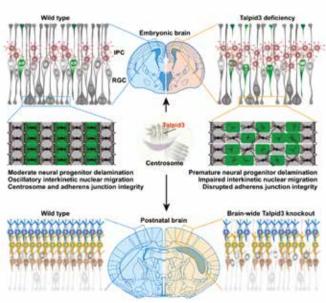


Figure: A schematic model showing the molecular mechanism of Talpid3 in regulating cortical neurogenesis





### Publication

Zhao, Z.Q., Shang, Z.W., Vasconcelos, Z., Li, C.F., Jiang, Y.S., Zu, S.L., Zhang, J.Y., Wang, F.C., Yao, L., Jung, J.U., Brasil, P., Moreira, M.E., Qin, C.F., Kerin, T., Nielsen-Saines, K., Cheng, G.H., Zhang, X.H., and Xu, Z.H. (2020). Zika virus infection leads to variable defects in multiple neurological functions and behaviors in mice and children. Advanced Science, 7 (18).

### Signal Transduction Diseases & Development

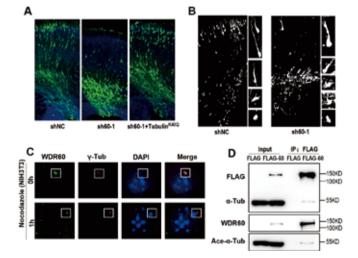
Zhiheng Xu, Principal Investigator, Ph.D. (1999, Rutgers University, USA); Postdoc and Research Associate (1999-2005, Columbia University, USA).

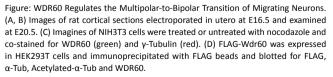
Research Interests: The laboratory is mainly interested in studying the underlying mechanisms involved in brain development, and in different diseases including microcephaly, schizophrenia, autism, and neurodegenerative diseases.

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## WDR60 regulates the multipolar-to-bipolar transition of migrating neurons during brain development

Mutations of repeat domain 60 (WDR60) have been identified in short-rib polydactyly syndromes (SRPS I–V), a group of lethal congenital disorders. However, the underlying mechanism is still unclear. WDR60 is a member of the WD40 repeat protein family. We have previously reported that the mutations of the same family member, affects neurogenesis and leads to microcephaly. Our current study demonstrates that Loss of WDR60 perturbs the multipolar-to-bipolar transition and migration of neuron during cortical development. We show that WDR60 is highly expressed in the pericentrosome and the basal body of cilia in the neuronal precursors and migrating neurons. Mechanically, we demonstrate that WDR60 controls microtubule organization and possibly, the trafficking of cellular components. Importantly, the migration defect caused by Wdr60 knockdown could be rescued by the stable form of  $\alpha$ -Tubulin,  $\alpha$ -Tubulin K40Q (an acetylation-mimicking mutant). These findings identified a non-cilia function of WDR60 and provided insight into its biological function, as well as the pathogenesis of WDR60 deficiency associated with SRPS.







## **Molecular Genetics of Sexual Plant Reproduction**

**Weicai Yang**, Principal Investigator, Director of the Institute, Ph.D. (1994, Wageningen University, The Netherlands); Postdoctoral Fellow (1994-2000, Wageningen University; Cold Spring Harbor Laboratory, USA; The Institute of Molecular Agrobiology, Singapore); Senior Scientist and Principal Investigator (2000-2003, Temasek Life Sciences Laboratory, Singapore).

Research Interests: The laboratory aims to decipher molecular mechanisms controlling male-female interactions, sperm delivery, fertilization, and early embryo development in plants using multi-disciplinary approaches; to explore molecular mechanism that maintains the hypoxia microenvironment for symbiotic nitrogen fixation in legumes and the possibility to engineer nitrogen fixation in nonlegumes.

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## Integration of ovular signals and exocytosis of a Ca<sup>2+</sup> channel by MLOs in pollen tube guidance

The spatiotemporal regulation of Ca<sup>2+</sup> channels at the plasma membrane in response to extracellular signals is critical for development, stress response and reproduction, but is poorly understood. During flowering-plant reproduction, pollen tubes grow directionally to the ovule, which is guided by ovule-derived signals and dependent on  $Ca^{2+}$  dynamics. However, it is unknown how ovular signals are integrated with cytosolic Ca<sup>2+</sup> dynamics in the pollen tube. Here, we show that MILDEW RESISTANCE LOCUS O 5 (MLO5), MLO9 and MLO15 are required for pollen tube responses to ovular signals in Arabidopsis thaliana. Phenotypically distinct from the ovule-bypass phenotype of previously identified mutants, mlo5 mlo9 double-mutant and mlo5 mlo9 mlo15 triple-mutant pollen tubes twist and pile up after sensing the ovular cues. Molecular studies reveal that MLO5 and MLO9 selectively recruit Ca<sup>2+</sup> channel CNGC18-containing vesicles to the plasma membrane through the R-SNARE proteins VAMP721 and VAMP722 in trans mode. This study identifies members of the conserved seven transmembrane MLO family (expressed in the pollen tube) as tethering factors for Ca<sup>2+</sup> channels, reveals a novel mechanism of molecular integration of extracellular ovular cues and selective exocytosis, and sheds light on the general regulation of MLO proteins in cell responses to environmental stimuli.



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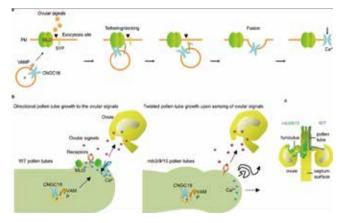


Figure: The working model of MLO-mediated vesicle fusion of CNGC18 to the plasma membrane in response to ovular signals. a, Model of the MLOs-mediated CNGC18-vesicle fusion through the syntaxin (SYP) and VAMP to the plasma membrane site where the ovular signals are sensed. b, Model of pollen tube response to the ovule. c, The diagram of the pollen tube twisting phenotype of mlo5/9/15 mutants.





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- Zhao, K., Hong, H., Zhao, L., Huang, S., Gao, Y., Metwally, E., Jiang, Y., Sigrist, S.J., and Zhang, Y.Q. (2020). Postsynaptic camp signalling regulates the antagonistic balance of drosophila glutamate receptor subtypes. Development (Cambridge, England) 147(24): 1-12.

## Molecular Mechanisms of Major Neurodevelopmental Diseases

**Yongqing Zhang**, Principal Investigator, Ph.D. (1991, China Agricultural University, China).

Research Interests: 1). Study the molecular and cellular bases of major neurological diseases including intellectual disability and autism in order to develop an intervention and/or a cure for the diseases, using non-human primates, domestic dogs and Drosophila melanogaster as model organisms. 2). Taking an interdisciplinary approach including molecular, cell, developmental and genetic methods to study brain development, structure and function, using Drosophila as a model system.

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## Structural remodeling of active zones is associated with synaptic homeostasis

Perturbations to postsynaptic glutamate receptors (GluRs) trigger retrograde signaling to precisely increase presynaptic neurotransmitter release, maintaining stable levels of synaptic strength, a process referred to as homeostatic regulation. However, the structural change of homeostatic regulation remains poorly defined. At wild-type *Drosophila* neuromuscular junction synapse, there is one Bruchpilot (Brp) ring detected by superresolution microscopy at active zones (AZs). In the present study, we report multiple Brp rings (i.e., multiple T-bars seen by electron microscopy) at AZs of both male and female larvae when GluRs are reduced. At *GluRIIC*-deficient neuromuscular junctions, quantal size was reduced but quantal content was increased, indicative of homeostatic presynaptic potentiation. Consistently, multiple Brp rings at AZs were observed in the two classic synaptic homeostasis models (i.e., *GluRIIA* mutant and pharmacological blockade of GluRIIA activity). Furthermore, postsynaptic overexpression of the cell adhesion protein Neuroligin 1 partially rescued multiple Brp rings phenotype. Our study thus supports that the formation of multiple Brp rings at AZs might be a structural basis for synaptic homeostasis.

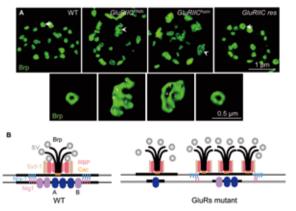




Figure: Multiple Brp rings cluster together in *GluRIIC*<sup>RMAI</sup> and *GluRIIC*<sup>PMPO</sup> mutants. A, Super resolution images of NMJ boutons of WT, *GluRIIC*<sup>PMPO</sup>, *GluRIIC*<sup>CMMAI</sup>, *GluRIIC*<sup>CMMAI</sup>, and *GluRIIC* res larvae stained with anti-Brp (green). Arrows indicate a single Brp ring. Arrowheads indicate multiple Brp rings. Scale bar, 1 um. B, Model describing the role of Nlg1/Nrx in controlling the formation of multiple T-bars at GluRIIC mutant NMJs.

The center for molecular systems biology was established in 2006. The mission of the center is to pursue research related to human health and agricultural development using multidisciplinary approaches comprising computational biology, bioinformatics, systems biology, structural biology, evolutionary genomics. Research at the center focuses on the hidden regulatory mechanisms underlying gene expression, the assembly, modification and dynamics of macromolecules, the noise and robustness of biological systems. Currently, the center has six group leaders, including one recipient of the *National Science Fund for Distinguished Young Scholars*, three recipients of the *National Science Fund for Excellent Young Scholars*, three fellows of the *National One Thousand Talents Program for Young Scientists*, one fellow of the *National Ten Thousand Talents Program for Scientific and Technological Innovation Leading Talents*, and three fellows of the *CAS One Hundred Talents Program*. In 2020, the center has received research funds from the Ministry of Science and Technology, National Natural Science Foundation of China, Chinese Academy of Sciences. The center has made substantive progresses in a variety of research directions with 22 SCI papers published. The center has 3 patents approved of which one is PCT patent, and submitted one patent application.

**Single Cell Transcriptome:** Dr. Wenfeng Qian's group recapitulated the cellular and developmental responses to abiotic stresses in rice using Single-cell transcriptome analyses. The metabolism, environmental adaptation, and reproduction of plants depend on the division of labor among individual cells. However, cells with various functions are often studied as a bulk where their specificities cannot be distinguished. Based on the large-scale single-cell transcriptome method which can assist in identifying cell types, the functional heterogeneity of different types of cells can be explored. Here, Dr. Wenfeng Qian's group applied a barcode-based single-cell RNA sequencing to the aerial organs and the underground root tip of rice seedlings grown in four different environments (control, low-nitrogen, iron-deficiency, and high-salinity). They captured the transcriptomes of ~200 thousand cells and identified all major cell types, particularly classified the subpopulations of mesophyll cells in monocot for the first time. They observed significant cell-type heterogeneity in gene expression changes, clarifying the specific roles of each type of cell in stress response. Moreover, they also found that abiotic stresses impacted the composition of cell populations, which could be explained by the differences of development through reconstructing trajectories of cell differentiation. Collectively, their study represents a benchmark-setting data resource of single-cell transcriptome atlas for rice seedlings and an illustration of exploiting such resources to drive discoveries in plant biology.

Dr. Qiang Tu's group systematically profiled the gene expression and chromatin accessibility of the medaka ovary using single-cell RNAseq and single-cell ATAC-seq technologies. Germ cells transfer genetic information from one generation to the next. In mammalian testes, germline stem cells (GSCs) produce sperms continually, while there is no direct evidence for the presence of GSCs in mammalian ovaries. Some lower vertebrates with high fecundity, such as fish and amphibia, possess GSCs in their mature ovaries. Medaka is the first vertebrate in which ovarian GSCs have been identified. Therefore, the medaka ovary provides an important model system for studying vertebrate GSC development. Currently, little is known about molecular regulatory mechanisms underline this process, except a few marker genes have been identified. In this study, they were able to distinguish all cell types along the ovarian GSC developmental trajectory. For each cell type, they identified specifically expressed genes and specifically active open chromatin regions. By integrating two single-cell data sets, they inferred the gene regulatory network underlining the developmental trajectory of ovarian GSCs. Finally, they discovered multiple transcription factors and associated *cis*-regulatory elements, which could be the master controls of the GSC maintenance, meiosis initiation, and oocyte differentiation. In short, the integration of single-cell RNA-seq and single-cell ATAC-seq data described the gene expression and genomic regulatory landscape of ovarian GSC development, shed light on the big picture of regulatory mechanisms underline this process.

**Functional Proteomics:** Dr. Yingchun Wang's group evaluated the potential risk of the advanced peak determination (APD) algorithm of mess spectrum (MS) in distorting isobaric labeling-based single-shot proteome quantitation. The recent development and implementation of APD algorithm with MS instrument dramatically increased the sampling quantity of low abundance features for MS/MS fragmentation. After in-depth evaluation, they found that with APD on, many chimeric spectra were acquired through co-fragmentation of high abundance contaminants with low abundance targets, and such co-fragmentations were largely avoided when APD was off. To evaluate whether such a co-fragmentation could significantly distort the accuracy of the isobaric-labeling based quantitation of the low abundance target, a single-shot TMT experiment was performed using a two-proteome model, whereby each TMT channel contained premixed peptides from human and cyanobacterium with a known ratio. Unexpectedly, they found that APD did not significantly distort TMT ratios, probably because the majority of the APD-specific chimeric spectra were not identifiable. Nevertheless, a few examples of significant distortion of TMT ratios of low abundance peptides caused by APD were found through manual inspection. These studies suggest that APD should be off in a single-shot TMT experiment to improve the accuracy of MS data analysis.

**Structural Biology:** Dr. Yuhang Chen's group carried out the structural and functional analyses on the SLAC1 channel which is directly involved in regulating stomatal pores. The stomatal pore, formed by two specialized guard cells, serves as the gateway for water transpiration and atmospheric CO<sub>2</sub> influx for photosynthesis. Environmental stimuli such as drought, darkness, ozone, low air humidity

and high levels of carbon dioxide can induce the closure of stomatal pores. The guard cells respond to a wide range of environmental stimulus and convert them into appropriate osmotic pressure change to regulate the openness of stomatal pore aperture. When the osmotic pressure increases, these cells will swell and the pore will open. And when the osmotic pressure decreases, these cells will collapse and the pore will close down. Plant hormone abscisic acid (ABA) plays a central role in controlling stomatal closure via activation of a complex signaling pathways that are mediated by receptors, kinase/phosphatases, and ion channels. Dr. Yuhang Chen's group obtained a 2.97 Å cryo-EM structure of SLAC1 witch was purified from *Barachypodium distachyon* (*Bd*SLAC1), and further characterized the channel activity of SLAC1 in *Arabidopsis*. They employed a novel SLAC1::OST1 fusion design for examining SLAC1 phosphorylation, whereby they systematically characterized critical sites responsible for channel activation. Inspired by the structure, they also conducted mutagenesis on the highly conserved phenylalanine residues related to channel gating. Altogether, their structural and functional analyses provide insights into SLAC1 gating and activity modulation. These findings allow them to propose a mechanism for finely tuned SLAC1 control of the stomatal pore in response to environmental stimuli.

**Developmental Regulatory Network Biology:** *Caenorhabditis elegans* is the first multicellular organism to have its cell atlas fully mapped. However, the molecular processes underlying the cell atlas remains elusive. To model cellular state-fate relationships, Dr. Zhuo Du's team generated a protein expression atlas encompassing 266 transcription factors (TFs) in precisely lineaged embryonic cells using imaging-based single-cell analysis. Tracing the dynamics of cellular states as defined by combinatorial TF expression reveals intricate state-fate relationships. Cellular states change continuously but at a varying speed across lineage and time. Intriguingly, a cell's trajectory from initial to terminal states undergoes extensive detours in which state transitions are generally not directional towards the terminal state but take a "winding" path, except for upon terminal differentiation. Single-cell analysis also discovers unexpected diversity of cellular states. In the process of tissue formation, cellular states from multiple lineages converge but exhibit intra-tissue diversities according to lineages, shaped by lineage-restricted expression of tissue-specific TFs. Furthermore, TFs exhibit widespread transient expression that increases the temporal state diversity and fate complexity. They constructed spatiotemporal cascades and identified new functions of TFs in neurogenesis and muscle differentiation, demonstrating high functional relevance and utility. Collectively, this study presents a valuable protein atlas and unravels complex state-fate relationship during metazoan embryogenesis.

**Small-Molecule Drug Repurposing:** At the early stage of 2019-nCoV outbreak, with the hope to identify candidate drugs for 2019-nCoV, Dr. Xiu-Jie Wang's group predicted potential inhibitors against SARS-CoV-2 coronavirus M protease from clinically approved medicines. Basing on the structural information of clinical effective medicines, they adopted a computational approach and identified 10 available commercial medicines which may function as inhibitors for the main protease M<sup>pro</sup> of SARS-CoV-2. Compared to lopinavir/ritonavir, most of these predicted drugs could form more hydrogen bonds with SARS-CoV-2 M<sup>pro</sup>, thus may have higher mutation tolerance than lopinavir/ritonavir. Moreover, the binding pockets of these drugs on M<sup>pro</sup> are conserved between SARS-CoV M<sup>pro</sup> and SARS-CoV-2 M<sup>pro</sup>, indicating the potential of these drugs as inhibitors for other coronaviruses with similar M<sup>pro</sup> binding sites and pocket structures.

## Structural and Functional Analysis on Biological Macromolecules

Yuhang Chen, Principal Investigator, Ph.D. (2002, Tsinghua University, China), Postdoctoral fellow and associate research scientist (2002-2012, Columbia University, USA).

Research Interests: Structural and functional analysis on membrane transport proteins; Structural and functional analysis on epigenetic regulators involved in chromatin assembly; Structure-based seed design for herbicide-resistance crop.

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## Structure and activity of SLAC1 Channels for stomatal signaling in leaves

The stomatal pore, formed by two specialized guard cells, serves as the gateway for water transpiration and atmospheric CO<sub>2</sub> influx for photosynthesis. Environmental stimuli such as drought, darkness, ozone, low air humidity and high levels of carbon dioxide can cause stomatal pores to close. The guard cells respond to a wide range of environmental stimulus and convert them into appropriate osmotic pressure change to regulate the openness of stomatal pore aperture. When the osmotic pressure increases, these cells will swell and the pore will open. When the osmotic pressure decreases, these cells will collapse and the pore will close down. Plant hormone abscisic acid (ABA) plays a central role in controlling stomatal closure via activation of a complex signaling pathways that are mediated by receptors, kinase/phosphatases, and ion channels. We solved the cryo-EM structure of SLAC1 from Barachypodium distachyon (Bd SLAC1) at 2.97 Å resolution (Figure 1A), and further characterized the channel activity on Arabidopsis SLAC1. We employed a novel SLAC1::OST1 fusion design (Figure 1B) for examining SLAC1 phosphorylation, whereby we systematically characterized critical sites responsible for channel activation (Figure 1C). Inspired by the structure, we also conducted mutagenesis on the highly conserved phenylalanine residues related to channel gating. Altogether, our structural and functional analyses provide insights into SLAC1 gating and activity modulation. These findings allow us to propose a mechanism for finely tuned SLAC1 control of the stomatal pore in response to environmental stimuli.



