



第四届全国玉米生物学学术研讨会

The 4th Maize Biology Conference of China

April 19-22, 2019

Program & Abstracts

主 办

中国作物学会玉米专业委员会

承 办

省部共建小麦玉米作物学国家重点实验室

河南省粮食作物协同创新中心

河南农业大学农学院

国家玉米改良（郑州）分中心

Endorsed by

The Crop Science Society of China

Organized by

National Key Laboratory of Wheat and Maize Crop Science

Collaborative Innovation Center of Henan Grain Crops

College of Agronomy, Henan Agricultural University

Zhengzhou Subcenter of National Maize Improvement Center

会务信息

欢迎您来到郑州参加第四届全国玉米生物学学术研讨会。本次会议于 2019 年 4 月 19 日-21 日在河南省黄河迎宾馆召开。为了保证会议的顺利进行，请认真阅读以下信息：

- **会议注册**

会议注册时间为 4 月 19 日 8:30-22:00，注册地点位于郑州黄河迎宾馆 9 号楼 1 楼大厅。

- **报告**

所有会议报告均在河南省黄河迎宾馆中华厅进行。会议日程和摘要详见内页。

- **会议墙报**

墙报环节在郑州黄河迎宾馆梅花厅进行，欢迎与会代表积极参与交流讨论。4 月 19 日 15:00 开始张贴墙报。4 月 20 日 16:15-18:15 进行正式墙报交流，请墙报作者注意奇偶序号和时间，于墙报前同参会代表讨论，18:15-22:00 为墙报自由交流时间。墙报请于 4 月 21 日 19:00 前收回。

- **住宿**

本次会议住宿地点为河南省黄河迎宾馆（郑州市迎宾路 1 号）。

- **餐饮**

黄河迎宾馆 9 号楼 1 楼天香苑餐厅、10 号楼 1 楼富丽宫餐厅为本次会议提供午餐、晚餐，请凭餐券按时用餐。早餐由入住酒店提供。在墙报交流期间，会务组提供茶点和饮料。

- **联系信息**

如有任何疑问或需要帮助，请联系黄河迎宾馆 9 号楼的大会工作人员。

General Information

Welcome to Zhengzhou for the 4th Maize Biology Conference of China. The conference will be held on 19-21 April, 2019 in Yingbin Hotel of the Yellow River Henan. To ensure the smooth progress of the conference, please read the following information carefully.

- **Meeting Registration**

From 8:30 AM to 22:00 PM on April 19. The registration site is located in the lobby of Building 9.

- **Lectures**

All lectures will be presented in Zhonghua Hall.

- **Poster Session**

Posters will be presented in Meihua Hall. Posters should be hung up starting at 15:00 PM on April 19, and removed after 19:00 PM on April 21. During the poster sessions, presenters should stand by the poster to answer the questions for the participants.

- **Accommodation**

The hotel for this meeting is Yingbin Hotel of the Yellow River Henan (No.1, Yingbin Road, Zhengzhou).

- **Meals**

Yingbin Hotel of the Yellow River Henan provides the lunch and dinner at Tianxiangyuan Restaurant in Building 9 (Lobby Level) and Fuligong Restaurant in Building 10 (Lobby Level). Please have the breakfast at the hotel where you live. Tea and snack will be provided at informal poster time.

- **Contact information**

The lobby of Building 9 is for conference staffs. Please feel free to contact us for any questions or if you need any help.

会议地址和地图

(Hotel Information and Maps)

会议地址：河南省黄河迎宾馆（郑州市迎宾路 1 号）

(Address: Yingbin Hotel of the Yellow River Henan (No.1, Yingbin Road, Zhengzhou.)

电 话(Tel): +86+371+66778888



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第四届全国玉米生物学学术研讨会

组委会名单

(Organizing Committee)

主办单位 (Sponsoring Organization): 中国作物学会玉米专业委员会

承办单位 (Sponsoring Units): 省部共建小麦玉米作物学国家重点实验室

河南省粮食作物协同创新中心

河南农业大学农学院

国家玉米改良（郑州）分中心

组委会主席 (Chair):

谭保才 (山东大学, SDU; bctan@sdu.edu.cn)

组委会秘书长 (Secretary-general):

汤继华 (河南农业大学, HENAU; tangjihua1@163.com)

组委会委员 (Members):

代明球 (华中农业大学, HZAU; mingqiudai@mail.hzau.edu.cn)

赖锦盛 (中国农业大学, CAU; jlai@cau.edu.cn)

李 林 (华中农业大学, HZAU; hzaulilin@mail.hzau.edu.cn)

李培金 (安徽农业大学, AHAU; Peijin.li@ahau.edu.cn)

李文学 (中国农科院作物科学研究所, CAAS; liwenxue@caas.cn)

卢艳丽 (四川农业大学, SICAU; yanli.lu82@hotmail.com)

秦 峰 (中国农业大学, CAU; qinfeng@ibcas.ac.cn)

宋任涛 (中国农业大学, CAU; rentaosong@cau.edu.cn)

谭保才 (山东大学, SDU; bctan@sdu.edu.cn)

汤继华 (河南农业大学, HENAU; tangjihua1@163.com)

田 丰 (中国农业大学, CAU; ft55@cau.edu.cn)

王柏臣 (中国科学院植物研究所, IB-CAS; wangbc@ibcas.ac.cn)

王海洋 (华南农业大学, SCAU; whyang@scau.edu.cn)

王振华 (东北农业大学, NEAU; zhenhuawang_2006@163.com)

巫永睿 (中国科学院上海生命科学研究院, SIBS; yrwu@sibs.ac.cn)

杨 琴 (西北农林科技大学, NWSUAF; yq_smile_745@163.com)

张宪省 (山东农业大学, SDAU; zhangxs@sdau.edu.cn)

(按姓氏首字母排序)

执行委员会主席(Chair):

巫永睿 (中国科学院上海生命科学研究院, SIBS; yrwu@sibs.ac.cn)

执行委员会委员 (Members):

李 林 (华中农业大学, HZAU; hzaulilin@mail.hzau.edu.cn)

卢艳丽 (四川农业大学, SICAU; yanli.lu82@hotmail.com)

秦 峰 (中国农业大学, CAU; qinfeng@ibcas.ac.cn)

孙 峰 (山东大学, SDU; epusun@sdu.edu.cn)

田 丰 (中国农业大学, CAU; ft55@cau.edu.cn)

汤继华 (河南农业大学, HENAU; tangjihua1@163.com)

巫永睿 (中国科学院上海生命科学研究院, SIBS; yrwu@sibs.ac.cn)

(按姓氏首字母排序)

地方组委会 (Local Organizing Committee):

陈彦惠, 刘宗华, 李玉玲, 吴建宇, 汤继华, 胡彦民, 席章营, 库丽霞, 苟明月, 王桂凤, 吴连成, 吴刘记, 丁俊强, 丁冬, 付志远, 李卫华, 李浩川, 董永彬, 郭战勇, 薛亚东, 史勇, 陈甲法, 张战辉, 徐莉萍, 张雪海, 陈晓阳, 王洪秋, 母小焕, 李见坤, 姚文, 陈洪宇, 张向歌, 杨孟莉, 李敏, 赵晨云

摘要编辑 (Abstract Editors):

丁俊强, 董永彬, 陈晓阳, 张战辉, 李见坤

大会秘书处 (Secretariat of the Conference):

大会秘书:	张雪海 17729756037
	李卫华 15838022312
食宿负责人:	吴连成 13733178282
财务负责人:	徐莉萍 17131530966
后勤负责人:	李浩川 15136176256
报到/注册负责人:	王桂凤 18321367864
接待调度负责人:	苟明月 13592581762
会场秩序负责人:	郭战勇 15037294889
墙报负责人:	丁俊强 13838542670

第四届全国玉米生物学学术研讨会日程表

The 4th Maize Biology Conference of China

Schedule of Events (April 19-22)