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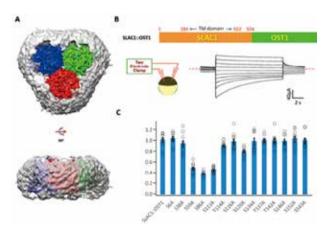


Figure: Three-dimensional cryo-EM structure of plant SLAC1 and electrophysiological analysis of phosphorylation sites. A, Three-dimensional cryo-EM structure with a resolution of 2.97 Angstroms in plant. B,Electrophysiological analysis of the fusion system of SLAC1::OST1. C,Electrophysiological analysis of SLAC1 phosphorylation site mutants.





## **Dynamic Regulation of Embryogenesis**

**Zhuo Du**, Principal Investigator. Ph.D. (2008, China Agricultural University, China). Postdoctoral researcher (2008-2009, Albert Einstein College of Medicine, USA; 2009-2014, Memorial Sloan Kettering Cancer Center, USA).

Research Interests: 1) Spatiotemporal regulation of gene expression during embryogenesis. 2) Mechanism of cell fate determination and differentiation. 3) Molecular and cellular basis of robustness and plasticity of embryogenesis

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## Extensive dynamics and diversity of cellular regulatory states during differentiation of an invariant embryonic lineage

Caenorhabditis elegans is the first multicellular organism to have its cell atlas fully mapped; however, the molecular processes underlying the cell atlas remains elusive. To model cellular state-fate relationships, we generated a protein expression atlas encompassing 266 transcription factors (TFs) in precisely lineaged embryonic cells using imaging-based single-cell analysis. Tracing the dynamics of cellular states as defined by combinatorial TF expression reveals intricate state-fate relationships. Cellular states change continuously but at a varying speed across lineage and time. Intriguingly, a cell's trajectory from initial to terminal states undergoes extensive detours in which state transitions are generally not directional towards the terminal state but take a "winding" path, except upon terminal differentiation. Single-cell analysis also discovers unexpected diversity of cellular states. When producing cells constituting a tissue, cellular states from multiple lineages converge but exhibit intra-tissue diversities according to lineages, shaped by lineage-restricted expression of tissue-specific TFs. Furthermore, TFs exhibit widespread transient expression that increases

the temporal state diversity and fate complexity. We constructed spatiotemporal cascades and identified new functions of TFs in neurogenesis and converging muscle differentiation, demonstrating high functional relevance and utility. Collectively, we present a valuable protein atlas and unravel complex statefate relationship during metazoan embryogenesis.

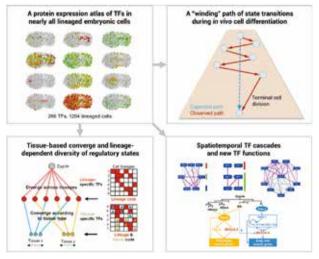




Figure: Complex state-fate relationships and spatiotemporal TF cascades during *C. elegans* cell lineage differentiation.

## **Quantitative Functional Genomics**

Wenfeng Qian, Principal Investigator, Ph. D. (2012, University of Michigan, USA).

Research Interests: The long term goal of our lab is to understand basic rules in genome evolution with both high-throughput experimental and computational approaches. There are four major ongoing research topics: 1) translational regulation and protein homeostasis; 2) the construction of fitness landscape and adaptation mechanisms during the climbing of such landscape; 3) directed evolution and how epistasis affects evolutionary trajectory; 4) cell lineage tracing during the development of multicellular organism.

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## Single-cell transcriptome analyses recapitulate the cellular and developmental responses to abiotic stresses in rice

The metabolism, environmental adaptation, and reproduction of plants depend on the division of labor among individual cells. However, cells with various functions are often studied as a bulk where their specificities cannot be distinguished. Based on the large-scale single-cell transcriptome method that can assist in identifying cell types, the functional heterogeneity of different types of cells can be explored. Here, we apply a barcode-based single-cell RNA sequencing to the aerial organs and the underground root tip of rice seedlings grown in four environments (control, low-nitrogen, iron-deficiency, and high-salinity). We capture the transcriptomes of ~200 thousand cells and identify all major cell types, particularly classify the subpopulations of mesophyll cells in monocot for the first time. We observe significant cell-type heterogeneity in gene expression changes, clarifying the specific roles of each type of cell populations, which can be explained by the differences in development through reconstructing trajectories of cell differentiation. Collectively, our study represents a benchmark-setting data resource of single-cell transcriptome atlas for rice seedlings and an illustration of exploiting such resources to drive discoveries in plant biology.



### Publications

Zhao, T.L., Zhang, S., and Qian, W.F. (2020). Cisregulatory mechanisms and biological effects of translation elongation. Yi Chuan, 42 (7): 613-631.

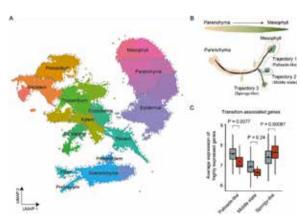


Figure: Single-cell transcriptome atlas of rice. (A) Dimension reduction and clustering map of rice cells. Each dot stands for a cell. Colored cell clusters in different regions indicate different cell types. (B-C) Abiotic stress alters the differentiation of cell populations. Reconstruction of cell differentiation trajectory from parenchyma towards mesophyll cells in rice. Each dot stands for a cell, and the direction of arrows indicates the path of cell differentiation (B). The expression levels of transitionassociatedgenes are changed in abiotic stress-treated samples, explaining the delayed development towards mesophyll cells (C). *P* values are given by the Mann-Whitney U tests.





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- Li, Y., Liu, Y., Yang, H., Zhang, T., Naruse, K., and Tu, Q. (2020). Dynamic transcriptional and chromatin accessibility landscape of medaka embryogenesis. Genome Research, 30 (6): 924-937.

## Gene Regulatory Networks Controlling Development and Regeneration

**Qiang Tu**, Principal Investigator, Ph.D. (2003, Institute of Biochemistry and Cell Biology, SIBS, CAS, China); Postdoctoral scholar, Senior research fellow and Professional Staff (2004-2014, California Institute of Technology, USA).

Research Interests: Our lab is interested in developmental systems biology. We are striving to understand how thousands of genes logically interact with each other to drive various development and regeneration processes. Currently we focus on resolving gene regulatory networks that control sex determination using medaka fish as the model, and heart regeneration using zebrafish as the model.

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## Integrative single-cell RNA-seq and ATAC-seq analysis of ovarian germline stem cell development

Germ cells transfer genetic information from one generation to the next. In mammalian testes, germline stem cells (GSCs) produce sperms continually, while there is no direct evidence for the presence of GSCs in mammalian ovaries. Some lower vertebrates with high fecundity, such as fish and amphibia, possess GSCs in their mature ovaries. Medaka is the first vertebrate in which ovarian GSCs have been identified. Therefore, the medaka ovary provides an important model system for studying vertebrate GSC development. Currently, little is known about molecular regulatory mechanisms underline this process, except a few marker genes have been identified. In this study, we systematically profiled the gene expression and chromatin accessibility of the medaka ovary using single-cell RNA-seq and single-cell ATAC-seq technologies. We were able to distinguish all cell types along the ovarian GSC developmental trajectory. For each cell type, we identified specifically expressed genes and specifically active open chromatin regions. By integrating two single-cell data sets, we inferred the gene regulatory network underlining the developmental trajectory of ovarian GSCs. Finally, we discovered multiple transcription factors and associated *cis*-regulatory elements, which could be the master controls of the GSC maintenance, meiosis initiation, and oocyte differentiation. In short, the integration of double single-cell omics data sets described the gene expression and genomic regulatory landscape of ovarian GSC development, shed light on the big picture of regulatory mechanisms underline this process.

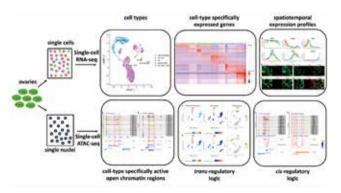




Figure: Overview of the study. The medaka ovary sample was profiled by single-cell RNA-seq and single-cell ATAC-seq. The integration of two data sets revealed all cell types during ovarian germline stem cell development, identified cell-type specifically expressed genes, active open chromatin regions, and inferred *trans-/cis-* regulatory logic as well.

### **Functional Proteomics**

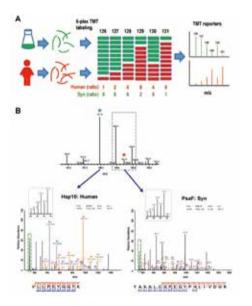
**Yingchun Wang**, Principal Investigator, Ph.D. (2003, Genetics/Bioinformatics and Computational Biology, Iowa State University, USA).

Research Interests: The current focus of our research is to discover novel mechanisms that regulate cell polarization and pseudopodia dynamics of migrating cells using functional proteomics, and to identify new proteins and protein modifications that are critical for cell polarization and migration. The current focus is to quantitatively identify proteins on different membranes, including outer membrane, cytoplasmic membrane, and thylakoid membrane, of a model photosynthetic organism *Synechocystis sp.* PCC 6803. We will build a membrane proteome atlas for this organism. The same approach and workflow could also be applied for creating protein atlas for other organisms.

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## Evaluation of the potential risk of advanced peak determination in distorting isobaric labeling-based single-shot proteome quantitation

The recent development and implementation of the Advanced Peak Determination (APD) algorithm with MS instrument dramatically increased the sampling of low abundance features for MS/MS fragmentation. After in-depth evaluation, it is found that with APD on, many chimeric spectra are acquired through co-fragmentation of high abundance contaminants with low abundance targets, and such co-fragmentations are largely avoided when APD is off. To



evaluate whether such a co-fragmentation could significantly distort the accuracy of the isobaric-labeling based quantitation of the low abundance target, a singleshot TMT experiment is performed using a two-proteome model, whereby each TMT channel contains premixed peptides from human and a cyanobacterium with a known ratio. Unexpectedly, it is found that APD does not significantly distort TMT ratios, probably because the majority of the APD-specific chimeric spectra are not identifiable. Nevertheless, a few examples of significant distortion of TMT ratios of low abundance peptides caused by APD is found through manual inspection, and suggests that APD should be off in a single-shot TMT experiment to avoid the laborious and timecosting manual inspection.



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Figure: Evaluation of the effect of APD on the TMT-based quantitative proteomics. (A) The diagram shows the strategy to evaluate the effect of APD on the distortion of TMT ratios. The two-proteome model (human proteome and *Synechocystis* proteome) was used in the experiment, whereby the tryptic peptides from the two proteomes were TMT-labeled and mixed with designated ratios in each TMT channels as indicated. Peptides in all channels were then mixed with an equal ratio and analyzed with LC-MS/MS. (B) An example showing the significant distortion of the TMT ratio caused by APD. A tryptic peptide from the *Synechocystis* protein PsaF (precursor, red star) was co-isolated and co-fragmented with a higher abundance peptide from the human protein Hsp10 (contaminant, green star) when APD is on. Note that the distribution pattern of the TMT reporters of the PsaF peptide resembles that of the Hsp10 and is actually reversed from the theoretical distribution pattern for *Synechocystis* protein (A). The peptide was not isolated for fragmentation when APD was off.





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## **Bioinformatics and Systems Biology**

#### Xiujie Wang, Principal Investigator, Ph.D. (2004, The Rockefeller University, USA).

Research Interests: The current focus the lab is to develop novel computational methods to analyze the fast increasing genomics, transcriptomics, proteomics and other large-scale biological data, to identify new non-coding regulatory RNA genes in eukaryotic genomes, to decipher their transcription regulatory mechanisms, and to study the functions of non-coding RNAs in regulating embryonic stem cell pluripotency and targeted differentiation of embryonic stem cells towards defined organs.

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## Potential inhibitors aganist 2019-nCoV coronavirus M protease from clinically approved medicines

Starting from December 2019, a novel coronavirus, which was later named 2019-nCoV ('n'stands for novel), was found to cause Severe Acute Respiratory (SARI) symptoms, including fever, dys-pnea, asthenia and pneumonia among people in Wuhan, China. With the hope to identify candidate drugs for 2019-nCoV, we adopted a computational approach to screen for available commercial medicines which may function as inhibitors for the M<sup>pro</sup> of 2019-nCoV. Based on the structural information of clinical effective medicines for 2019-nCoV, we have predicted a list of commercial medicines which may function as inhibitors for 2019-nCoV, we have predicted a list of commercial medicines which may function as inhibitors for 2019-nCoV by targeting its main protease M<sup>pro</sup>. Compared to lopinavir/ritonavir, most of these predicted drugs could form more hydrogen bonds with 2019-nCoV M<sup>pro</sup>, thus may have higher mutation tolerance than lopinavir/ritonavir. It should be noted that these results were obtained solely by in silico predictions, further experiments are needed to validate the efficacy of these drugs. The binding pockets of these drugs on M<sup>pro</sup> are conserved between SARS-CoV M<sup>pro</sup> and 2019-nCoV M<sup>pro</sup>, indicating the potential of these drugs as inhibitors for other coronaviruses with similar M<sup>pro</sup> binding sites and pocket structures.

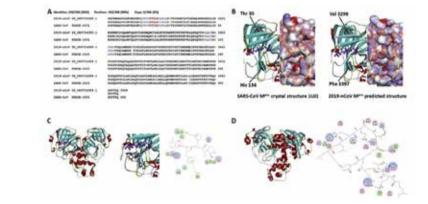


Figure: Screen for potential 2019-nCoV M<sup>pro</sup> inhibitors from commercial medicines.



The center's strategy is at ensuring the national demands on grain yield, water resources and ecological protection, focusing on the high efficient use of agricultural water resources; developing the innovative ecological resource theory and the modern agricultural technology systems for resource preservation; strengthening technological achievements transformation and service to society; gradually making the center to be an internationally well-known innovative institution.

The Center for Agricultural Resources Research has five key laboratories (i.e Key Laboratory for Agricultural Water Resources, CAS, Engineering Laboratory of Efficient Utilization of Saline-alkali Soils Resources, CAS, Hebei Key Laboratory of Water-saving Agriculture, Hebei Key Laboratory of Soil Ecology and Hebei Engineering Laboratory of Breeding and Germplasm Innovation for Plant Stress Tolerance). The center has five field stations (i.e Luancheng Agro-ecosystem Experimental Station, CAS, Nanpi Eco-agricultural Experimental Station, CAS, Taihang Mountain Mountain Ecosystem Experimental Station, CAS, Nandagang Ecological Research Station of Coastal Wetland, CAS and Saibei Ecology Experimental Station, CAS). There are three main research fields in the center (i.e water resource and water-saving agriculture, ecology and environment, and plant genetics and breeding). There are four research laboratories (i.e water resource and water-saving agriculture, ecology and environment, plant genetics and breeding, regional agriculture and agricultural policy). Now, the center has one professor from "Thousand Young Talents Program of the China's government", two professors from "National High-level Personnel of Special Support Program" and eight professors from the "Hundred Talents Program of CAS". In 2020, the center has achieved significant progresses in farmland hydrological process and high efficient use of agricultural water resources, crop germplasm selection and breeding, ecosystem process and management.

Water resources and water-saving agriculture: Yanjun Shen's group integrated water consumption management measures of "top-down" at county scale and "bottom-up" at field scale, and proposed County Agricultural Water Consumption Management System. Yonghui Yang's team recommend that further improvement on retrieval algorithm is needed by considering topographical impacts for satellite precipitation products in the future. Xiying Zhang's group found that winter wheat-summer maize cropping system under minimum irrigation had the potential to sustain the groundwater balance in the region North China Plain now and under climate change. Mengyu Liu's group found that 20-30 m<sup>2</sup> was the most water-saving and high-efficiency plot specification for surface irrigation. Shiqin Wang's group found that accumulated high nitrate in surface soil in a normal year and leaching after extreme precipitation lead to increased nitrate concentration in groundwater. Hongyong Sun's group found that 10 days after physiological maturity is the proper time to conduct mechanical grain harvesting for spring maize in the North China. Yanjun Shen's Group developed a high resolution net surface radiation dataset , based of which the spatio-temporal characteristics of net surface radiation over mainland China was revealed from 1982 to 2017. Xiuwei Liu's group found that stable monomer compound isotope can more accurately indicate the conclusion of crop water use efficiency. Baodi Dong's group found that low light led to an increase in the number of regenerated effective tillers, and further expand glume opening angle of spikes for accepting external pollens.

**Plant genetics and breeding:** Junming Li's group found that *QMrI-7B* confers wheat upon larger root system which is favorable to nitrogen uptake and grain production. Diaoguo An's group identified 17 stable QTLs related to kernel traits which can be used for further positional cloning and marker-assisted selection in wheat breeding programs. Zhengbin Zhang's group demonstrated that shortening the sgRNA-DNA interface enables SpCas9 and eSpCas9(1.1) to nick the target DNA strand and the system provided a novel tool for achieving trait stacking in plants. Dongping Lv's group revealed that miR172b-TOE1/2 module regulates plant innate immunity in an age-dependent manner.

**Ecology and environment:** Chunsheng Hu's group found that the conversion of native saline-alkaline grassland with low N to Tamarix or cropland is likely to result in increased soil N<sub>2</sub>O emission to the atmosphere and also contribute significantly to the depletion of the <sup>15</sup>N isotopic composition of atmospheric N<sub>2</sub>O. Xiaojing Liu's group found that local desalination could alleviate the salt stress of *Hibicus* moscheutos. Jintong Liu's group delimited the Farming-grazing Transitional Belt with the county boundaries in north China, considering irrigation as one of the key factors. Lin Ma's group proposed 10 key elements of green agricultural development and five "one" scientific and technological projects for agricultural green development. Binbin Liu's group found that phytoremediation significantly changed the soil microbial community characteristics of archaea, bacteria and fungi, which exploring the mechanisms of plant-micribobe interaction on amelioration of saline-alkali soil. Feng Zhu's group explored the functions of microorganisms and their potential plant-microbe interaction mechanisms by using integrated approach. Jiansheng Cao's group found that the conversion of grassland to construction land had the largest contribution rate to the decrease of ESV while the conversion of grassland to waters had the largest contribution rate to the upstream of Xiong'an New Area. Lipu Han's group developed a new type of pelletized preparation which was through the cascade pelleting process under the mixed experimental design method. Zhaohai Bai's group provided that spatial planning was needed to drastically reduce nitrogen and phosphorus surplus in China's agriculture.

In addition, the center was awarded a first prize of Science and Technology Progress Award of Hebei Province and a second prize of Natural Science Award of Hebei Province; published 129 papers, among which 85 were included in SCI Journals; granted 27 patents; draw 4 local standard and 1 industry standard; registered 3 softwares in 2020.



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## Genetic Improvement & Germplasm Enhancement in Wheat

Diaoguo An, Principal Investigator, Ph.D. (2006, Research Center for Ecoenvironmental Sciences, CAS, China).

Research Interests: The laboratory is mainly interested in the development of new wheat germplasm resources by molecular chromosomesengineering; identifying, fine mapping and cloning of important genes/QTLs for disease resistance and important agronomic traits; molecular design breeding.

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## Identification and validation of quantitative trait loci for kernel traits in common wheat (*Triticum aestivum* L.)

Kernel weight and morphology are important traits affecting cereal yields and quality. In this study, we performed quantitative trait loci (QTL) analyses using recombinant inbred lines derived from the cross 'PB3228 × Gao 8901' (PG-RIL) to dissect the genetic basis of kernel traits. A total of 17 stable QTLs related to kernel traits were identified, notably, two stable QTLs *QTkw.cas-1A.2* and *QTkw.cas-4A* explained the highest phenotypic variance for thousand kernel weight and kernel length (KL), and other two stable QTLs *QTkw.cas-6A.1* and *QTkw.cas-7D.2* and *QKw.cas-7D.2* contributed greater effects on kernel width (KW). The QTLs *QTkw.cas-7D.2* and *QKw.cas-7D.1* associated with TKW and KW were delimited to the physical interval of approximately 3.82 Mb harboring 47 candidate genes. Among them, the candidate gene *TaFT-D1* had a 1 bp insertions/deletion (InDel) within the third exon, which might be the cause of the diversity in TKW and KW between the two parents. A KASP marker of TaFT-D1allele was developed and verified by PG-RIL and a natural population consisted of 141 cultivar/lines. It was found that the favorable *TaFT-D1* (G)-*allele* has been positively selected during Chinese wheat breeding. Thus, these results can be used for further positional cloning and markerassisted selection in wheat breeding programs.



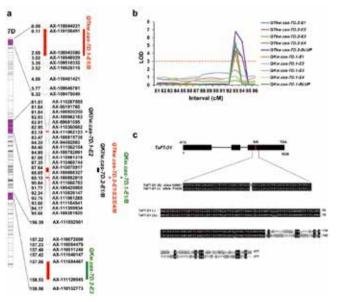


Figure: Distribution of QTLs for kernel-related traits on 7D chromosomes and prediction of candidate gene for QTkw.cas-7D.2 and QKw.cas-7D.1

## Sustainable Development of Agriculture and Livestock System

Zhaohai Bai, Young Investigator, Ph.D. (2015, China Agiculture University)

Research Interests: Mainly focus on the research of multi-index quantitative evaluation of agricultural and livestock systems, and nutrient management of livestock manure and urine.

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## Spatial planning needed to drastically reduce nitrogen and phosphorus surplus in China's agriculture

China's fertilization practices contribute greatly to theglobal biogeochemical nitrogen (N) and phosphorus (P) flows, which have exceeded the safe-operating space. Here, we quantified the potentials of improved nutrient management in the food chain and spatial planning of livestock farms on nutrient use efficiency and losses in China, using a nutrient flow model and detailed information on >2300 counties. Annual fertilizer use could be reduced by 26 Tg N and 6.4 Tg P following improved nutrient management. This reduction N and P fertilizer use would contribute 30% and 80% of the required global reduction, needed to keep the biogeochemical N and P flows within the planetary boundary. However, there are various barriers to make this happen. A major barrier is the transportation cost due to the uneven distributions of crop land, livestock, and people within the country. The amounts of N and P in wastes and residues are larger than the N and P demand of the crops grown in 30% and 50% of the counties, respectively. We argue that a drastic increase in the recycling and utilization of N and P from wastes and residues can only happen following relocation of livestock farms to areas with sufficient cropland.

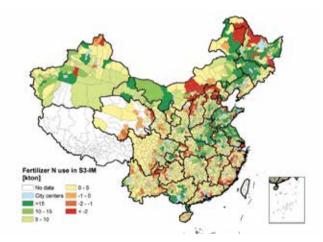


Figure: Fertilizer N demand in improved whole food system strategy



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## Mountain Ecological Hydrological Process and Regulation Mechanism

Jiansheng Cao, Principal Investigator, Ph.D. (2012, University of Chinese Academy of Sciences, China), Deputy Head of Ecological Station in Taihang Mountain.

Research Interests: 1) Ecological hydrological process on slope for the rocky mountain area in North China. 2) Mechanism of flood and drought disastersfor the rocky mountain area in North China. 3) Improvement of water conservation and regulation technology of rainfall. 4) Degraded mountain ecological restoration.

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## The impact of land use change on ecosystem service value in the upstream of Xiong'an New Area

The development of the Xiong'an New Area is a crucial strategy for the next millennium in China. The ecosystem upstream of the Xiong'an New Area, serving for the development of the Xiong'an New Area, changed with land use changes. To analyze the contribution rate of the land use change to the ecosystem service value, we analyzed the land use changes of three small watersheds (7318.56 km<sup>2</sup>) upstream of the Xiong'an New Area based on a total of six phases of land use data from 1980 to 2015. Then, the ecosystem service value (ESV) was calculated using the equivalent factor method. The results showed that the construction land and arable land were the largest land use types that increased and decreased in the area of the study area, respectively. The grassland and construction land were the land use types with the largest transfer-out area and transfer-in area, respectively. The regulating services accounted for the largest proportion of total ESV among different ecosystem service functions, and the grassland and woodland accounted for the largest proportion of total ESV among different land use types. ESV in the study area fluctuated slightly from 1980 to 2015. The decrease of ESV mainly occurred in the surrounding areas of mountain towns, and the conversion of cultivated land to construction land was the main reason for the decrease of ESV in this area. The conversion of grassland to construction land had the largest contribution rate to the decrease of ESV in the study area, while the conversion of grassland to waters had the largest contribution rate to the increase of ESV. In conclusion, controlling the development of construction land and strengthening water resources' protection may improve the ecosystem service value in the upstream Xiong'an New Area in the future.



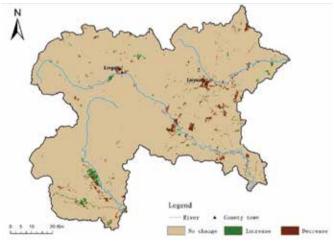


Figure: Spatial distribution map of ecosystem service value changes

## **Regulation Mechanisms and Technologies for Highly Efficient and Eco-friendly Water Use of Crops**

Baodi Dong, Young Investigator, Ph.D. (2008, University of Chinese Academy of Sciences)

Research Interests: Physio-ecological regulation mechanisms; Water saving technology; Rainfed agriculture; Improving crop water use efficiency.

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### Effects of low light in early reproductive stage on the grain number of wheat (*Triticum aestivum* L.) and the responding mechanism of wheat plants

Under the background of low light stress caused by global climate change, different wheat cultivars with low light tolerance were subjected to different degrees of low light stress at the early stage of reproductive growth. The results showed that low light reduced photosynthesis, significantly decreased the soluble sugar content in different organs, increased the ABA content of spike, inhibited the transport of nutrients to spike coordinated by IAA, caused the development stagnation of spike, thus the limited assimilate could be transported to the spike for development needs. Grain number was reduced by 1.7%-72.7% under low light. However, the respond of crops to low light by self-regulation, it was mainly by that low light led to an increase in the number of regenerated effective tillers, and further expand glume opening angle of spikes for accepting external pollens. Under low light for 1 day to 7 days, the total tiller number was increased by 21.1%-123.9%, grain umber was increased by 4.3%-81.4%. This eventually alleviated the adverse effect of decreased grain number.



### Publication

Yang, H., Dong, B., Wang, Y., Qiao, Y., Shi, C., Jin, L., and Liu, M. (2020). Photosynthetic base of reduced grain yield by shading stress during the early reproductive stage of two wheat cultivars. Scientific Reports, 10 (1): 14353.

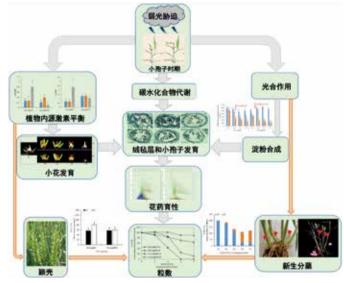


Figure: Proposed model in physiological pathway response to YM-stage shading stress in wheat





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## **Ecological Processes and Resource Utilization of Marginal Land**

### Lipu Han, Young Investigator, Ph.D. (2011, China Agricultural University).

Research Interests: we are focus on: (1) Ecological improvement mechanism of the interaction between saline-alkali lands and tolerant plants; (2) Research on key techniques of planting forage and energy plants; (3) Study for ecological response mechanism and sustainable development of forage and energy plants in sandy wasteland land; (4) Development and utilization for waste resource and plant residues in wetland-saline-alkaline land-farmland ecosystem.

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## Evaluation and optimization of the formulation selection of sorghum hybrid sudangrass seed pelletizing preparation

In this study, the method of mixture experiment design combined with seed pelleting technology was used to design {3,3} simplex center of gravity design with polymer absorbent resin and composite inert materials (two or more) as material factors, and seven pelleting formulations were obtained for pelleting and coating of Sorghum Hybrid Sudangrass seeds. Using 14 indexes related to physical characteristics, germination ability and agronomic characters of pelleted seeds, combined with the establishment of mixture regression model, analysis of variance of regression model, analysis of contour map and 3D diagram, and comparison of measured and predicted values of indexes, the optimal proportion and response value of high molecular absorbent resin and composite inert material compound formula were predicted and an optimized formula which is beneficial to improve the quality (y) of pelleted seeds of Sorghum Hybrid Sudangrass was gained. When seeds are treated by pelleting, on the one hand, the components in the preparation will form a porous film on the surface of the seed, and the effective / specific components and inert materials are connected together through the pelleting agent membrane net. After the seeds are sown into the soil, the membrane network will absorb water and expand without cracking, forming a protective barrier around the seeds, thus preventing the invasion of external microorganisms; on the other hand, the seeds germinate at the seedling growth stage, the active components in pelleting preparation gradually contact with seeds and their adjacent soil through the gap of membrane net and membrane net itself, and then participate in the physiological and biochemical process of plant growth and continue to play a role. Thus, it can provide

the necessary nutrients for seeds, promote seed germination and improve the ability of resistance to stress.



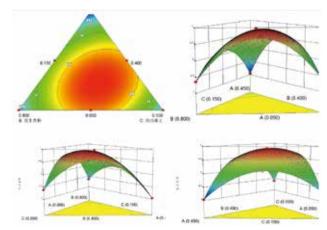


Figure: Contour map and 3D diagram of the interaction of various material factors on comprehensive quality.

## Agrosystem Nutrient Cycling and Environmental Impacts

Chunsheng Hu, Principal Investigator Ph.D. (1996, Shenyang Institute of Applied Ecology, CAS, China).

Research Interests: The in situ processes and mechanisms of water and nitrogen leaching are studied using the isotope tracing technology and modeling. The study can provide scientific evidence for adopting reasonable water and fertilizer management strategies. Nitrogen nutrition diagnosis and transformation of middle-yielding and low-yielding lands are studied. The preferred fertilization plan is acquired based on the long-term fertilizer application experiments for the sustainable land use in northern China.

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### Nitrogen isotopic signatures and fluxes of $N_2O$ in response to landuse change on naturally occurring saline-alkaline soil

The conversion of natural grassland to semi-natural or artificial ecosystems is a large-scale land-use change (LUC) commonly occurring to natural saline-alkaline land. So, this study assesses the impact of LUC on N<sub>2</sub>O emission and <sup>15</sup>N isotopic signatures of N<sub>2</sub>O emitted from naturally occurring saline-alkaline soil when changing from natural grassland (*Phragmites australis*) to seminatural (*Tamarix chinensis* (Tamarix)) and artificial cropland (*Gossypium* spp). The natural grassland and semi-natural ecosystems weren't subject to any management practice and anthropogenic nitrogen (N) addition, while the cropland received. Overall, median N<sub>2</sub>O flux was significantly different among the ecosystems with the highest from the cropland, intermediate from the Tamarix and the lowest from the grassland ecosystem. The <sup>15</sup>N isotopic signatures of N<sub>2</sub>O emitted from the soil were also significantly affected by the LUC with more depleted from cropland and less depleted when emitted from grassland soil. Our results suggest that the conversion of native saline-alkaline grassland with low N to Tamarix or cropland is likely to result in increased soil N<sub>2</sub>O emission to the atmosphere and also contribute significantly to the depletion of the <sup>15</sup>N isotopic composition of atmospheric N<sub>2</sub>O.

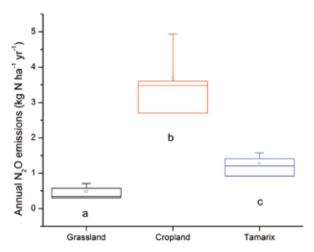


Figure: Box plot for annual  $N_2O$  emissions from three different ecosystems (*n*=4). Different letters indicate significant difference and square represents mean values.



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- Song, L., Zhao, H., Zhang, Z., Zhang, S., Liu, J., Zhang, W., Zhang, N., Ji, J., Li, L., and Li, J. (2020). Molecular cytogenetic identification of wheat-Aegilops Biuncialis 5M disomic addition line with tenacious and black glumes. International Journal of Molecular Sciences, 21 (11): 4053.
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## Wheat Molecular Breeding and Genetics

Junming Li, Principal Investigator.

Research Interests: Fine-maping QTL of yield- and root-related traits and identifying the foverable alleles; Developing novel germplasm through exploitation of alien resistance genes and breeding new varieties with marker-assisted selection.

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## Fine mapping and characterization of *QMrI-7B*, a major QTL controlling the maximum root length in wheat

Root is the major organ for plant to absorb water and mineral nutrients. On the basis of identifying a major QTL *QMrl-7B* controlling the maximum root length in wheat, we developed a set of near isogenic lines from a residual heterozygous line KJ-RIL239. With the help of BSR-seq analysis, markers were designed to draw the fine map of *QMrl-7B*. Eight recombinants in the target region were selected and *QMrl-7B* was narrowed down to the physical distance of 0.22 Mb using the secondary mapping population. According to KN9204 genome sequence assembly, there are 6 genes annotation in the *QMrl-7B* target interval. Among these candidate genes, *TraesKN7B01HG37540*, corresponding to *TraesCS7B02G339100*, was differentially expressed between the parents as well as the near isogenic lines. The effects of *QMrl-7B* were dissected under both low and high nitrogen environments. Trials in two consecutive growing seasons showed that *QMrl-7B* confers wheat upon larger root system which is favorable to nitrogen partial factor productivity. This research provides a fundamental basis for molecular improvement of root traits.

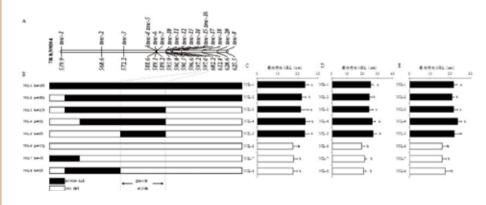




Figure: Physical map, genotypes and phenotypes of the NILs within *QMrI-7B* target interval based on KN9204 genome sequence assembly Notes: Fig. A indicates the physical map of target interval in KN9204; Fig. B indicates the genotype of the NILs, in which the black indicates the genotype of KN9204 and the white indicates the genotype of J411; NIL-1 ~ 8 indicate different recombinants and "n" indicates the number of the recombinants; Fig. C ~ E indicate the MRL of the recombinants growing at vermiculite culture for 10 days, low and normal nitrogen hydroponic culture for 14 days, respectively; Different lowercases upon them indicate significant differences at p<0.05.

## **Microbial Ecology**

Binbin Liu, Principal Investigator, Ph.D. (2006, Shanghai Jiao Tong University, China).

Research Interests: Dr. Liu's laboratory is mainly interested in using high throughput methods to investigate the microbial processes, focusing on soil microbial diversity, functional gene dynamics and expression related to nutrient turnover in agricultural ecosystems.

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## Long-term phytoremediation of coastal saline soil reveals plant species-specific patterns of microbial community recruitment

Soil salinization is one of the major land degradation processes that decreases soil fertility and crop production worldwide. In this study, a long-term coastal saline soil remediation experiment was conducted with three salt-tolerant plant species: Lycium chinense Mill. (LCM), Tamarix chinensis Lour. (TCL) and Gossypium hirsutum Linn. (GHL). All three plants successfully remediated the saline soil but showed different efficacies. The archaeal, bacterial and fungal communities in barren soil and in four rhizocompartments of the three plants were assessed. All three plants significantly decreased the richness of the archaeal communities but increased that of the bacterial and fungal communities in both the rhizosphere and rhizoplane compared with those in the barren soil. The archaeal and bacterial community structure was strongly influenced by the rhizocompartment, while specific fungal communities were recruited by different plant species. Soil electrical conductivity (EC) was identified as the main factor driving the variation in microbial community composition between the remediated and barren soil, and total nitrogen (TN), total carbon (TC), and available potassium (AK) were the main factors driving the differences among plant species. This study provides new insights into the responses of the root zone microbial communities of different salt-tolerant plant species during phytoremediation.

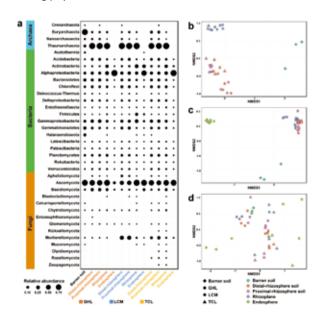


Figure: The composition of archaeal, bacterial, and fungal communities in the barren soil and in the four rhizocompartments of the three salt-tolerant plants (a). NMDS plot of archaeal (b), bacterial (c), and fungal communities (d) based on Bray-Curtis distances.



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## Sustainable Management and Ecological Engineering for Ecosystem

**Jintong Liu**, Principal Investigator, Ph.D. (2000, Beijing Forestry University, China), Senior Visiting Scholar (1999-2000, Danish Royal University of Pharmaceutical Sciences, Denmark), Postdoctoral Fellow (2002-2004, Center for Central Agricultural Research, Japan).

Research Interests: Eco-engineering for drought prone mountain region, north China. Eco-engineering for coastal saline-alkali soil. Management for resources and ecosystem information.

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## Definition of agro-pastoral ecotone in North China based on irrigation corrections

Agro-pastoral ecotope in North China is a typical ecologically fragile zone. Scientific and reasonable definition of this ecotone is of great significance to the sustainable development of regions based on resource utilization. The water condition is the most basiccondition of the definition of agro-pastoral ecotone in North China . The previous studies used precipitation as the water condition and did not consider the impact of irrigation on the formation of the agropastoral ecotone. Based on the traditional method, this research adds irrigation corrections and uses the corrected water condition as a definition indicator, combined with the water variability and dryness indicators, to define the agro-pastoral ecotone in North China with the support of the fuzzy comprehensive evaluation method. After that, the research further combined the administrative boundaries to divide the counties in the agro-pastoral ecotone. The results show that the agro-pastoral ecotone in North China presents a banded distribution of northeast-southwest direction, with a total area of 659 000 km<sup>2</sup>; there are 123 counties (cities, banners) in the agro-pastoral ecotone in North China, with a total area of 660 800 km<sup>2</sup>, distributed in 9 Provinces (autonomous regions) in North China. The number of counties in the agropastoral ecotone is the most in Inner Mongolia Autonomous Region, Shanxi Province and Qinghai Province, and the total area of the counties in the agro-pastoral ecotone is the largest in Inner Mongolia Autonomous Region. The results of this study shifts slightly northwest in position from the traditional result, and extends northeast and southwest respectively in scope. Three provinces, Heilongjiang, Jilin, and Qinghai were added, and Liaoning province was removed. There has been a decrease in the total number of counties. The total number of counties (cities, banners) in the agro-pastoral ecotone in Heilongjiang, Jilin, Inner Mongolia, and Qinghai province has increased, while the total number of counties (cities) in the agropastoral ecotone in Hebei. Shanxi, Shaanxi, Gansu and Ningxia province reduced. And some counties in the Inner Mongolia Hetao Irrigation District and Ningxia Hetao Irrigation District have been included. Analysis of the hydrothermal conditions in the agropastoral ecotone shows that the annual accumulated temperature in most areas of the agro-pastoral ecotone is between 2 000~3 500°C, which shows a trend of high in the middle and low in the northeast and southwest. The water conditions (precipitation and irrigation) in most areas of the agro-pastoral ecotone are in the range of 300~450mm, decreasing along the southeast-northwest direction. The content of top soil organic carbon in most areas of the agro-pastoral ecotone is between 0~1%, showing a trend of low in the middle and high in the northeast and southwest. This study incorporates irrigation indicators into the definition index system of the agro-pastoral ecotone which makes up for the shortcomings of the traditional climate definition method and is a scientific supplement and development to the definition of the agro-pastoral ecotone. The obtained county-scale agro-pastoral ecotone can provide a scientific reference for the adjustment of the agricultural-pastoral structure and precise sustainable development in the agro-pastoral ecotone if combined with the study of the distribution of hydrothermal conditions in the agropastoral ecotone.