		会议安排 Program	地点 Location
4.19	08:30-22:00	会议报到注册 Registration	黄河迎宾馆 9 号楼一层大厅 The lobby of Building 9
	08:30-24:00	酒店入住 Hotel Check-in	各自入住酒店
	17:30-19:30	自助晚餐 Dinner (Buffet)	9 号楼 1 楼天香苑餐厅 10 号楼 1 楼富丽宫餐厅 Tianxiangyuan, Building 9 (Lobby Level) Fuligong, Building 10 (Lobby Level)
	19:30-21:00	特邀报告 Plenary Lecture - Session 1	中华厅 Zhonghua Hall
	15:00-23:00	墙报张贴及交流 Informal poster viewing	梅花厅 Meihua Hall
4.20	08:30-08:45	开幕式 Opening Ceremony	中华厅 Zhonghua Hall
	08:45-09:50	专题报告 2: 基因克隆及作物遗传改良 I Session 2: Gene Cloning & Crop Genetic Improvement I	中华厅 Zhonghua Hall
	09:50-10:25	茶歇、合影留念 Tea Break & Group Photo	中华厅室外, 梅花厅和荷花厅门口 Entrance of Zhonghua Hall (Outdoor) & Entrance of Meihua Hall and Hehua Hall
	10:25-12:00	专题报告 3: 植物发育 I Session 3: Plant Development I	中华厅 Zhonghua Hall
	12:00-14:00	自助午餐 Lunch (Buffet)	9 号楼 1 楼天香苑餐厅 10 号楼 1 楼富丽宫餐厅 Tianxiangyuan, Building 9 (Lobby Level) Fuligong, Building 10 (Lobby Level)
	14:00-16:15	专题报告 4: 多维组学 Session 4: Multi-Omics	中华厅 Zhonghua Hall

	16:15-17:15	墙报交流 (奇数编号) Poster Session (Odd-numbered posters) & Hospitality	梅花厅 Meihua Hall
	17:15-18:15	墙报交流 (偶数编号) Poster Session (Even-numbered posters) & Hospitality	梅花厅 Meihua Hall
	18:15-20:00	自助晚餐 Dinner (Buffet)	9 号楼 1 楼天香苑餐厅 10 号楼 1 楼富丽宫餐厅 Tianxiangyuan, Building 9 (Lobby Level) Fuligong, Building 10 (Lobby Level)
	18:15-22:00	墙报自由交流时间 Poster Session & Reception Party - Informal poster time	梅花厅 Meihua Hall
	20:00-21:30	组委会会议 Committee Meeting	迎宾会堂第一会议室 1 st Conference Room
4.21	08:30-10:10	专题报告 5: 基因克隆及作物遗传改良 II Session 5: Gene Cloning & Crop Genetic Improvement II	中华厅 Zhonghua Hall
	10:10-10:35	茶歇 Tea Break	梅花厅和荷花厅门口 Entrance of Meihua Hall and Hehua Hall
	10:35-12:00	专题报告 6: 生物及非生物抗性 Session 6: Biotic & Abiotic Resistance	中华厅 Zhonghua Hall
	12:00-14:00	自助午餐 Lunch (Buffet)	9 号楼 1 楼天香苑餐厅 10 号楼 1 楼富丽宫餐厅 Tianxiangyuan, Building 9 (Lobby Level) Fuligong, Building 10 (Lobby Level)
	14:00-15:05	专题报告 7: 植物发育 II Session 7: Plant Development II	中华厅 Zhonghua Hall
	15:05-16:35	专题报告 8: 育种技术 Session 8: Breeding Technology	中华厅 Zhonghua Hall
	15:35-15:50	茶歇 Tea Break	梅花厅和荷花厅门口 Entrance of Meihua Hall and Hehua Hall
	16:35-17:00	闭幕式 Closing Ceremony	中华厅 Zhonghua Hall

	17:30-19:30	自助晚餐	9 号楼 1 楼天香苑餐厅 10 号楼 1 楼富丽宫餐厅 Tianxiangyuan, Building 9 (Lobby Level) Fuligong, Building 10 (Lobby Level)
4.22	08:30-19:00	离会 Adjournment	