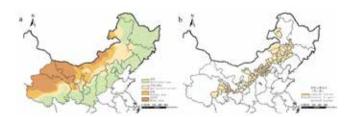


Figure: Agro-pastoral ecotone in North China (a) and distribution of counties (cities, banners) in agro-pastoral ecotone in North China (b) defined in this paper

## Efficient Water Use of Crops and Water-Saving Technology

Mengyu Liu, Principal Investigator, Ph.D. (2000, Hokkaido University, Japan).

Research Interests: Special efforts are devoted to eco-physiological mechanism of efficient water use, water saving and regulating technology, high efficient and safe production of vegetables, and relations between climate change and water use of agro-ecosystems.

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## The technology to increase water use efficiency and decrease water use amount by shrinking irrigation unit size

The high yield and high quality of winter wheat in Hebei and even the whole north China are at the expense of excessive consumption of groundwater. Improving irrigation technology is one of the important ways to save water. Besides automatic control technology, it is also a feasible method to control irrigation quantity and quality by irrigation area. In this study, the relationship between irrigation amount and irrigation unit size was determined by combining different check lengths (4, 6, 8, 10m) with check widths (2, 3, 5, 6, 8 m). Irrigation water amount increased linearly with the increase of border field area. For every 1m<sup>2</sup> increase of irrigation check area, irrigation water amount increased by about 1.667%. In production, we recommend 20-30m<sup>2</sup> (5-6m long and 4-5 m wide) as the most water-saving and efficient irrigation unit specification, where winter wheat achieved the highest water use efficiency. Beside this, the concept of "micro-flow irrigation" is put forward. Micro-flow irrigation is a kind of multi-outlet irrigation technology. Generally, the check width is 4m, the check length is 25-35m, and a hose with a certain number of outlet holes is laid in the middle of the check. The appearance of micro-flow irrigation greatly improves the practicability of the former technique.

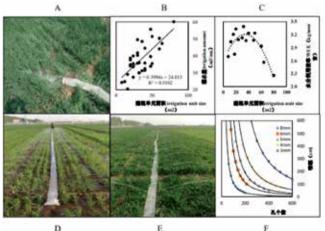


Figure: The relationship between irrigation size and irrigation amount (A-C) & the technical index and field irrigation operation of micro-flow irrigation. A: the irrigation operation; B: For every  $1m^2$  increase of irrigation check area, irrigation water amount increased by about 1.667%; C: winter wheat achieved the highest water use efficiency at the irrigation unit size around 20-30 m<sup>2</sup>. D & E: Field operation sight view. F: Under the condition of 30 m<sup>3</sup>/hr water output, the relationship between the outlet apertures, the number of outlet holes and the spray amplitude of the micro outflow water.





### Publication

Yang, H., Dong, B.D., Wang, Y.K., Qiao, Y.Z., Shi, C.H., Jin, L.L., and Liu, M.Y. (2020). Photosynthetic base of reduced grain yield by shading stress during the early reproductive stage of two wheat cultivars. Scientific Reports, 10 (1): 143-153.



## **Efficient Utilization of Saline Resources**

Xiaojing Liu, Principal Investigator, Ph.D. (2006, Tokyo University of Agriculture, Japan).

Research Interests: Reclamation and utilization of saline soils, saline water irrigation, and ecophysiology of plant salt tolerance.

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### **Publications**

- Feng, X., Tabassum, H., Guo, K., An, P., and Liu, X. (2020). Physiological, morphological and anatomical responses of Hibiscus moscheutos to non-uniform salinity stress. Environmental and Experimental Botany, DOI: 10.1016/j.envexpbot.2020.104301.
- Guo, K. and Liu, X. (2020). Salt leaching process in coastal saline soil by infiltration of melting saline ice under field conditions. Journal of Soil and Water Conservation, 75 (4): 549.
- Hussain, T., Koyro, H.W., Zhang, W., Liu, X., Gul, B., and Liu, X. (2020). Low salinity improves photosynthetic performance in *Panicum antidotale* under drought stress. Frontiers in Plant Science, 11: 481.
- Ju, Z., Sun, H., and Liu, X. (2020). Thermotime domain reflectometry to evaluate unsaturated soils contaminated with nonaqueous phase liquids. Vadose Zone Journal, 19 (1): e20016.
- Li, X., liu, X., and Liu, X. (2020). Long-term fertilization effects on crop yield and desalinized soil properties. Agronomy Journal, 112 (5): 4321.

## Physiological, morphological and anatomical responses of *Hibiscus moscheutos* to non-uniform salinity stress

In this study, swamp rose mallow (*Hibiscus moscheutos, SRM*) seedlings grown in split-root pots irrigated with different concentration of NaCl solutions were studied. Under non-uniform salinity, the total biomass, shoot height, and leaf area of *SRM* seedlings were significantly higher and the leaf Na<sup>+</sup> and Cl<sup>-</sup> contents were significantly lower than under uniform salinity. Once the parts of the roots were in the salt-free zone, the leaf water potential and the daily water consumption of the seedlings under non-uniform salinity were much higher than the uniform salinity treatment. In terms of leaf anatomical traits, that the palisade and spongy parenchyma was significantly thicker than the uniform salinity. More roots, especially fine roots, were distributed in the non-saline zone. These results indicate that under non-uniform salinity, *SRM* seedling growth was significantly inhibited, however, the salt stress was alleviated by the partial salt-free root zone compared to uniform the salinity. Non-uniform salinity led to moderate Na<sup>+</sup> accumulation in the leaves, which in turn caused slight oxidative and osmotic stress, but had little influence on the morphological or anatomical traits of the leaves. *SRM* could adapt to the saline soil where partial zone desalinated or partial zone replaced by the non-saline soil.

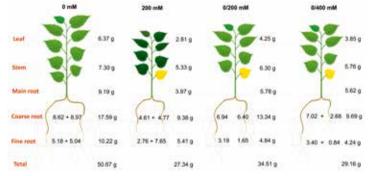


Figure: The biomass accumulation for different parts of the Swamp Rose Mallow seedlings under uniform and non-uniform salinity



## Using Compound-Specific Isotopes to Evaluate Crop Water Use Efficiency

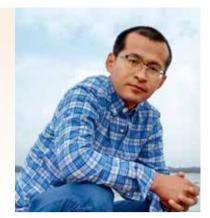
Xiuwei Liu, scientist, graduated from the Center for Agricultural Resourses Research, Institute of Genetics and Developmental Biology, CAS in 2015. He was a post-doc at Texas A&M University and Noble Research Institute from 2015 to 2019.

Research Interests: Evaluating the leaf n-alkane carbon isotope as a surrogate for crop yield and water use efficiency, Investigating the effects of root phenes on root distribution, Using electrical capacitance to detect cotton roots.

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## Dynamic changes in leaf wax n-alkanes and d13C during leaf development in winter wheat under varied irrigation experiments

Leaf wax n-alkanes serve as biomarkers that record environmental information with their carbon isotopic composition ( $\delta^{^{13}}C_{_{alk}}$ ) recording water availability. Agricultural and paleoclimate applications would benefit from a greater understanding of how water shortages are recorded in  $\delta^{13}C_{alk}$  across the growing season, and controlled experiments can contribute to understanding as they constrain cause and effect, although they represent highly simplified versions of natural ecosystems. In this study, we use fieldgrown winter wheat (Triticum aestivum L.) under three water treatments (full irrigation, deficit irrigation and dry/rain-fed) to investigate the changes of flag (top) leaf traits including leaf mass per area, wax n-alkanes and carbon composition of bulk leaf and n-alkanes ( $\delta^{13}C_{alk}$ ) during leaf development. We find that leaf wax n-alkane synthesis ceases when the leaf reaches its maximum area (fully expanded) under dry conditions. However, fully expanded leaves can still increase their n-alkane concentrations by 42% and 70% under deficit and full irrigation, respectively. Total leaf wax n-alkane concentrations and  $\delta^{13}C_{alk}$  show no difference atleaf flushbetween rain-fed (dry) and deficit irrigation treatments, but  $\delta^{13}C_{alk}$  under full irrigation is more depleted as expected, with more pronounced offsets between the experiments developing during the growing season.  $\delta^{13}C_{alk}$  decreases logarithmically during leaf development perhaps indicatinga shift from stored carbohydrate to fresh photosynthate and shoot growth related responses.Our results indicate that the water information at leaf flush is not 'locked-in' to the  $\delta^{13}C_{alk}$  signal carried by the leaf wax n-alkanes.Instead,the first flush likely reflects the signal of stored carbohydratemore so than ambient environmental conditions. Total leaf wax n-alkane concentrations and  $\delta^{13}C_{alk}$ at the mature stage carry a more complete record of differences in water use efficiency experienced during the growing season.



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Jiang, H., Feakins, S.J., Sun, H., Feng, X., Zhang, X., and Liu, X. (2020). Dynamic changes in leaf wax n-alkanes and δ 13C during leaf development in winter wheat under varied irrigation experiments. Organic Geochemistry, 146: 104054.

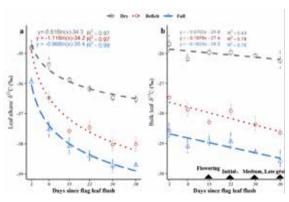


Figure: Logarithmic decrease in the leaf alkane  $\delta^{13}$ C (a) and linear decrease in the bulk leaf  $\delta^{13}$ C (b) during leaf development (key crop growing stages are marked with solid dark triangle)





- Zou Y, Wang S, Lu D. (2020) MiR172b-TOE1/2 module regulates plant innate immunity in an age-dependent manner. Biochemical and Biophysical Research Communications, 531(4):503-507.
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- Fan F, Zhang Q, Lu D. Identification of N-glycosylation sites on AtERO1 and AtERO2 using a transient expression system. Biochemical and Biophysical Research Communications, 533(3):481-485.

## The Molecular Genetics of Plant Defense

**Dongping Lv**, Principal Investigator. Ph.D. (2007, University of Hawaii, USA). Research Interests: Study on the plant pathogen resistance mechanism. Email: dplu@sjziam.ac.cn

## MiR172b-TOE1/2 module regulates plant innate immunity in an age-dependent manner.

Plant innate immunity varies with age and plant developmental stages. We reported previously that *Arabidopsis thaliana* microRNA miR172b regulates *FLS2* transcription through two transcription factors: TARGET OF EAT1 (TOE1) and TOE2. Although the flg22- triggered immune responses were investigated in 2-d-old or even younger *toe1/toe2* mutant and miR172b over expression (OE) transgenic plants, the *FLS2*-mediated immune responses in older plants remain uncharacterized yet. Here, we analyzed the flg22- triggered immune response in 6-d-old *toe1/toe2* and miR172b OE plants. We found that unlike 2-d-old plants, 6-d-old Col-0, *toe1/toe2* and miR172b OE plants exhibit comparable flg22-triggered immune responses. Strikingly, miR172b precursor in 6-d-old Col-0 plants upon flg22 treatment reached to a very high level, consequently, the TOE1/2 protein level under this condition was very low or almost undetectable, which explains why 6-d-old WT seedlings are very similar to *toe1/toe2* seedlings or miR172b OE plants with respect to the flg22-triggered immune responses. Taken together, our study reveals that miR172b-TOE1/2 module regulates plant innate immunity in an age-dependent manner.

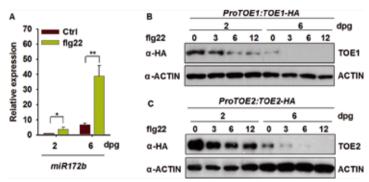
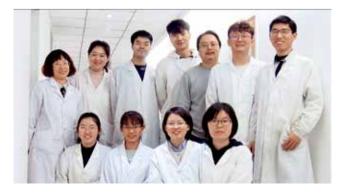


Figure: Measurement of the *pre-miR172b* and detection of TOE1/2 proteins in 2- or 6-d old seedlings treated with flg22. (A) Measurement of miR172b precursor levels. TOE1/2 protein accumulation in *ProTOE1:TOE1-HA* (B) or *ProTOE2:TOE2-HA* (C) transgenic plants. Two- or 6-d-old transgenic plants were treated with 1 µM flg22 for the indicated times.



## Agro-Ecology and Nutrient Management

Lin Ma, Principal Investigator, Ph.D. (2010, Agricultural University of Hebei, China). Postdoctoral fellow (2010-2014, Wageningen University, the Netherlands).

Research Interests: Sustainable development of agriculture and food systems by analyzing complex "soil-crop production-animal production-food processinghousehold consumption" chains, Mitigation options of non-point sources pollution from crop and animal production system, Techniques of manure management from "housing-storage-processing-application" chain.

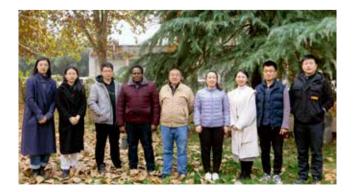
Email: malin1979@sjziam.ac.cn

## Theoretical framework and realization pathway of agricultural green development

Agricultural green development, as a novel concept, technical pattern and systematic procedure, is the new direction of China's agricultural development, but many theoretical and scientific questions about agricultural green development remain unsolved. In this study, we described the theoretical framework and the realization route of agricultural green development in China. Compared with sustainable agricultural development overseas, China's agricultural green development is considered to focus more on the synergy of "green" and "development", and emphasis the characterized as "green" led by "development" and "development" promoted by "green", which in turn poses difficult challenges for China's agricultural green development. Ten key points of agricultural green development are proposed in this paper, namely: focusing on food systems; persisting in the two themes, i.e., green and development; reinforcing the three pillars, i.e., society, economy and the ecological environment; focusing on the four interfaces, i.e., resources, production, consumption and the environment; coordinating the relationships among the five stakeholders, i.e., the government, farmers, companies, retailers and consumers; following the eight principles, which are input control, resource cycling for improving efficiency, the potential realization through integrated management, emission reduction, integration for value appreciation, diet optimization for environmental pressure alleviation, policy mechanism guarantee, and local implementation; respecting the rules of the ten processes, including materials, energy, information and value flows; innovating the regulatory measures in the seven sections of green development, i.e., policy, capital, services, technology, products, knowledge and engineering; taking the three paths of transforming towards, triggering of and the synergic advance of green development, as secured by ecological environmental control mechanisms based on green development checkpoints, the implementation of the whole industrial chain green policy and technology, the optimization and regulation of food supply and demand; and realizing these paths through the nine approaches. Eventually, the coordination of the six agriculture-related goals in the social, economic, productivity, ecological, environmental, and resource sectors will be achieved. We propose to enhance key research on agricultural green development theory, technology, and strategy. We also propose to implement the "five-one" agricultural green development technical engineering projects that promote China's agricultural green development as early as possible, namely, "one basic database, one batch of technical equipment, one set of science and technology platforms, one batch of demonstration projects and one series of think tank achievements".



Figure: The schematic diagram of agriculture green development





- Liu, L., Guo, Y., Tu, Y., Zhang, N., Bai, Z., Chadwick, D., Dou, Z., and Ma, L. (2020). A higher water-soluble phosphorus supplement in pig diet improves the whole system phosphorus use efficiency. Journal of Cleaner Production, 272: 122586.
- Liu, Z., Wang, X., Wang, F., Bai, Z., Chadwick, D., Misselbrook, T., and Ma, L. (2020). The progress of composting technologies from static heap to intelligent reactor: Benefits and limitations. Journal of Cleaner Production, 270: 122328.
- Cao, Y., Wang, X., Liu, L., Velthof, G.L., Misselbrook, T., Bai, Z., and Ma, L. (2020). Acidification of manure reduces gaseous emissions and nutrient losses from subsequent composting process. Journal of Environmental Management, 264: 110454.



## Eco-Hydrological Modelling and Water Environmental Management

Yanjun Shen, Principal Investigator, Ph.D. (2018 Friedrich Schiller Universität Jena, Germany)

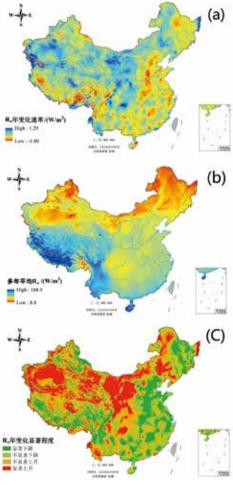
Research Interests: Eco-hydrological simulation based on hydrological models; Water quantity and quality response to agricultural activities; Agricultural water resources management.

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## Estimation of surface net radiation and its spatio-temporal variation characteristics over mainland China

 ${\sf CMFD}\ reanalysis\ dataset\ and\ {\sf GLASS}\ surface\ albedo\ dataset\ were\ used\ to\ estimate\ daily\ {\sf Rn}\ over\ mainland\ China\ during\ 1982-2017.\ {\sf GIS}\ spatial\ analysis\ technology,\ {\sf R/S}\ method\ and$ 

Mann-Kendall trend analysis method were used to reveal Rn spatiotemporal variation characteristics. The results show that: (1) Daily temperature, relative humidity and downward shortwave radiation products provided by CMFD reanalysis dataset, and estimated high spatial and temporal resolution Rn dataset have good applicability over mainland China. (2) Rn shows a slight upward trend over mainland China during 1982-2017. Rn in Northwest Inland Basin and Yellow River Basin shows an upward trend, while it in the other basins shows a downward trend with the most significant trend in Haihe Basin and Huaihe Basin. (3) The Rn interannual change is normally distributed, and the maximum Rn appears in June. (4) Rn over mainland China drops the fastest in August and rises the fastest in April. It is of great significance to the estimation of evapotranspiration, plant growth process and global climate change.



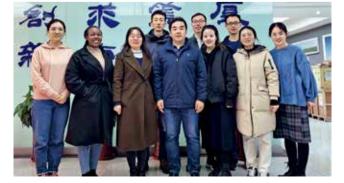


Figure: (a) Spatial distributions of mean annual Rn;(b) Spatial distributions of Rn change rates;(c) Spatial distributions of significance levels

## Agricultural Hydrology and Water Resources

Yanjun Shen, Principal Investigator, Ph.D. (2004, Chiba University, Japan).

Research Interests: Our group is mainly interested in eco-hydrological processes, especially the processes related to agricultural water use and ecological conservation. By field observation, environmental tracing, remote sensing, and numerical modeling, we focus on the water cycles at sites, watershed, and regional scales. We are also involved in the research on hydrology and water resources change and adaptation to climate warming.

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### Optimization of agricultural water consumption management based on water balance of ecosystems

Water balance characteristics of different agricultural ecosystems are important to the optimization of planting structure and green transformation under the constraints of sustainable water resources utilization in the agricultural region of North China. It is found that vegetables and fruit trees consume a large amount of water, around 900 mm, which is related to the type of facility and maturation. Evapotranspiration of winter wheat - summer maize is 715 mm, while the cotton field consumes the least water. The characteristics of irrigation and water consumption are consistent. Based on the results of water balance, by the measures such as total amount control, structural optimization and efficiency improvement, an "appropriate water" grain crop, cash crop and forage rotation allocation system with the amount of renewable agricultural water resources at the county level as the constraint has been constructed. Deficient irrigation, critical period-irrigation and dry / semi-dry farming technologies were carried out. A field water consumption management technology model with high efficiency under reasonable yield targets has been established. Integrating the water consumption management measures of the county scale "top-down" and "bottom-up" in a field scale, the agricultural water consumption management system for county - level is proposed.



- Li, C., Zhang, Y., Shen, Y., and Yu, Q. (2020). Decadal water storage decrease driven by vegetation changes in the Yellow River Basin. Science Bulletin, 65 (22): 1859.
- Liu, M., Min, L., Shen, Y., and Wu, L. (2020). Evaluating the impact of alternative cropping systems on groundwater consumption and nitrate leaching in the piedmont area of the North China Plain. Agronomy, 10 (11): 1635.
- Min, L.L., Vasilevskiy, P.Y., Wang, P., Pozdniakov, S.P., and Yu, J.J. (2020). Numerical approaches for estimating daily river leakage from arid ephemeral streams. Water, 12 (2): 19.

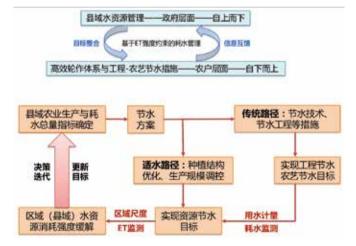


Figure: County-level agricultural water consumption management optimization system





Dong, X.L., Sun, H.Y., Wang, J.T., Liu, X.J., and Singh, B.P. (2020). Wheat-derived soil organic carbon accumulates more than its maize counterpart in a wheat-maize cropping system after 21 years. European Journal of Soil Science, 71 (4): 695-705.

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## Water and Salt Movement Process and Regulation in Agro-Ecosystems

**Hongyong Sun**, Professor, Ph.D. (2007, Institute of Geographic Sciences and Natural Resources Research, CAS), Research Fellow (2009-2010, Commonwealth Scientific and Industrial Research Organisation, Australia).

Research Interests: 1) Water and salt movement process in GSPAC; 2) Mechanism of crop adaption to water and salt; 3) Water and salt regulation of field in water shortage saline area.

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### Analysis of kernel dry down process after physiological maturity of spring maize based on diffusion theory in the North China

Moisture content of grains after physiological maturity (MCAM) is the key determinant of the quality ofmechanical grain harvesting(MGH), which can significantly improve the production efficiency of maize. Using the data of a two-year spring maize experiment conducted on 4 sites of North China with 7 common maize varieties and 3 planting densities, a model based on the diffusion theory was built. The results showed that 10 days after physiological maturity, the MCAM could be reduced to 28% in almost all circumstances and to 25% in half of the circumstances. Therefore, 10 days after physiological maturity was an appropriate time to conduct MGH for spring maize in North China. Climate and variety had significant influence on the grain moisture content at physiological maturity (M) and the moisture diffusion rate (k). ET, max-temperature, and irrigation amount at grain-filling stage had significant positive effects on K. Rainfall during the entire growth period had a significant negative effect on k. Number of husk layers and leaves had the greatest influence on M (positive effect) and k (negative effect), respectively.

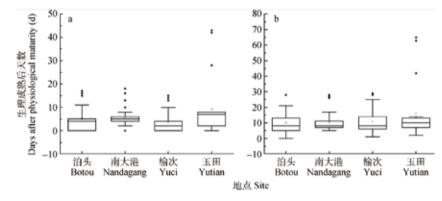


Figure: Days after physiological maturity when spring maize grain moisture drying to 28% (a) and 25% (b) at each experiment site



### Water Cycle and Groundwater Environment

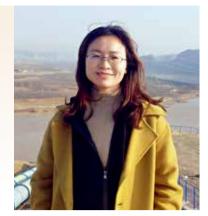
Shiqin Wang, Principal Investigator, Ph.D. (Institute of Geographical Sciences and Natural Resources Research, CAS).

Research Interests: Hydro-geochemical processes of earth critical zone; Mechanism of water cycle, and nitrate transport within unsaturated zone and saturated zone; Interaction of surface water and groundwater; Sustainable development of groundwater resources based on the water quantity and water quality simulation.

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### Nitrate accumulation and leaching potential is controlled by landuse and extreme precipitation in a headwater catchment in the North China Plain

Nitrate in groundwater is increasing in hilly areas of the world due to diverse land-uses and intensified anthropogenic activity. However, the key factors that control nitrate in groundwater as it leaches through the thin vadose zone are still poorly understood. In this study, the behavior of nitrate in the vadose zone during a normal year (2015) and a wet year (2016) were investigated in a cultivated farmland (FL) under wheat-maize double cropping and a field of natural vegetation (NV) in a headwater region of the Taihang Mountain. Water chemistry and N balances were quantified to estimate the accumulation and leaching of nitrate. Transport and fate of nitrate in the vadose zone were identified using stable isotopes of nitrate. Accumulation of NO<sub>3</sub><sup>-</sup> was mainly in the shallow layer (0–50 cm) under both NV and FL in 2015. After large rain events (20-50 mm/day) during the rainy season in 2015, the leaching rates of NO<sub>3</sub><sup>-</sup> at NV and FL sites were 86.04 and 9.61 kg/hm<sup>2</sup>, respectively. The accumulation of NO<sub>3</sub><sup>-</sup> decreased 30% and 7% at NV and FL, respectively. However, theleaching rates of NO<sub>3</sub><sup>-</sup> were 63.8 and 22.63 kg/hm<sup>2</sup>, and the accumulation of NO<sub>3</sub><sup>-</sup> decreased 84% and 43% atNV and FL after extreme precipitation in 2016, respectively. Additionally, nitrate isotopes indicated that the different sources of nitrate in soil water were due to the land-use. However, nitrate isotopes showed that increased nitrate concentrations in groundwater at two sites in 2016 were due to the extreme precipitation. Accumulated high nitrate in surface soil in a normal year and leaching after extreme precipitation lead to increased nitrate concentration in groundwater, which poses a major threat in the future. The results are critical for informing land use and water management when mitigating groundwater contamination in hilly area.



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- Yuan, R., Wang, M., Wang, S., and Song, X. (2020). Water transfer imposes hydrochemical impacts on groundwater by altering the interaction of groundwater and surface water. Journal of Hydrology, 583: 124617.
- Hou, X., Wang, S., Jin, X., Li, M., Lv, M., and Feng, W. (2020). Using an ETWatch (RS)-UZF-MODFLOW coupled model to optimize joint use of transferred water and local water sources in a saline water area of the North China Plain. Water, 12 (12).

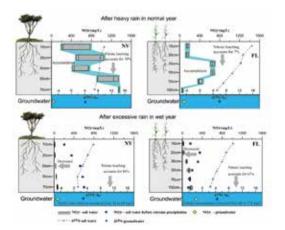


Figure: The migration mechanism in natural vegetation land and farmland after heavy precipitation in dry and wet years





- Yu, L.F., Ma, L.X., Li, H.L., Zhang, Y.Q., Kong, F.C., and Yang, Y.H. (2020). Assessment of high-resolution satellite rainfall products over a gradually elevating mountainous terrain based on a high-density rain gauge network. International Journal of Remote Sensing, 41 (14): 5620. DOI: 10.1080/01431161.2020.1734255.
- Yu, L., Zhang, Y., and Yang, Y. (2020). Using High-Density Rain Gauges to Validate the Accuracy of Satellite Precipitation Products over Complex Terrains. Atmosphere, 11 (6): DOI: 10.3390/atmos11060633.

## Water Resources and Food Security

**Yonghui Yang**, Principal Investigator, Ph.D. (2002, Chiba University, Japan), Vice Director of Center for Agricultural Resources Research of IGDB.

Research Interests: Accurate evaluation of key elements of hydrological cycle; Effect of human and climate change on hydrological cycle; Development of methods for agricultural water assessment; Land Food Water Nexus.

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## Using high-density rain gauges to validate the accuracy of satellite precipitation products over complex terrains

Topography and precipitation intensity are important factors that affect the quality of satellite precipitation products (SPPs). The objective of this study is to evaluate three SPPs—CMORPH CRT, IMERG, and TRMM 3B42V7, against a high-density network of 104 rain gauges over the Taihang Mountains. The results show that three SPPs slightly overestimate daily precipitation, compared to rain gauge observations. In terms of accuracy, 3B42 slightly outperforms CRT and IMERG. As for different elevation ranges, three SPPs show better performance in terms of accuracy in low and moderate elevation (0–500 m) regions. Better precipitation detecting performance of three SPPs was discovered in high-elevation (>1000 m) regions. In the accuracy statistical metrics, the errors of 3B42 and CRT showed significantly positive correlations with elevation variations. Three SPPs slightly overestimated the frequency of heavy rain events; CRT and 3B42 tended to underestimate the frequency of no rain events, while IMERG generally overestimated the frequency of no rain events, but also recommend that further improvement on retrieval algorithm is needed by considering topographical impacts for SPPs in the future.

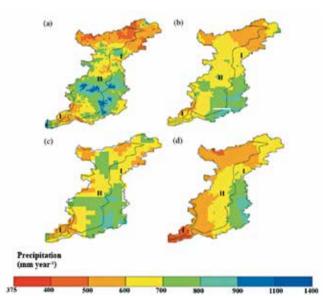




Figure: Spatial variability of the average annual total rainfall in 2016 and 2017 over the TaihangMountains as measured by (a) CMORPH CRT, (b) IMERG, (c) 3B42, and (d) rain gauges.

## **Improving Farmland Water Use Efficiency**

Xiying Zhang, Principal Investigator. Ph.D. (2006, University of Tokyo, Japan).

Research Interests: Focusing on crop-water relationship, root growth and soil water utilization, deficit irrigation scheduling, cultivars characters related to drought resistance and managing practices to improve crop water use efficiency.

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### Assessment of the sustainability of different cropping systems under three irrigation strategies in the North China Plain under climate change

The annual double cropping system of winter wheat and summer maize requires a large amount of irrigation which has led to the rapid depletion of groundwater resources in the North China Plain (NCP). Alternate cropping systems and limited irrigation strategies should be developed for the purposes of maintaining sustainable groundwater use now and in the future. In this study, the water use and crop production of seven cropping systems under three irrigation strategies were assessed using the Agricultural Production Systems SIMulator (APSIM) during 1987-2017 as a baseline and in 2040, 2060, and 2080 under climate change conditions at a typical site in the NCP. The results showed that under current growing conditions, sustainable groundwater use could be achieved with the current double annual cropping system under minimum irrigation strategy, or reduced cropping intensity under full irrigation strategies. The traditional cropping system under minimum irrigation had the potential to sustain the groundwater balance in the region and concurrently resulted in higher grain production and WUE than that of the systems with reduced cropping intensity, both now and under climate change, and therefore, this system should be prioritized in this region.



- Yan, Z., Zhang, X., Rashid, M.A., Li, H., Jing, H., and Hochman, Z. (2020). Assessment of the sustainability of different cropping systems under three irrigation strategies in the North China Plain under climate change. Agricultural Systems, 178: 102745.
- Chen, S., Zhang, X., Shao, L., Sun, H., Niu, J., and Liu, X. (2020). Effects of straw and manure management on soil and crop performance in North China Plain. Catena, 187: 104359.
- Lu, Y., Yan, Z., Li, L., Gao, C., and Shao, L. (2020). Selecting traits to improve the yield and water use efficiency of winter wheat under limited water supply. Agricultural Water Management, 242: 106410.
- Liang, S., Zhang, X., Lu, Y., An, P., Yan, Z., and Chen, S. (2020). Performance of double cropping silage maize with plastic mulch in the North China Plain. Agronomy Journal, 112 (5): 4133.

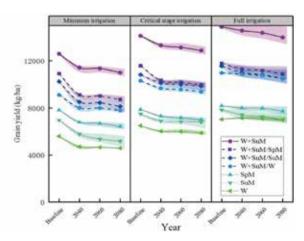


Figure: Simulated average annual grain yield for the seven cropping systems during the baseline period (1987-2017) and in 2040, 2060 and 2080.





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- Baillo, E.H., Hanif, M.S., Guo, Y.H., Zhang, Z.B., Xu, P., and Algam, S.A. (2020). Genomewide Identification of WRKY transcription factor family members in sorghum (*Sorghum bicolor*(L.) moench). PLoS One, 15 (8): e0236651.
- Fan, R., Chai, Z.Z., Xing, S.N., Chen, K.L., Qiu, F.T., Chai, T.Y., Qiu, J.L., Zhang, Z.B., Zhang, H.W., and Gao, C.X. (2020). Shortening the sgRNA-DNA interface enables SpCas9 and eSpCas9(1.1) to nick the target DNA strand. Science China-Life Sciences, 63 (11): 1619.
- Tolosa, L.N. and Zhang, Z.B. (2020). The Role of Major Transcription factors in solanaceous food crops under different stress conditions: Current and future perspectives. Plants-Basel, 9 (1).

### Genetics and Breeding of Wheat Drought Resistance and Water Saving

Zhengbin Zhang, Principal Investigator, Ph.D. (1998, Northwest A&F University, China).

Research Interests: The cloning and function analysis of drought resistance and water ues efficiency related genes in wheat. The breeding of wheat drought resistance and water saving

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# Shortening the sgRNA-DNA interface enables SpCas9 and eSpCas9(1.1) to nick the target DNA strand

The length of the sgRNA-DNA complementary sequence is a key factor influencing the cleavage activity of Streptococcuspyogenes Cas9 (SpCas9) and its variants. The detailed mechanism remains unknown. Here, based on in vitro cleavage assays and base editing analysis, we demonstrate that reducing the length of this complementary region can confer nickase activity on SpCas9 and eSpCas9(1.1). We also show that these nicks are made on the target DNA strand. These properties encouraged us to develop a dual-functional system that simultaneously carries out double-strand DNA cleavage and C-to-T base conversions atseparate targets. This system provides a novel tool for achieving trait stacking in plants.

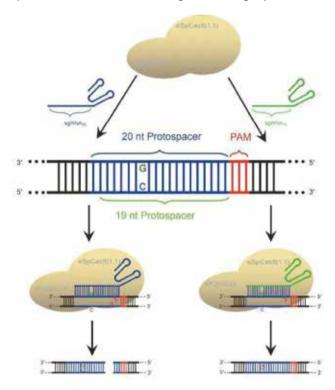




Figure: Mode of the APOBEC3A-eSpCas9(1.1)-UGI dual-functional gene editing system

### Center for Agricultural Resources Research

### **Terrestrial Ecology and Plant Sciences**

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Research Interests: Plant-mediated above- and below-ground species interactions: plant-soil feedback effects on aboveground plant-insect interactions; plant volatilemediated species interactions up to the fourth trophic level; mechanisms of plant responses to biotic and abiotic stresses.

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# Beyond plant microbiome composition: exploiting microbial functions and plant traits via integrated approaches

Plants recruit specific microorganisms to live inside and outside their roots that provide essential functions for plant growth and health. The study of the microbial communities living in close association with plants helps in understanding the mechanisms involved in these beneficial interactions. Currently, most of the research in this field has been focusing on the description of the taxonomic composition of the microbiome. Therefore, a focus on the plant-associated microbiome functions is pivotal for the development of novel agricultural practices which, in turn, will increase plant fitness. Recent advances in microbiome research using model plant species started to shed light on the functions of specific microorganisms and the underlying mechanisms of plant-microbial interaction. Integrated approaches, such as different multi-omics and microbiome engineering strategies, have greatly contributed to a better understanding of the and dynamics of plant-associated microbial communities. In addition, plant-soil feedbacks opened a new avenue for improvement of agricultural practices using knowledge obtained from natural ecosystems. Plant breeding programs have traditionally focused on exploring genetic variability of the crops for higher productivity and stress resistance, often neglecting the importance of beneficial interactions between microorganisms and plants. Therefore, future strategies for plant breeding should take plant microbial symbionts as lifelong bodyguards into consideration. These proposed integrated approaches will provide solutions for exploring and exploiting plant-microbiome interactions for improving the sustainability and productivity of global agriculture.

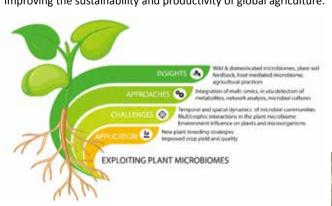


Figure: Summary of integrated approaches to explore and exploit plant microbiomes for new agricultural strategies.





### **Publications**

- Heinen, R., Hannula, S.E., De Long, J.R., Huberty, M., Jongen, R., Kielak, A., Steinauer, K., Zhu, F., and Bezemer, T.M. (2020). Plant community composition steers grassland vegetation via soil legacy effects. Ecology Letters, 23 (6): 973.
- Song, C., Zhu, F., Carrion, V.J., and Cordovez, V. (2020). Beyond plant microbiome composition: exploiting microbial functions and plant traits via integrated approaches. Frontiers in Bioengineering and Biotechnology, 8: 896.

Center for Core Facility & Advanced Technologies (CCFAT) were organized follow the principle that the center should prove technical service for the major project and tasks of the Country and the National Academy of Sciences, for the development of subjects and new layout for the development of the basic research capacity of the institute. CCFAT is currently composed of six specialized technical platforms (lipidomic analysis, proteomics analysis, metabolomics analysis, plant hormone analysis, biological big data analysis), and animal experiment center, which own the facilities and technologies that can strongly support genomics, proteomics, metabonomics, cell biology and mostly the whole life sciences research.