报告日程

Conference Schedule

2019 年 4 月 19 日 星期五	
08:30-22:00 会议报到注册 Registration	
地点: 9 号楼一楼大厅	
19:30-21:00 特邀报告	Plenary Lecture - Session 1
地点: 中华厅	Chair: 谭保才 (山东大学 教授)
19:30-20:15 PT1	
Prof. L. Brian A. Larkins (University of Nebraska-Lincoln, USA)	Maize kernel development: Still much to learn!
20:15-21:00 PT2	
Prof. A. Mark Settles (University of Florida, USA)	Endosperm cell differentiation requires efficient splicing of minor introns
15:00 - 23:00 墙报张贴及交流	Informal poster viewing
地点: 梅花厅	
2019 年 4 月 20 日 星期六	
08:30 - 08:45 开幕式 Opening Ceremony	Chair: 吴建宇 (河南农业大学 教授)
地点: 中华厅	河南农业大学张改平校长致开幕词 The opening statement by President Gaiping Zhang of Henan Agricultural University
08:45 - 09:50 专题报告	专题报告 2: 基因克隆及作物遗传改良 I
地点: 中华厅	Session 2: Gene Cloning & Crop Genetic Improvement I
	Chair: 巫永睿 (中国科学院上海植物生理生态所 研究员)
08:45 - 09:30 PT3	
韩斌 (中国科学院上海植物生理生态所 研究员)	水稻复杂性状和杂种优势的遗传基础
09:30 - 09:50 IT1	
陈化榜 (中国科学院遗传与发育生物学研究所 研究员)	玉米 <i>Ga1</i> 和 <i>Ga2</i> 位点的分子遗传
09:50 - 10:25	茶歇及合影留念 (Tea Break & Group Photo)
10:25 - 12:00 专题报告	专题报告 3: 植物发育 I
地点: 中华厅	Session 3: Plant Development I
	Chair: 李小琴 (仲恺农业工程学院 教授)
10:25 - 11:10 PT4	
李建生 (中国农业大学 教授)	玉米品质的分子育种
11:10 - 11:30 IT2	

王桂凤（河南农业大学 教授）	Evolutionary divergence of the plant MCIA complex that is required for mitochondrial complex I assembly and seed development
11:30- 11:45 ST1	
黄永财（中国科学院上海植物生理生态所）	Maize <i>VKSI</i> Regulates Mitosis and Cytokinesis during Early Endosperm Development
11:45 - 12:00 ST2	
陈永强（河南农业大学）	<i>ZmcytMdh4</i> is essential for storage reserve synthesis and seed development in maize
14:00 - 16:15 专题报告 地点：中华厅	专题报告 4: 多维组学 Session 4: Multi-Omics Chair: 赖锦盛（中国农业大学 教授）
14:00 - 14:45 PT5	
Prof. Xiuren Zhang（Texas A&M University, USA）	Mechanism of miRNA production in plants
14:45 - 15:05 IT3	
李林（华中农业大学 教授）	利用基因组大数据系统解析玉米株高变异分子机制
15:05 - 15:25 IT4	
杨万能（华中农业大学 教授）	室内/室外高通量表型技术研究及玉米遗传解析
15:25 - 15:45 IT5	
王向峰（中国农业大学 教授）	玉米理想基因组智能设计
15:45 - 16:00 ST3	
罗成（华中农业大学）	Single gametophyte sequencing reveals that crossover events differ between sexes in maize
16:00 - 16:15 ST4	
许光辉（中国农业大学）	Metabolome divergence during maize domestication
16:15- 22:00 墙报交流环节 地点：梅花厅	Poster Session
16:15 - 17:15	墙报交流（奇数编号，请墙报作者到场）
17:15 - 18:15	墙报交流（偶数编号，请墙报作者到场）
18:15 - 22:00	墙报自由交流时间（会务组提供茶点及饮料）
20:00 - 21:30	组委会会议（迎宾会堂第一会议室）

2019 年 4 月 21 日 星期日	
08:30 - 10:10 专题报告 地点: 中华厅	专题报告 5: 基因克隆及作物遗传改良 II Session 5: Gene Cloning & Crop Genetic Improvement II Chair: 卢艳丽 (四川农业大学 教授)
08:30 - 09:15 PT6 曹晓风 (中国科学院遗传与发育生物学研究所 研究员)	Mechanisms and functions of dynamic histone methylation in higher plants
09:15 - 09:35 IT6 杨小红 (中国农业大学 教授)	高油玉米人工选择的遗传基础
09:35 - 09:50 ST5 刘红军 (山东农业大学 教授)	Measurement of high frequency and variability rearrangement at the 27-kDa γ -zein locus creates a novel allele for Quality Protein Maize breeding
09:50 - 10:10 IT7 王海洋 (华南农业大学 教授)	玉米现代育种过程中的全基因组选择与遗传改良
10:10 - 10:35	茶歇 (Tea Break)
10:35 - 12:00 专题报告 地点: 中华厅	专题报告 6: 生物及非生物抗性 Session 6: Biotic & Abiotic Resistance Chair: 徐明良 (中国农业大学 教授)
10:35 - 10:55 IT8 苟明月 (河南农业大学 教授)	自发免疫突变体-解析玉米抗病机理的有利工具
10:55 - 11:15 IT9 杨琴 (西北农林科技大学 教授)	重要玉米真菌病害抗病基因挖掘及利用
11:15 - 11:30 ST6 邓穗宁 (中国农业大学)	A helitron-induced <i>RabGDIa</i> variant causes quantitative recessive resistance to maize rough dwarf disease
11:30 - 11:45 ST7 周子键 (河南农业大学)	Variations in an LRR-Receptor Like Kinase Gene, <i>SRR1</i> , Impart Different Quantitative Resistance to Seed and Ear Rot in Maize Heterotic Groups
11:45 - 12:00 ST8 骆美洁 (北京市农林科学院)	Cloning and characterization of a salt tolerance related gene in maize
14:00 - 15:05 专题报告 地点: 中华厅	专题报告 7: 植物发育 II Session 7: Plant Development II Chair: 严建兵 (华中农业大学 教授)
14:00 - 14:20 IT10 贺岩 (中国农业大学 教授)	玉米减数分裂: 知否, 知否?
14:20 - 14:35 ST9 宁强 (华中农业大学)	A new role for ethylene as a developmental signal controlling ear length and kernel number in maize

14:35 - 14:50	ST10 李洪超（中国农业大学）	Perturbation of the C terminus of <i>ZmSDW3</i> alters plant architecture in maize (<i>Zea mays</i> L.)
14:50 - 15:05	ST11 王乐乐（雷根斯堡大学）	The RALF-LRX-ANX/BUPs signaling pathway is conserved during pollen tube growth in maize
15:05 - 16:35	专题报告 地点：中华厅	专题报告 8: 育种技术 Session 8: Breeding Technology Chair: 程备久（安徽农业大学 教授）
15:05 - 15:20	ST12 谢传晓（中国农业科学院 研究员）	Genome Editing and Double Fluorescence Proteins Enable Robust Maternal Haploid Induction and Identification in Maize
15:20 - 15:35	ST13 王宝宝（中国农业科学院）	Accelerate maize breeding by Inducer Mediated Genomic Editing (IMGE)
15:35 - 15:50		茶歇（Tea Break）
15:50 - 16:35	PT7 朱健康（中国科学院上海植物逆境生物学研究中心 研究员）	Developing gene editing tools for functional genomics research and crop breeding
16:35 - 17:00	闭幕式 Closing Ceremony 地点：中华厅	Chair: 汤继华（河南农业大学 教授）
16:35 - 16:50		- 颁奖典礼 Award ceremony
16:50 - 16:55		- 下届承办单位致欢迎词 Welcome remark by next organizer
16:55 - 17:00		- 谭保才主席致闭幕词 Closing remark
2019 年 4 月 22 日 星期一		
离会 Adjournment		

Abstract List

Abstract	Submitted Author	Title
A1	安婷婷	Maize genotypes with contrasting root system architecture responded differently to drought stress
A2	安学丽	<i>ZmMs30</i> Encoding a Novel GDSL Lipase Is Essential for Male Fertility and Valuable for Hybrid Breeding in Maize
A3	曹岩	MicroRNAs Are Involved in Maize Immunity against <i>F. verticillioides</i> -associated Ear Rot
A4	曾兴	Effects of temperature, water content, pH and soil sterilization on the degradation of CP4-EPSPS protein released from herbicide-tolerant corn leaves in the soil
A5	陈甲法	Inheritance of resistance to <i>Rhopalosiphum maidis</i> in maize
A6	陈青云	玉米中 <i>STOMAGEN-Like</i> 基因的功能分析
A7	陈曙	8 个玉米泛素结合酶家族基因对低氮胁迫的响应模式分析
A8	陈思佳	Translatome Profiling Shows Widespread Complete Dominance associated with Heterosis in Maize
A9	陈应龙	Phenotyping root system architecture in maize
A10	陈永强	<i>ZmcytMdh4</i> encoding a cytosolic malate dehydrogenase is essential for storage reserve synthesis and seed development in maize
A11	陈园园	Genome wide association study revealing the genetic basis of ionomic variations in maize
A12	代明球	Functional characterization of <i>ZmMAP3K1</i> and the association of its natural variations in maize drought tolerance
A13	戴大伟	Maize <i>Dek33</i> encodes a pyrimidine reductase in riboflavin biosynthesis essential for oilbody formation and ABA biosynthesis during seed development
A14	邓穗宁	A <i>helitron</i> -induced RabGDIa variant causes quantitative recessive resistance to maize rough dwarf disease
A15	杜邓襄	Construction and application of a genome-wide maize overexpression mutant library
A16	杜晓夏	Genetic architecture of natural variation in maize embryo size
A17	杜艳芳	<i>KRN4</i> controls kernel row number by long-distance regulation of <i>UB3</i> expression in maize