Lipidomic Analysis Platform continuously developed the quantitative and semi-quantitative analysis and detection methods of various lipids. The service scope of the lipidomics platform covers many domestic research units: In addition to more than 20 research groups in the Institute of Genetics Developmental Biology (IGDB), there are also Peking University, Tsinghua University, Fudan University, Nanjing University, West China Hospital, Institute of Zoology, Chinese Academy of Sciences, and Chinese Academy of Sciences Institute, Shanghai Institute of Plant Physiology, Beijing Institute of Life Sciences, Beijing University of Traditional Chinese Medicine, Beijing Academy of Agriculture and Forestry Sciences and many other well-known universities / research institutes in China. The subjects of lipidomics tests include human, mouse, monkey, fruit fly, nematode, cave fish, rice, Arabidopsis, green pepper and many other species. In 2020, the total time of the LC quadrupole linear ion trap tandem mass spectrometer was 1462.10 hours, the use efficiency was 72.81%, the total shared machine time was 909.64 hours, and the sharing efficiency was 45.30%. The annual total time of the ion chromatography system is 1150.55 hours, the use efficiency is 57.30%, the annual shared time is 329.74 hours, and the sharing efficiency is 16.42%. The mass spectrometry pre-treatment system (bead mill / vacuum centrifuge / lyophilizer / ultracentrifuge) has a total time of 1257.48 hours, a use efficiency of 62.62%, a total shared machine time of 28.57 hours, and a sharing efficiency of 1.42%. The total time of the macro zoom microscope is 49.26 hours, the use efficiency is 2.45%, the total shared time is 2.5 hours, and the sharing efficiency is 0.12%. The total time of the seahorse is 62.29 hours, and the use efficiency is 3.10%. The TripleTOF 5600+ has an operating time of 169.17 hours and an operating efficiency of 8.42%. A total of 26 articles have been published in 2020.



**Proteome Analysis Platform** had been improving the LC gradient and MS dissociation energy and being able to identify more than 7000 phosphopeptides by a single-shot LC-MS analysis, a coverage that has never been achieved before by any lab under the similar experiment conditions. Proteome Analysis Platform also evaluated the performance of the Advanced Peak Determination (APD) algorithm in proteome identification and TMT-labeling based proteomic quantitation, and found the potential risk in distorting TMT ratios in some specific cases although overall effect is not significant (Proteomics, 2020, Cover story). The platform have provided proteomic analysis services for many research groups in IGDB, as well as for many other institutions and universities, such as China Agricultural University, TianTan Hospital, Academy of Agricultural Sciences, etc. In 2020, the total operation time of the Orbitrap Fusion Lumos is 3850 hours, the service efficiency is 270%, the total sharing time is 3138 hours, and the sharing efficiency is up to 181.51%. The total operation time of the LTQ Orbitrap Elite is 3288 hours, the service efficiency is 230%, the total sharing time is 2843 hours, and the sharing efficiency is 92.43%, the annual sharing time is 1030 hours, and the sharing efficiency is 78.12%. The platform severed more than 1000 samples, and got about 1 million Yuan income in this year. The platform and its collaborators published 11 articles in Nature Plant, Molecular Plant and etc.



**Metabolomics Analysis Platform** focuses on profiling large numbers of metabolites to gain a high-resolution view of the biochemical phenotypes of living organisms. These quantitative and qualitative chemical data are highly valuable in research focused on gene validation, biomarker discovery, and the elucidation of uncharacterized metabolic pathways, as well as in studies searching for mechanistic insights about the regulation of metabolism. The platform has provided analytical services for more than one hundred customers, from both inside and outside IGDB, and published more than 30 papers with co-authorship in high-level journals including Nature Plants, Genome Biology, and PNAS. In 2020, the platform developed new methods for the study of metabolomics based on high throughput and high sensitive chromatographic mass spectrometry: (1) Based on LC/MS-QQQ technology, a highly sensitive method for the detection of lactucocirin and lactucrin sulfate in lettuce was developed; (2) A highly sensitive, high-throughput and specific quantitative method for flavonoid metabolites based on LC/MS-QQQ technology; (3) The cyanobacteria metabonomics analysis method based on GC/MS technology overcame the problem of poor repeatability of cyanobacteria sample derivation. The total service time of the four instruments on the platform was 2052 hours in 2020, a total of 5474 analytical samples were tested. The total service time of external service machine is 235 hours, and the total number of external test samples is 440.Two research articles and 3 collaborative papers were published this year.



**Plant Hormone Analysis Platform** has several sets of chromatograph and mass spectrometer and all kinds of equipment equipped in a standard analytical chemistry laboratory. The platform has long been committed to the development of high-efficiency and high-sensitivity phytohormone analysis methods and technologies. The quantitative analysis methods established by the platform have been widely used in the related research on the molecular mechanism of plant hormones, which has greatly promoted the publication of a number of high-level original achievements in the field of plant hormones in China. Based on the chemical structure and physiochemical properties of all known types of plant hormones, a sample pretreatment technology was designed to cover all the classical plant hormones and new hormones, including strigolactones and peptide hormones. Combined with LC-MS/MS technique, only 100 mg plant sample was needed to achieve accurate simultaneous quantitative analysis of all known classes of phytohormones, including indole acetic acid (IAA), cytokinins (CKs), ethylene, abscisic acid (ABA), jasmonic acid (JA), salicylic acid (SA), gibberellins (GAs), brassinosteroids (BRs), strigolactone (SL) and some plant peptide hormones (PSK) 44 compounds. This method covering all known types of phytohormones is the most comprehensive analytical method reported so far, and successfully solves the problem of simultaneous absolute quantitative analysis of plant hormones. The application of this method will provide more powerful technical support for the study of plant hormone network interaction. The platform shared the techniques and methods, developed deep cooperation with other research groups and provided technical services

and support to more than 50 research institutes. This year, platform received more than 3000 samples for phytohormone and related metabolite analysis. The platform instrument sharing time was 9883 hours.18 co-author articles have been published at Nature. Angewandte Chemie-International Edition. PNAS and other high-level magazines in 2020. The platform also held a professional technical lecture for the interested teachers and students in the institute.



**Bio-imaging Platform** has a variety of CT imaging systems and high-end fluorescence microscopes, which can meet the imaging needs of different applications. The platform insists on R&D of instruments to meet frontier scientific research needs: (1) To exceed the limitations of poor stability and complex operation of commercial microscopes, the platform proposed a two-photon fluorescence correlation spectroscopy technique; (2) Towards the urgent need of nondestructive method to acquire the internal structure and characteristics of seeds for micro cutting technology in molecular precision breeding, the platform designed a high-throughput seed phenotype identification scheme by combining optical with CT imaging technologies. This method can nondestructively obtain seed internal structure and external traits, and can precisely position specific organ; (3) The platform continues to develop new imaging methods. The best imaging conditions under different soil and soil moisture conditions were studied by using plant CT system, and the root CT images were obtained by using self-developed image processing method, which laid a foundation for studying the internal phenotype shape of crops in different periods. The time-lapse imaging system and self-developed long-life gold nanoparticles were used to image tobacco leaves to effectively remove spontaneous fluorescence. In 2020, the total service hours of the four main instruments in the platform were 2,483 hours, with a sharing rate of 74%. The platform held five technical training sessions, including one network conference, covering a total number of 210 trainees. This year, the platform cooperated with other research groups to publish 3 high-level scientific research papers and was granted 1 patent.



**Biological Big Data Platform** developed to achieve high quality assembly and analysis of plant genomes by which combines single molecule real-time sequencing, fosmid library sequencing, BioNano optical mapping, genetic mapping and other technologies and new software. Biological big data platform's high-performance computer cluster run 3,874,809 hours and accomplished 2,304,692 tasks for 40 groups of IGDB and many other institutes of scientific research in2020. A series of lectures and training

center on the data analysis requirement of each group were held during the past year. Specifically, a number of speakers gave lectures on the principle and analysis process of BSA, and then different speakers stated the key points of BSA analysis and the problems that should be paid attention to from different angles. Researchers had a profound understanding of the development process and current situation about the third-generation sequencing technology through PacBio sequencing principle and application lecture. Furthermore, it was helpful for researchers to choose the appropriate data acquisition scheme. The training of Single-Cell Sequencing data analysis and application presented the steps of Single-Cell data analysis and how to interpret the analysis results in detail. 3D genomics technology lecture introduced the principles of HiC, ATAC-seq and cut & tag technology and their application in plant field. These lectures and training not only enhanced the researchers' theoretical knowledge of cutting-edge technology, but also improved their practical ability.



The Experimental Animal Center ensured that the average rabbit, guinea pig breeding facilities, aquatic animal breeding facilities and the barrier facility running well for the entire year. The Center completed the renewal of the "License for use of laboratory animals" in July 2020. The new permit includes the barrier environment for mice and rats, and the conventional environment for rabbits, guinea pigs, and laboratory fish. The new animal facility has been completed and passed the Acceptance by four sides in November 2020. The new facilities are over 3865 square meters, of which the barrier facilities has1053 square meters. The barrier will all uses the IVC system, and also equipped with modern equipment such as video surveillance system, animal drinking system, tunnel-type cage washing machine, bedding dumping machine, xenon light transmission window. When the new facility was used, it can triple to amount of mice availability (increase 19,026 units), and further improves the service capacity. In 2020, the Experimental Animal Center have provided foster care services for 52 labs in 17 different institutions including Chinese Academy of Sciences, Peking University, and 301 Hospital; Provided antibody producing services for 30 labs in 10 institutions including the Chinese Academy of Sciences, Chinese Academy of Agricultural Sciences, and China Agricultural University. Provide



mice defecate services for 12 labs in 4 institutions including the Institute of Genetics and Development of the Chinese Academy of Sciences, Beijing Institute of Genomics, Peking University People's Hospital; Provide mice embryos transfer services 15 labs in 7 institutions including the Chinese Academy of Sciences, Beijing Ditan Hospital, and Beijing People's Hospital. The Experimental Animal Center has a qualified for isolation facility for imported mice and has provided 7 batches of imported animal isolation services for Guangzhou Medical University, Tsinghua University, and West Lake University.

The above-mentioned professional technology platforms take their own characteristics and advantages to provide strong guarantee for the major scientific research tasks of the Institute, and at the same time achieve the goals of scientific and technological innovation and social service. In 2020, CCAFT has made substantial progress in technological innovation and development, technical services and support, and technical lectures and training. 103 sets of equipment were shared use in IGDB. The total machine time was 12,1045 hours, the total utilization efficiency was 74.3%, the total sharing time was 43845 hours, and the total sharing efficiency was 26.1%. Six platforms provided technical support and services for the publication of 67 papers, and obtained one patent authorization. The CCAFT organized 13 technical lectures and training sessions, and trained nearly 1000 people.

### **State Key Laboratory of Plant Genomics**

The State Key Laboratory of Plant Genomics was approved in 2003 by the Ministry of Science and Technology of China, and roots back to the Key Laboratory of Plant Biotechnology of the Chinese Academy of Sciences (CAS), founded in 1990. The Laboratory has passed the Nationwide Evaluation of State Key Laboratories (5-year-peroid), and ranked outstanding (Rank A) in 2006 and 2011, and excellent (Rank B) in 2016.

The Laboratory combines multidisciplinary approaches to address fundamental questions in plant growth and development, with an emphasis on dissecting the molecular mechanisms of complex agronomic traits of crops. A mission of the Laboratory is to extend knowledge from basic research to crop improvement. Using rice and *Arabidopsis* as model systems, main research fields of the Laboratory include, but not limited to, the structure and function of plant genomes, functional genomics of important agronomic traits, molecular mechanisms of plant signal transduction, plant-environment and plant-pathogen interactions, and molecular breeding.

DIRECTOR: Jianru Zuo DEPUTY DIRECTORS: Chengcai Chu, Wei Qian

GROUP LEADERS: Yang Bai, Xiaofeng Cao, Mingsheng Chen, Shouyi Chen, Zhukuan Cheng, Chengcai Chu, Rongxiang Fang, Huishan Guo, Yantao Jia, Danhua Jiang, Yuling Jiao, Zhaosheng Kong, Chuanyou Li, Jiayang Li, Xiang Li, Chengzhi Liang, Shaoyang Lin, Jun Liu, Dongping Lv, Wei Qian, Wenfeng Qian, Jinlong Qiu, Bo Ren, Guodong Wang, Guixian Xia, Qi Xie, Cao Xu, Yongsheng Yan, Shanguo Yao, Jian Ye, Jie Zhang, Jinsong Zhang, Lili Zhang, Jian-min Zhou, Yihua Zhou, Lihuang Zhu, Zhen Zhu, Jianru Zuo

SCIENTIFIC STEERING COMMITTEE

CHAIR: Bin Han

VICE CHAIRS: Huishan Guo and Kang Chong

MEMBERS: Xiaofeng Cao, Xiaoya Chen, Jia Li, Jiayang Li, Qian Qian, Jianmin Wan, Weihua Wu, Jian-min Zhou, Yuxian Zhu, Jianru Zuo SCIENTIFIC ADVISORY BOARD: Shouyi Chen, Rongxiang Fang, Yaoguang Liu, Zhihong Xu, Qifa Zhang, Lihuang Zhu, Zhen Zhu HOMEPAGE: http://plantgenomics.genetics.cas.cn/

#### **MAJOR RESEARCH PROGRESSES**

In 2020, scientists in the State Key Laboratory of Plant Genomics published 136 papers in *Nature*, *Cell*, *Cell Host Microbe*, *Trends Plant Sci*, *FEMS Microbiol Rev*, *Nat Plants*, *Nat Commun*, *Mol Plant*, *Nucleic Acids Res*, *PNAS*, *EMBO J*, *Curr Biol*, *Plant Cell*, *Dev Cell* and etc. 11 varieties of rice and sorghum have obtained national or provincial approvals, and 23 patents have been granted.

In the past year, scientists in the Laboratory have made important advances in several research fields. In the area of plant genomics, Jingsong Zhang's group and collaborators revealed that asymmetric subgenome evolution and improvement of peanut by comparison of *Arachis monticola* with diploid and cultivated tetraploid genomes (Yin et al., *Adv Sci*, 2020). Lihuang Zhu's group and collaborators revealed important genetic variations and different heterosis patterns by resequencing 1143 *indica* rice accessions (Lv et al., *Nat Commun*, 2020). Chengzhi Liang's group and collaborators provided genomic resources for alfalfa research by the chromosome-level genome sequence of the autotetraploid alfalfa and resequencing of core germplasms (Shen et al., *Mol Plant*, 2020). Xiaofeng Cao's group and collaborators uncovered the protein arginine methyltransferase 3 fine-tunes the assembly/ disassembly of pre-ribosome to repress nucleolar stress by interacting with RPS2B in *Arabidopsis* (Hang et al., *Mol Plant*, 2020). The Cao group and collaborators demonstrated that RNA G-quadruplex structures exist and function in vivo in plants (Yang et al., *Genome Biol*, 2020). Zhukuan Cheng's group and collaborators revealed that 21-nt phasiRNAs direct target mRNA cleavage in rice male germ cells (Jiang et al., *Nat Commun*, 2020). The Cheng group and collaborators found that *Oryza sativa* RNA-dependent RNA polymerase 6 contributes to double-strand break formation in meiosis (Liu et al., *Plant Cell*, 2020).

In the area of functional genomics, Yihua Zhou's group and collaborators found that *MYB61* is regulated by GRF4 and promotes nitrogen utilization and biomass production in rice (Gao et al., *Nat Commun*, 2020). The Zhou group and collaborators also found that the phenylpropanoid derivatives are essential components of sporopollenin in vascular plants (Xue et al., *Mol Plant*, 2020). Shanguo Yao's group and Chengcai Chu's group identified the *SLG1* gene, and found that natural variations in *SLG1* confer high-temperature tolerance in *indica* rice (Xu et al., *Nat Commun*, 2020). Jiayang Li's group and collaborators found that the SL biosynthesis gene *HTD1* contributed to the improvement of plant architecture in modern rice varieties (Wang et al., *Mol Plant*, 2020). The Li group and collaborators found the regulation of rice tillering by RNA-directed DNA methylation at miniature

### Key Laboratories

inverted-repeat transposable elements (Xu et al., Mol Plant, 2020).

In the area of plant signal transduction, Jiayang Li's group revealed the transcriptional regulation of strigolactone signalling in Arabidopsis (Wang et al., Nature, 2020); further elucidated the molecular mechanism of the synergistic regulation of strigolactone and abscisic acid on tiller (Liu et al., Mol Plant, 2020); and uncovered the convergent pathway in SL- and KARregulated gene expression and hypocotyl elongation (Wang et al., Plant Cell, 2020). It has important guiding significance for the improvement of plant type and nutrient utilization efficiency, and the cultivation of parasite resistant crops. Qi Xie's group demonstrated that the UBC27-AIRP3 ubiquitination complex modulates ABA signaling by promoting the degradation of ABI1 in Arabidopsis (Pan et al., PNAS, 2020); and enunciated ESCRT-I component VPS23A is targeted by E3 ubiquitin ligase XBAT35 for proteasome-mediated degradation in modulating ABA signaling (Yu et al., Mol Plant, 2020); also demonstrated that VPS23A is a critical regulator of the SOS pathway that positively modulates the regulation of salt tolerance (Lou et al., Mol Plant, 2020). Yuling Jiao's group established a self-activation loop that maintains stem cells to enable branching (Cao et al., Curr Biol, 2020). The Jiao group and collaborators revealed that microtubule-mediated mechanical feedback amplifies an initial asymmetry and maintained directional growth (Zhao et al., Curr Biol, 2020). Chuanyou Li's group uncovered that SEUSS integrates transcriptional and epigenetic control of root stem cell organizer specification (Zhai et al., EMBO J, 2020); the Li group and collaborators demonstrated that mediator subunit MED25 couples alternative splicing of JAZ genes with fine-tuning of jasmonate signaling (Wu et al., Plant Cell, 2020). Jingsong Zhang's group and Shouyi Chen's group demonstrated the mechanistic details of ethylene signaling for root growth regulation (Zhao et al., Nat Commun, 2020); they also revealed a mechanism by which MHZ11 participated in ethylene signaling for regulation of root growth in rice (Zhao et al., Plant Cell, 2020). Jianru Zuo's group identified a transnitrosylase ROG1, positively regulating the Nitric Oxide-based redox signaling by promoting the degradation of its target, GSNOR1 (Chen et al., Dev Cell, 2020).

In the area of plant-environment and plant-pathogen interactions, Jian-Min Zhou's group and collaborators revealed how a plant defense metabolite protects plants by specifically disarming pathogen virulence machinery rather than non-selectively kill microbes (Wang et al., *Cell Host Microbe*, 2020). The Zhou group and collaborators provided evidence that pathogen proteins triggered resistosome formation *in vivo* and that this is required for disease resistance (Hu et al., *Mol Plant*, 2020). Zhaosheng Kong's group indicated that the transfer cells mediate nitrate uptake to control root nodule symbiosis (Wang et al., *Nat Plants*, 2020). Huishan Guo's group and collaborators demonstrated that a *Brassica* miRNA regulates plant growth and immunity through distinct modes of action (Cui et al., *Mol Plant*, 2020). The Guo's group and collaborators demonstrated DNA geminivirus infection induces an imprinted E3 ligase gene to epigenetically activate viral gene transcription (Chen et al., *Plant Cell*, 2020). Jun Liu's group uncovered the mechanism by which LORE mediates signaling in response to 3-OH-C10:0 in *Arabidopsis* (Han et al., *EMBO J*, 2020); and demonstrated that host RLKs could modify pathogen effectors to dampen their virulence and undermine their ability in suppressing PTI (Xu et al., *Mol Plant*, 2020). Yang Bai's group and collaborators demonstrated a crucial role of the bacterial microbiota in shaping rhizobia-host interactions in soybean (Han et al., *ISME J*, 2020).