A18	段志坤	玉米 PYS1 蛋白调控叶绿体发育的分子机理研究
A19	冯文奇	玉米转录因子 <i>BES1/BZR1-7</i> 基因克隆及功能分析
A20	冯宣军	Variation of <i>ZmDRO1</i> gene to ABA response confers the difference of drought avoidance between modern maize and <i>Zea mays</i> ssp. <i>mexicana</i>
A21	冯雪	Characterization and fine mapping of stay-green loci for leaf senescence in maize (<i>Zea mays</i> L.)
A22	冯阳	Maize <i>Dek45</i> encodes a non-zein PB protein that interacts with 22kD zein
A23	冯雨	A new subclass of the mitochondrial-localized RNA-binding proteins required for seed development in maize and <i>Arabidopsis</i>
A24	付春祥	玉米棕色叶脉突变体基因鉴定及功能分析
A25	付修义	Genetic basis of selection for kernel nutritional traits during maize domestication and improvement
A26	高景阳	Linkage mapping and GWAS reveal candidate genes conferring thermo-tolerance of seed-set in maize
A27	宫捷	甜玉米自然老化其生理生化机制分析
A28	龚央庆	The host-virus protein interaction network in maize resistance to sugarcane mosaic virus
A29	苟明月	New insight into lignin biosynthesis: P450 complex formation and electron shuttle
A30	顾日良	Identification of Quantitative trait loci associated with drought tolerance at reproductive stage in maize (<i>Zea mays</i> . L) across multi-environments
A31	郭明月	Two Transcripts of <i>ZmHsf12</i> Have Distinct Functions in <i>Arabidopsis</i> Under Heat Stress
A32	郭新梅	Cloning and expression of <i>ZmXTH23</i> in maize (<i>Zea mays</i>) and its response to salt and drought stress
A33	郭战勇	Cloning and characterization of <i>ZmELS2</i> gene that regulates leaf pre-senescence in maize
A34	韩兆雪	Phylogenetic analysis and synteny network characterize the conserved and diverse evolutionary patterns of <i>NAC</i> gene families in plants
A35	何永辉	Maize <i>Dek15</i> encodes the cohesin-loading complex subunit SCC4 and is essential for chromosome segregation and kernel development

A36	陈翠霞	玉米苞叶层数主效 QTL- <i>qHN1</i> 的精细定位
A37	胡龙	玉米抗灰斑病主效 QTL <i>qRglsB11</i> 的定位
A38	胡明建	Cloning and function analysis of <i>ZmCTL</i> gene associated with seed development in maize
A39	胡书婷	Genome-wide association study deciphers the genetic architecture of carbohydrate partitioning in maize kernels
A40	胡小娇	Genome-wide proteomic profiling reveals the role of dominance protein expression in heterosis in immature maize ears
A41	胡晓丹	Assessment of phytochemical biosynthesis and accumulation in black sweet corn during kernel development
A42	黄帝	玉米根系响应干旱胁迫的研究进展
A43	黄圣禅	The creation of high amylose maize material by CRISPR/Cas9 gene editing approach
A44	黄永财	Maize <i>VKS1</i> is Essential for Early Endosperm Development by Regulating Mitosis and Cytokinesis
A45	冀晨	The molecular mechanism of the maize PLATZ transcription factor family involved in maize endosperm development and storage filling
A46	贾宏	Positional cloning of <i>qDTA6</i> controlling flowering time in maize
A47	贾利强	An evolutionary and expressional pattern of the bZIP family in maize
A48	姜程淋	Detailed characterization of non-crossover recombination event spectrum by single-microspore sequencing in maize and rice
A49	姜露光	Characterization of proteome variation during modern maize breeding
A50	金立方	Editing of 19-kD and 22-kD α -zein gene families by CRISPR/Cas9 gene editing approach in maize
A51	金敏	Genome wide association study underlying the genetic basis of RFOs metabolic pathway in maize
A52	金如昌	Specific expression of <i>GS5</i> gene in endosperm increases maize yield
A53	晋高锐	Stable-isotope labeling for quantitative peptidomics of maize treated by <i>Rhizoctoniasolani</i> stress and <i>Bipolaris maydis</i> stress