In the area of plant biotechnology and molecular breeding, Jiayang Li's group bred and released new rice elite cultivars "Zhongkefa 7", "Zhongkefa 8", "Zhongkefa 176", "Zhonglonggeng 107" and "Quan 9 You 220". Shanguo Yao's group bred and released new rice elite cultivar "Zhongke 613". Zhukuan Cheng's group bred and released new rice elite cultivars "Zhongkeyan 3", "Zhongkeyan 5" and "Songkegeng 108". Chuanyou Li's group bred and released new tomato cultivars "Zhongqinghong 1" and "Zhongqingyingfen 1". All these varieties have obtained national or provincial approvals in 2020.

To recognize the contributions in their research fields, Profs. Jiayang Li, Jian-min Zhou, Chengcai Chu, Jie Zhang, Qi Xie, Yuling Jiao, Chuanyou Li, Zhen Zhu and Guodong Wang have been invited to write 23 review papers, spotlight papers or perspective papers for *Cell*, *Nat Plants, Trends Plant Sci, Curr Opin Plant Biol, Mol Plant, FEMS Microbiol Rev, New Phytol, J Exp Bot, J Integr Plant Biol*, etc.

### AWARDS AND RECOGNITIONS

In the past year, Xiaofeng Cao was elected a Foreign Member of the US National Academy of Sciences; and awarded the second prize of the National Natural Science Award in 2019 and the Pollyanna Chu Outstanding Teacher Award of the Chinese Academy of Sciences in 2020. The plant immunity research team, led by Jian-min Zhou, was awarded the 2020 Outstanding Science and Technology Achievement Prize of the Chinese Academy of Sciences. Yuling Jiao was honored with the 16th China Youth Science and Technology Award. Taolan Zhao, Jingying Zhang and Fangfang Wang were elected to the CAS Youth Innovation Promotion Association. Chengzhi Liang received the 2020 Outstanding Mentors Award of Chinese Academy of Sciences. Naiqin Zhong was awarded the Second prize of University Education and Teaching Achievement Award of Chinese Academy of Sciences. Naiqin Zhong was awarded the "Grassland talents of Inner Mongolia Autonomous Region". Jiayang Li, Chengcai Chu, Chuanyou Li and Jian-Min Zhou were recognized as Highly-Cited Researchers in 2020 by Clarivate Analytics.



### State Key Laboratory of Plant Cell and Chromosome Engineering

The State Key Laboratory of Plant Cell and Chromosome Engineering (PCCE) was established in 1989. Over the past 30 years, PCCE has become one of the most influential state key laboratories in China for its tremendous impact on advancing scientific research in crop genetics and breeding. PCCE is committed to three main research areas: (1) Dissecting the genetic basis of important agronomic traits of major crops systematically and identifying gene function underlying the control of plant growth, development, and environmental adaption. (2) Developing efficient gene pyramiding technologies, such as chromosome engineering, genome editing, and molecular design breeding, to facilitate an effective assembly of beneficial alleles for the breeding of elite varieties. (3) By combining the understanding in the genetics of agronomic traits and gene pyramiding technologies, PCCE designs and creates novel germplasms with favorable agronomic traits including high yield, superior quality, high nutrient efficiency, and improved tolerance to biotic and abiotic stresses. PCCE is leading breakthrough studies to address the most important challenges in plant science to expedite crop breeding and improve wellbeing.

DIRECTOR: Xiangdong Fu VICE DIRECTORS: Fangpu Han, Caixia Gao, Zhixi Tian PRINCIPAL INVESTIGATORS: Huabang Chen, Xiangdong Fu, Caixia Gao, Fangpu Han, Zanmin Hu, Junming Li, Yunhai Li, Zhensheng Li, Hongqing Ling, Cuimin Liu, Xignag Liu, Zhiyong Liu, Fei Lu, Qianhua Shen, Zhixi Tian, Yiping Tong, Daowen Wang, Lei Wang, Jun Xiao, Yongbiao Xue, Aimin Zhang ACADEMIC COMMITTEE CHAIR: Weihua Wu VICE CHAIR: Kang Chong MEMBERS: Xiangdong Fu, Bin Han, Bao Liu, Qian Qian, Qixin Sun, Daowen Wang, Yongbiao Xue, Fusuo Zhang, Xueyong Zhang, Jianmin Zhou, Yuxian Zhu ADVISORY COMMITTEE: Rongxiang Fang, Tingyun Kuang, Zhensheng Li

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#### MAJOR RESEARCH PROGRESS

In 2020, PCCE scientists have published 64 scientific papers in peer-reviewed academic journals, including *Cell, Science, Nature Biotechnology, Nature Genetics, Genome Biology* etc. PCCE scientists obtained 32 domestic and 8 international authorized invention patents. PCCE received approval for 5 new crop varieties, including three rice cultivars, a soybean cultivar, and a wheat cultivar. PCCE scientists have made important progresses in decoding complex agronomic traits, inventing genome-editing technologies and improving nitrogen use efficiency, etc. The representative achievements are as follows.

### Genetic Basis and Regulation Network of Important Agronomic Traits

Fei Lu's Lab performed whole-genome sequencing of 414 accessions from 25 subspecies of wheat relatives in the genus *Triticum* and *Aegilops* and constructed a genus-level genetic variation map of wheat (VMap 1.0). This is the first time to construct a genus-level whole-genome genetic variation map in all crops and revealed the genetic mechanism of the successful adaptation of wheat to both natural environments and human needs. This study provided new perspectives on crop improvement (*Zhou* et al., *Nature Genetics*, 2020). Yongbiao Xue's lab has finished the epigenome and transcriptome from a large spectrum of wheat samples under diverse developmental and environmental conditions, and found distinct epigenetic architectures of epiREs representing different levels of subgenome divergence. These results define an epigenomic atlas of subgenome-divergent regulation in hexaploid wheat and provide new clues for dissecting the genetic and epigenetic interplay in determining polyploid wheat heterosis (Wang et al., *The Plant Cell* (in press)). Zhixi Tian's Lab completed *de novo* genome assemblies and comparative genomic analysis for 26 most representative soybeans, and firstly realized the construction of a graph-based pan-genome in plants (Liu et al., *Cell*, 2020). They also found the feedback regulatory network of *Pd1-Ps-P1* controlling the pubescence development (Liu et al., *Molecular Plant*, 2020) and the molecular mechanisms of *GmSWEET10a* and *GmSWEET10b* coordinating yield and quality in soybean (Wang et al., *National Science Review*, 2020), and identified the PHYA(E3E4)-Tof11/Tof12-LHY-E1-FT molecular regulatory network for photoperiodic control of flowering in soybean (Lu et al., *Nature Genetics*, 2020), and the main regulatory sites of plant height (Fang et al., *Frontiers in Plant Science*, 2020), oil content (Tian et al., *Frontiers in Genetics*, 2020),

100-seed weight (Qi et al., *Frontiers in Genetics*, 2020), and pod number (Song et al., *Molecular Breeding*, 2020). Aimin Zhang's Lab identified a novel bZIP family transcription factor TubZIP28 from *Triticum urartu*, and its homologue TabZIP28 from *Triticum aestivum*, which were specifically expressed in endosperm of wheat at grain filling stage. Manipulation them could increase starch content and yield, with no trade-off on other agronomic traits (Song et al., *New Physiologist*, 2020).

Xiangdong Fu's lab has made an important breakthrough in the field of nitrogen use efficiency regulation in rice, demonstrating a new mechanism of epigenetic regulation, and modulation of GID1-NGR5-DELLA module provide a novel breeding strategy for enhancing future agricultural sustainability and food security (Wu et al., Science, 2020). Hongging Ling's lab cloned and demonstrated that GLU1, a gene encoding a ferredoxin-dependent glutamate synthase, was involved in iron homeostasis through affecting glutamate synthesis, chelating iron in vivo and promoting iron transportation (Cui et al., Journal of Integrative Plant Biology, 2020). They finely mapped the awn suppressor B1 with HIFs and found the expression differences caused by the variation in its promoter region resulted in the difference of awn length in HIFs. At the same time, they found awn length had no significant effect on agronomic traits in HIFs (Niu et al., Crop Journal, 2020). Yiping Tong's lab found TaLAMP1, a gene encoding carboxypeptidase, which is critical for wheat adaptation to N availability, and shaping plant architecture by regulating plant height, spike number per plant and grain number per spike. They also found that the down-regulation of brassinosteroid receptor TaBRI1 reduced photosynthesis, tolerance to high light and high temperature stresses and grain yield in wheat (Fang et al., Plants, 2020). Yunhai Li's lab discovered the KIX-PPD-MYC-GIF1 and GSK2-LARGE1/OML4 pathways determine seed size (Liu et al., Nature Communications, 2020; Lyu et al., The Plant Cell, 2020), and revealed that CUC2/CUC3-DA1-UBP15 regulatory module builds up a novel molecular link between organ size and lateral branch formation (Li et al., The Plant Cell, 2020). Cuimin Liu's Lab solved the structures of caseinolytic protease (Clp) and Clp-Cpn11/20/23 using single particle cryo-electron microscopy, and improved plant photosynthesis by manipulating starch metabolic enzyme.

Daowen Wang's lab discovered a viral protein that could interfere with the normal growth of cereal plants. This viral factor and its homologs, representing a unique type of mitosis regulator, exist in many plant and animal viruses and play an important role in viral pathogenesis. Zhiyong Liu's lab cloned the powdery mildew resistance genes *Pm5e* and *Pm24* from Chinese landraces and *Pm41* from wild emmer wheat. *Pm24* encodes a tandem kinase protein WTK3 and a rare 6-bp natural deletion confers the broad spectrum powdery mildew resistance (Lu et al., *Nature Communications*, 2020). Both *Pm5e* and *Pm41* encode NLR receptor proteins that are valuable for modern wheat breeding via molecular marker assisted selection and genome editing (Xie et al., *New Phytologist*, 2020).



### Key Laboratories

### **Genome Editing and Precision Plant Breeding**

Caixia Gao's Lab developed saturated targeted endogenous mutagenesis editors for directed evolution of plant genes (Li et al., *Nature Biotechnology*, 2020); established plant prime editor systems enabling point mutations, small insertions and deletions (Lin et al., *Nature Biotechnology*, 2020); developed a suite of APOBEC-Cas9 complexes for predictable and targeted deletions in plant genomes (Wang et al., *Nature Biotechnology*, 2020); developed new APOBEC3B-based cytosine base editors to eliminate sgRNA-independent DNA off-target edits (Jin et al., *Molecular Cell*, 2020); developed a multiplexed orthogonal genome editing in plants with a Cas9 nickase and engineered CRISPR RNA scaffolds to simultaneously generate C-to-T, A-to-G and knock out mutations (Li et al., *Genome Biology*, 2020); and achieved fine-tuning quantitative traits in asexually reproducing strawberry and obtained novel mutants containing a continuum of sugar content using base editing (Xing et al., *Genome Biology*, 2020).

### Plant Chromosome Engineering and Crop Breeding

Fangpu Han's Lab identified multiple circular RNAs generated via trans-splicing from specific centromeric retrotransposons in maize, which binds to the centromere region through R-loop structure and affects the centromeric chromatin organization (Liu et al., *PLoS Biology*, 2020). As well, they found that, in addition to sister chromatid cohesion, ZmSMC3 participates in early meiotic centromere pairing (Zhang et al., *Plant Cell*, 2020). Zhensheng Li's Lab found that *TaVTE3* regulates photosynthetic efficiency and its favorable SNP alleles may be used for high photosynthetic efficiency in wheat breeding. Meanwhile, an uncharacterized *TaRKL1* encoding a receptor-like cytoplasmic kinase was found to regulate photosynthetic efficiency and H<sub>2</sub>O<sub>2</sub> homeostasis (Ying et al., *Photosynthetica*, 2020). In addition, a total of 90 stable QTL for 15 agronomical traits were detected on salt stress by using a RIL population derived from a cross between Zhongmai 175 and Xiaoyan 60 (Luo et al., *TAG*, 2020). Huabang Chen's lab revealed the molecular mechanism of maize S-type cytoplasmic male sterility gene orf355 and transcription factor ZmDREB1.7 in synergistic regulation of pollen fertility.

In addition, three rice varieties "Zhongheyou 3", "Zhongheyou 5", "Zhongheyou 6" developed by Xiangdong Fu's lab, and a soybean variety "Kedou 10" developed by Baoge Zhu's lab, were approved by the national variety committee. A wheat variety "Kenuo 2" developed by Aimin Zhang's lab was approved by the local variety committee of Shandong Province. Junming Li's lab bred a new high-yield and water-saving wheat variety Kenong 8162 through a distant hybridization between wheat and *Aegilops*.

# State Key Laboratory of Molecular Developmental Biology

The State Key Laboratory of Molecular Developmental Biology is hosted in the Institute of Genetics and Developmental Biology. Our mission is: 1) to address fundamental questions in development of both plants and animals using model organisms such as *C. elegans, Drosophila, Xenopus,* zebrafish, mouse, monkey, *Arabidopsis* and rice; and 2) to develop innovative technology to meet national needs in agriculture and human health.

DIRECTOR: Weicai Yang VICE DIRECTORS: Xun Huang, Fan Chen, Zhiheng Xu
PRINCIPAL INVESTIGATORS: Shilai Bao, Yuhang Chen, Jianwu Dai, Zhuo Du, Mei Ding, Weixiang Guo, Kangmin He, Yuqiang Jiang, Jiajia Liu, Falong Lu, Runlin Ma, Wenxiang Meng, Guanghou Shui, John R. Speakman, Ye Tian, Qiang Tu, Yingchun Wang,
Zhaohui Wang, Qingfeng Wu, Yongqing Zhang
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HOME PAGE: http://mdb.genetics.ac.cn

In 2020, the laboratory published 121 papers. Four eminent scientists spoke at the FORUM on DEVELOPMENT, GENETICS, AND DISEASE. During 2020, the laboratory achieved significant advances in the following fields:

#### **Cell Differentiation and Organ Formation**

Successful cloning by somatic cell nuclear transfer (SCNT) requires overcoming significant epigenetic barriers. Dr. Falong Lu with collaborators reports significant enhancement of SCNT efficiency by deriving somatic donor cells carrying simultaneous monoallelic deletion of four H3K27me3-imprinted genes Quadruple monoallelic deletion of the four genes increased fibroblast cloning efficiency to 14% compared with a 0% birth rate from wild-type fibroblasts while preventing the placental and body overgrowth defects frequently observed in cloned animals. These results show that lack of H3K27me3 imprinting in somatic cells is an epigenetic barrier that impedes post-implantation development of SCNT embryos and can be overcome by monoallelic imprinting gene deletions in donor cells (Cell Stem Cell, 2020). Dr. Wenxiang Meng's group finds CAMSAP1 plays a decisive role in the neuronal axon identification process by regulating the number of microtubules (MTs). Neurons lacking CAMSAP1 form a multiple axon phenotype in vitro, while the multipolar-bipolar transition and radial migration are blocked in vivo. They demonstrate that the polarity regulator MARK2 kinase phosphorylates CAMSAP1 and affects its ability to bind to MTs, which in turn changes the protection of MT minus-ends and also triggers asymmetric distribution of microtubules. These indicate that the polarized MT network in neurons is a decisive factor in establishing axon/dendritic polarity and is initially triggered by polarized signals (PNAS, 2020). Dr. Weicai Yang's lab show that the seven transmembrane protein MILDEW RESISTANCE LOCUS O (MLO) are required for pollen tube responses to ovular signals in Arabidopsis. This study identifies the pollen tube-expressed, MLOs act as tethering factors for membrane targeting of Ca<sup>2+</sup> channel CNGC18 to integrate extracellular ovular cues and intracellular selective exocytosis, this sheds light on the general regulation of MLO proteins in cell responses to environmental stimuli (Nature Plants, 2020).

### **Neurodevelopment and Regeneration**

Calcium-dependent proteolytic Calpains are implicated in a variety of physiological processes, as well as pathologies associated with calcium overload. Dr. Yongqing Zhang's lab discover that Calpain activity was inhibited upon knockdown of *Ttm50*. Their findings reveal the regulation of Calpain activation by Ttm50, and shed new light on Calpain-associated pathologies (*Cell Research*, 2020). Zika virus (ZIKV) has evolved into a global health threat because of its causal link to congenital Zika syndrome. Dr. Zhiheng Xu's lab finds although mouse brains infected later after birth do not have apparent abnormal brain structure, those mice still show significant impairments of visual cortical functions, circuit organization, and experience-dependent plasticity. The finding suggests that special attention should be paid to children born to ZIKV infected mothers for screening of abnormal behaviors and sensory function during childhood (*Advanced Science*, 2020). Neurogenesis in the developing neocortex relies on

### Key Laboratories

extensive mitosis of radial glial cells (RGCs) in the apical surface. Dr. Qingfeng Wu's lab finds Talpid3, related to Joubert syndrome, is localized to the mother centriole of RGCs and is required for their apical mitosis. Further analysis revealed that genetic ablation of Talpid3 results in synchronized, ectopic mitosis of neural progenitors and dysregulated neurogenesis. These findings provide an intriguing perspective for the non-ciliogenic role of centriolar proteins in mediating cortical neurogenesis (*Cell Reports*, 2020). Dr. Jianwu Dai's group have developed a biocompatible collagen scaffold that can be injected directly into the heart, which can provide stable extracellular matrix support for cells and effectively limit cell proliferation from the site of injury. The randomized, single-center clinical trial's result proved collagen gel may be a feasible and safe method to promote cell therapy and set the grounds for adequately powered efficacy studies (*JAMA Network Open*, 2020).

#### **Tissue Metabolism and Aging**

The coronavirus disease 2019 (COVID-19) pandemic presents an unprecedented threat to global public health. Dr. Guanghou Shui's lab with collaborator utilize a combination of targeted and untargeted tandem mass spectrometry to analyse the plasma lipidome and metabolome in mild, moderate and severe COVID-19 patients and healthy controls. Their findings suggest that gangliosides (GM3) -enriched exosomes may partake in pathological processes related to COVID-19 pathogenesis, and presents the largest repository on the plasma lipidome and metabolome distinct to COVID-19 (*Cell Metabolism*, 2020). Mild mitochondrial stress experienced early in life can have beneficial effects on the life span of organisms through epigenetic regulations. Dr. Ye Tian's lab find that acetyl-coenzyme A (CoA) represents a critical mitochondrial signal to regulate aging through the chromatin remodeling and histone deacetylase complex (NuRD) in *Caenorhabditis elegans*. The finding reveals the molecular mechanism of the metabolite-mediated epigenome for the regulation of organismal aging (*Science Advances*, 2020). The appearance of lipid droplets in neurons is not usually observed under physiological conditions, but is associated with neural diseases. Dr. Xun Huang's lab discover that mutations of two key lipolysis genes *atgl-1* and *lid-1* lead to LD appearance in neurons of *C. elegans*. This neuronal lipid accumulation protects neurons from hyperactivation-triggered neurodegeneration, with a mild decrease in touch sensation. These results suggest the crucial role of neuronal lipolysis in cell-autonomous regulation of neural functions and neurodegeneration (*EMBO Reports*, 2020).



### Luancheng National Station of Agricultural Ecosystem,China National Ecosystem Observation and Research Network

Luancheng station, established in 1981, is one of the field stations of the Chinese Ecosystem Research Network (CERN) and a member of Global Terrestrial Observation System (GTOS). The station is also a demonstration base for modern agricultural technologies in Hebei province. In 2005, the station became one of the stations of the Chinese National Ecosystem Observation and Research Network (CNERN). In 2016, the station joined the International Long TERM Ecological Research Network (ILTER).

The primary goals of the station are to implement long-term comprehensive observations on the structure, function, and evolvement of the agro-ecosystem; to clarify the mechanisms of the energy, water, and nutrient transfer processes of the farmland ecosystem and the theoretical basis for interface regulation; and to study the structural functions of the integrated system of agro-ecology and economy.

DIRECTOR: Yanjun Shen VICE DIRECTOR: Yan-jun Shen, Hongjun Li PRINCIPAL INVESTIGATOR: Chunsheng Hu, Xiying Zhang, Yanjun Shen, Mengyu Liu, Junming Li, Diaoguo An, Zhenngbin Zhang, Lin Ma, Shiqin Wang, Binbin Liu, Xiaofang Li, Hongyong Sun, Feng Zhu, Xiuwei Liu, Yan-jun Shen, Baodi Dong, Zhaohai Bai COMMITTEE OF SCIENCE

DIRECTOR:Academician Mingan Shao COMMISSARY: Academician Fusuo Zhang, Keling Wang, Ganlin Zhang, Chunming Liu, Chunsheng Hu, Xiying Zhang , Yi Luo, Yanjun Shen

### Main outputs in 2020

Totally, up to 72 papers were published in 2020, of which 53 were on the SCI indexed journals.

Twelve patents were granted in 2019, of which one were invention patents. And ten patents, including one inventive patent, have been applied for certification.

### Main progresses in 2020

The conversion of natural grassland to semi-natural or artificial ecosystems is a large-scale land-use change (LUC) commonly occurring to natural saline-alkaline land. Median  $N_2O$  flux was significantly different among the ecosystems with the highest from the cropland, intermediate from the Tamarix and the lowest from the grassland ecosystem. The <sup>15</sup>N isotopic signatures of  $N_2O$  emitted from the soil were also significantly affected by the LUC with more depleted from cropland and less depleted when emitted from grassland soil. Our results suggest that the conversion of native saline-alkaline grassland with low N to Tamarix or cropland is likely to result in increased soil  $N_2O$  emission to the atmosphere and also contribute significantly to the depletion of the <sup>15</sup>N isotopic composition of atmospheric  $N_2O$ .

Root is the major organ for plant to absorb water and mineral nutrients. According to KN9204 genome sequence assembly, there are 6 genes annotation in the QMrI-7B target interval. Among these candidate genes,TraesKN7B01HG37540, corresponding toTraesCS7B02G339100, was differentially expressed between the parents as well as the near isogenic lines. The effects of QMrI-7B were dissected under both low and high nitrogen environments. Trials in two consecutive growing seasons showed that QMrI-7B confers wheat upon larger root system which is favorable to nitrogen uptake and grain production, exhibiting higher harvest index, nitrogen harvest index and nitrogen partial factor productivity.

The length of the sgRNA-DNA complementary sequence is a key factor influencing the cleavage activity of Streptococcuspyogenes Cas9 (SpCas9) and its variants. Based on in vitro cleavage assays and base editing analysis, we demonstrate that reducing the length of this complementary region can confer nickase activity on SpCas9 and eSpCas9. We also show that these nicks are made on the target DNA strand. These properties encouraged us todevelop a dual-functional system that simultaneously carries out double-strand DNA cleavage and C-to-T base conversions atseparate targets. This system provides a novel tool for achieving trait stacking in plants.

# Key Laboratory of Agricultural Water Resources, Chinese Academy of Sciences Hebei Key Laboratiory for Water-Saving Agriculture

Heibei Key Laboratory for Water-saving Agriculture was founded in 2005 by the Science and Technology Department of Hebei Province. Based on this laboratory, Key Laboratory of Agricultural Water Resources of Chinese Academy of Science was established in 2008.

The laboratory focus on improving agricultural water use efficiency in individual, field and regional levels, guarantying food security in North China, achieving sustainable development of agriculture, carrying on theoretical and technological innovation of high efficient use on agricultural water resources, providing theoretical and technical support for regional shortage of agricultural water resources.

DIRECTOR: Yanjun Shen VICE DIRECTOR: Xiying Zhang, Shiqin Wang PRINCIPAL INVESTIGATORS: Diaoguo An, Zhaohai Bai, Jiansheng Cao, Baodi Dong, Chunsheng Hu, Xiaofang Li, Binbin Liu, Jintong Liu, Mengyu Liu, Xiaojing Liu, Xiuwei Liu, Lin Ma, Shenyan Jun, Shenyan Jun, Shiqin Wang, Yonghui Yang, Wanjun Zhang, Xiying Zhang, Zhengbin Zhang, Zhu Feng SCIENTIFIC ADVISORY COMMITTEE: DIRECTOR: Dahe Qin VICE DIRECTOR: Bojie Fu, Chunsheng Hu MEMBERS: Shaozhong Kang, Jun Xia, Fengchang Wu, Wenjun Ding, Xiaoyan Li, Xurong Mei, Jianhua Wang, Yanfen Wang, Yonghui Yang, Huijun Wang, Zhaoji Zhang, Zhenghui Xie WEBSITE: http://lawr.sjziam.ac.cn:8085/

### MAIN RESEARCH PROGRESSES:

Substantial progresses in several aspects have been achieved by the key laboratory in 2020: Hebei key laboratory for watersaving agriculture got the fifth "excellent grades" of the assessment continuously, which held by Science and Technology Development of Hebei Province. The hydro geochemistry test platform for vadose zone has passed the examination of the experts panel organized by the Chinese Academy of Science. One Second class Prize of Natural Science Award and one first class Prize of Scientific and Technological Progress Award in Hebei province were won by the key Lab. 58 SCI papers were published and eight excellent graduated doctors joined the key Lab. in this year.

### Crop physiological and genetic basis for high efficient water use

Professor An Diaoguo's group performed quantitative trait loci (QTL) analyses using recombinant inbred lines derived from the cross 'PB3228 × Gao 8901' (PG-RIL) to dissect the genetic basis of kernel traits. It was found that the favorable *TaFT-D1* (*G*)-allele has been positively selected during Chinese wheat breeding. Thus, these results can be used for further positional cloning and marker assisted selection in wheat breeding programs. Liu Xiuwei's group use field grown winter wheat under three water treatments to investigate the changes of flag (top) leaf traits including leaf mass per area, wax n-alkanes and carbon composition of bulk leaf and n-alkanes ( $\delta$ 13Calk) during leaf development. The study find that leaf wax n-alkane synthesis ceases when the leaf reaches its maximum area under dry conditions. However, fully expanded leaves can still increase their n-alkane concentrations by 42% and 70% under deficit and full irrigation, respectively. Liu Xiaojing's group studied the wamp rose mallow seedlings grown in split-root pots irrigated with different concentration of NaCl solutions. The results indicate that under nonuniform salinity, SRM seedling growth was significantly inhibited, however, the salt stress was alleviated by the partial salt-free root zone compared to uniform the salinity.

#### Hydrological process in farmland

Professor Liu Mengyu and his collaborators analyzed the relationship between irrigation amount and irrigation unit size was determined by combining different check lengths and widths, beside on the results, the concept of "micro-flow irrigation" is put forward. Micro-flow irrigation is a kind of multi-outlet irrigation technology, which could improve the WUE of winter wheat. Professor Sun Hongyong's group using the data of a two-year spring maize experiment built a model based on the

### Key Laboratories

diffusion theory. The results showed that 10 days after physiological maturity, the MCAM could be reduced to 28% in almost all circumstances and to 25% in half of the circumstances. Professor Sun Hongyong's group assessed the water use and crop production of seven cropping systems under three irrigation strategies using the Agricultural Production Systems SIMulator (APSIM) during 1987-2017 as a baseline and in 2040, 2060, and 2080 under climate change conditions at a typical site in the NCP. The results showed that under current growing conditions, sustainable groundwater use could be achieved with the current double annual cropping system under minimum irrigation strategy, or reduced cropping intensity under full irrigation strategies.

### Optimal allocation of regional agricultural water resources

Professor Shen Yanjun' s group studied the water balance characteristics of different agricultural ecosystems in North China Plain. Based on the results of water balance, by the measures such as total amount control, structural optimization and efficiency improvement, an "appropriate water" grain crop, cash crop and forage rotation allocation system with the amount of renewable agricultural water resources at the county level as the constraint has been constructed. Professor Yang Yonghui' s group evaluated three satellite precipitation products(SPPs) against a high-density network of 104 rain gauges over the Taihang Mountains. The result could not only give a detailed assessment of mainly current SPPs over the Taihang Mountains, but also recommend that further improvement on retrieval algorithm is needed by considering topographical impacts for SPPs in the future. Professor Shen Yanjun' s group used GIS spatial analysis technology, R/S method and Mann-Kendall trend analysis method revealed the Rn spatio- temporal variation characteristics. It is of great significance to the estimation of evapotranspiration, plant growth process and global climate change.

### Journal of Genetics and Genomics

Journal of Genetics and Genomics (JGG; known as Acta Genetica Sinica prior to 2007) is an international recognition journal in the field of genetics and genomics. The journal was founded in 1974 and co-sponsored by Institute of Genetics and Developmental Biology, Chinese Academy of Sciences and Genetics Society of China. The journal has been jointly published by Elsevier Ltd. and Science Press since 2006.

JGG aims to promote academic communications and foster scientific advances by publishing high-quality peer-reviewed articles of novel and significant scientific discoveries. The journal covers a broad range of research areas, including, but not limited to, molecular genetics, developmental genetics, epigenetics, medical genetics, population and evolutionary genetics, genomics and functional genomics, cell biology and signal transduction, omics and big data, computational biology, and other important fields in life sciences. Currently, JGG is indexed by Science Citation Index Expanded (SCIE), PubMed, Scopus, and other international databases. The current impact factor of JGG is 5.065, released by Journal Citation Reports (JCR) in 2019, ranking top 25% of all journals in the field of genetics (a.k.a Q1).

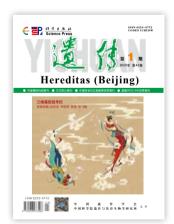


To further enhance competitiveness, JGG has adopted a series of organizational changes in 2020. The new Editor-in-Chief was appointed in July 2020 and subsequently, the new editorial board was established, absorbing more young scientists. Later, in November, the annual editorial meeting was held in Kunming, Yunnan to discuss the developing strategies for the future of JGG. In addition, JGG strengthened the editorial team by recruiting high-quality editors with solid scientific backgrounds and optimized its production and publication procedures to meet the growing needs of fast publication schedules and better publishing experiences. Meanwhile, JGG has strived to increase its influence through multiple channels, such as promoting articles and scientific progress in WeChat, attending academic conferences, and organizing special issues focusing on international cutting-edge research. Currently, two special issues of the journal, 'Cancer Genetics, Genomics and Epigenetics' and 'Microbiome' are launched and issued call for papers.

### Hereditas (Beijing)

Founded in 1979, *Hereditas (Beijing)* is sponsored by Institute of Genetics and Developmental Biology of Chinese Academy of Sciences and Genetics Society of China, and published monthly by Science Press. *Hereditas (Beijing)* is a national academic journal, Chinese core journal, and top-notch Chinese science and technology journal. Prof. Xun Huang is the Editor-in-Chief.

*Hereditas (Beijing)* has been indexed by more than 20 retrieval systems and databases, such as PubMed/MEDLINE, Biological databases, Index Medicus (IM), Russian Abstract Journal (AJ), Chemical Abstract (CA). *Hereditas (Beijing)* is also released on many databases and websites, such as CNKI, Wanfang Database, Chongqing VIP Database, and the website www.chinagene. cn. It publishes reviews and research articles in the fields of genetics, genomics, developmental biology, evolution, biotechnology, etc.



In 2020, the new editorial board of Hereditas (Beijing) was assembled and the editorial board

meeting was held at Kunming, China to discuss the development of the journal. In order to improve the academic quality of *Hereditas (Beijing)*, the special columns such as Invited Review, Frontier and Focus, and Resource and Platform were strengthened. Meanwhile, a special section on Three-Dimensional Genome was published. In order to enrich the content and increase the readability of this journal, new columns such as Editor Recommendations, Spotlights, and Research Frontiers were successively set up.

### **Chinese Journal of Eco-Agriculture**

*Chinese Journal of Eco-Agriculture* (CJEA, formerly known as Eco-Agriculture Research) was founded in 1993, and is sponsored by Institute of Genetics and Developmental Biology of Chinese Academy of Sciences (IGDB, Beijing), undertaken by Center for Agricultural Resources Research of IGDB (Shijiazhuang), published monthly by Sciences Press in Chinese or English. The Editor-in-Chief of CJEA is Academician LIU Changming, and Executive Editor-in-Chief is Prof. HU Chunsheng. CJEA is a per-reviewed academic journal. The goal of the journal is to provide a platform for scientists, academicians, and engineers to promote, share, and discuss various new issues and developments in different areas of eco-agriculture, such as agroecosystem and regulation, crop cultivation and physiological ecology, agricultural resources and environment, agricultural ecologic economics and ecoagriculture. CJEA has been included in various databases, such as CNKI, Wanfang Date, CSCD, VIP, Chaoxing, SCOPUS, CABI, CA, JST.

In 2020, the special column for "Green Development in Agriculture" and the "Special Issue for the 15<sup>th</sup> Anniversary of the Protection of GIAHS in China" were published in August and September.



### **Major Events**

### **3** January

2019 Annual Meeting of IGDB was held.



### 7 January

Center for Agricultural Resources Research held 2019 annual meeting.



### **10 January**

The State Key Laboratory of Plant Genomics held the annual academic meeting.



### **10 January**

The Discipline Inspection Commission of IGDB held 2019 annual meeting.



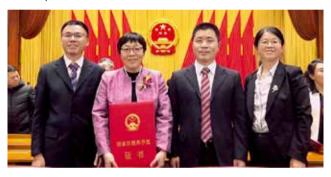
### **10 January**

The State Key Laboratory of Plant Cell and Chromosome Engineering held 2019 Academic Committee Meeting.



### **10 January**

"Mechanisms of histone methylation and small RNAs in regulating plant development and transposon activity" won the second prize of National Natural Science Award in 2019.



### 11 January

CAS Innovation Academy for Seed Design held 2019 annual meeting.



### 27 January

CAS Engineering Laboratory for Efficient Utilization of Saline Resources was launched.



### 27 April

Dr. Xiaofeng Cao and Dr. John Speakman were elected as Foreign Member of the National Academy of Sciences, USA.



### 27 April

CEPAMS signed the second five-year cooperation agreement.



### 30 April

IGDB held the first meeting of the Fifth Committee.



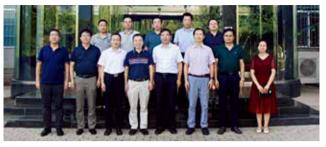
### 14 May

The sixth session of the fourth Labor Union Congress of IGDB was held.



### 24-26 August

Gansu Agricultural University visited Center for Agricultural Resources.



### **18 September**

The opening ceremony and entrance education for 2020 new students of IGDB were held.



### 20 October

IGDB held the Fifth Party Congress.



#### **3 November**

IGDB signed astrategic cooperation agreement with Beijing Children's Hospital.



#### 10-12 November

International Students Conference 2020 among NAIST, UC Davis and IGDB was held.



### Major Events

### **17 November**

The preparatory work of Innovation Academy for Seed Design passed the acceptance evaluation organized by CAS.



#### **18 November**

Five researchers from IGDB were recognized as Clarivate Analytics Hightly-Cited Researchers 2020.



### 25 November

Presentation Ceremony for YIHAI KERRY Awards in 2019 and 2020 was held.



### 28-29 November

The State Key Laboratory of Plant Genomics held 2020 Annual Forum of Young Scholars.



#### 30 November- 1 December

The Institute of Subtropical Agriculture CAS visited Center for Agricultural Resources.



### 2 December

IGDB and Beijing Genomics Institution established a multidimensionalomics joint center.

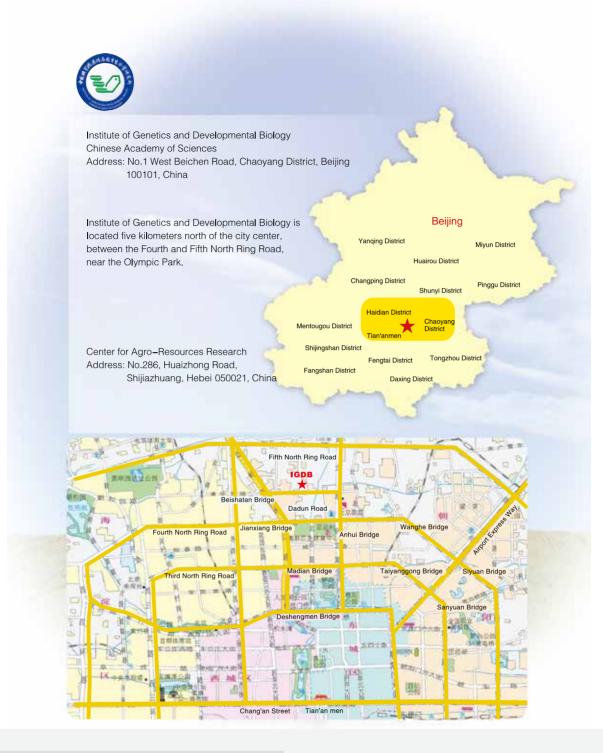


**10 December** Hou Jianguo, President of CAS, visited IGDB.



**15-17 December** IGDB held 2020 strategic planning and seminar.





### **Editorial Committee of Annual Report**

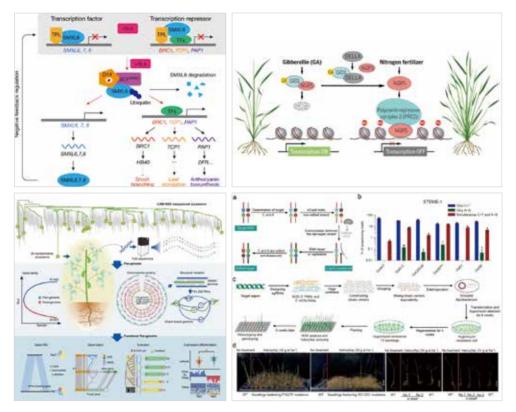
Chair: Xun Huang

Vice Chair: Xin Yu

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Hong Ji Guanghou Shui Jianru Zuo Hongqing Ling Limei Tan

Cover image: The second prize of photography competition of the Innovation Academy for Seed Design by Ke Huang



Figures are from the published papers by Jiayang Li's group (Wang et al., *Nature*, 2020), Xiangdong Fu's group (Wu et al., *Science*, 2020), Zhixi Tian's group and Chengzhi Liang's group (Liu et al., *Cell*, 2020), Caixia Gao's group and Jiayang Li's group (Wang et al., *Nature Biotechnology*, 2020).

### 2020 Annual Report

Institute of Genetics and Developmental Biology Chinese Academy of Sciences

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