A54	李圣彦	Different mechanisms regulate the expression of <i>TPS6</i> to improve the disease resistance and drought resistance in maize
A55	黎颀	Genetic analysis of <i>cis</i> -regulatory imprint control at the <i>Floury3</i> locus
A56	李保珠	Flavonols affects drought tolerance through regulating stomatal movement and alleviating oxidative damage in maize seedlings
A57	李朝斌	The <i>ZmbZIP22</i> transcription factor regulates 27-kD γ -zein gene transcription during maize endosperm development
A58	李洪超	Perturbation of the C terminus of ZmSDW3 alters plant architecture in maize (<i>Zea mays</i> L.)
A59	李见坤	<i>Mutator</i> 转座子介导的玉米籽粒发育突变体的基因克隆
A60	李结平	<i>ZmRAP2.7</i> , an AP2 transcription factor, is involved in maize brace roots development via <i>miR156-miR172</i> pathway
A61	李露露	玉米快速叶绿素荧光参数与叶片表型相关性状的 QTL 分析
A62	李曼菲	<i>KERNEL NUMBER PER ROW6</i> encodes a serine/threonine protein kinase to regulate grain yield by phosphorylating an ARF-GAPase activating protein in maize
A63	李圣彦	Different mechanisms regulate the expression of <i>TPS6</i> to improve the disease resistance and drought resistance in maize
A64	李婷	Combining genome-wide selected region sweep and association mapping to identify ear appearance genes in maize
A65	李文宗	在玉米籽粒中合成长链多不饱和脂肪酸 EPA 和 DHA 的研究进展
A66	李杨	份新的玉米矮秆突变体的遗传分析与图位克隆
A67	李玉斌	Serendipitous treasures: the maize transposable elements <i>Dotted</i> , <i>Mrh</i> and more
A68	李志刚	Genome-wide association study to dissect genetic basis of stay-green in maize under drought stress
A69	李志鹏	Genetic determinant controlling maize Rubisco activase gene expression and a comparison with rice counterpart
A70	李紫文	The ceRNA-miRNA-target gene regulatory networks contribute to anther development in maize
A71	梁磊	A sequence-indexed <i>Mutator</i> insertional library as an important resource for maize functional genomics study

A72	刘保兴	玉米/大豆间作下磷高效吸收的根系生物学机制研究进展
A73	刘柄良	Genome-wide analysis of LAZ1 gene family from maize
A74	刘博欣	The study of the molecular mechanism of drought stress affecting the anthesis and silking interval
A75	刘红军	High frequent DNA rearrangement at the 27-kD γ -zein locus creates a superior <i>o2</i> modifier for Quality Protein Maize breeding
A76	刘建菊	Identification and high-resolution mapping of quantitative trait loci related to grain water content in maize
A77	刘丽丽	Association mapping of stay-green traits for the panel adapted to China northern spring maize planting region
A78	刘升学	Genetic Components Controlling Drought Responsiveness in Maize Seedlings
A79	柳波娟	<i>Actin Depolymerizing Factor 5</i> was Identified Contribution to the Regulation of Drought Tolerance in Maize
A80	柳小庆	Engineering of ‘Purple Maize’ with a multigene expression system
A81	卢晓环	玉米株高 <i>qPH10</i> 的 QTL 定位及候选基因预测
A82	陆格格	Identification and characterization of three phytosterol synthesis related genes in Maize
A83	路风中	<i>ZmPP2Ca</i> gene regulating drought tolerance in maize by alternative splicing
A84	路小铎	基因索引的玉米突变体库助力功能基因组学研究
A85	罗成	Single gametophyte sequencing reveals that crossover events differ between sexes in maize
A86	骆美洁	Molecular dissection of maize seedling salt tolerance using a genome-wide association analysis method
A87	马彪	<i>ZmMs7</i> functions as a transcriptional activator to regulate anther and pollen development in maize
A88	马德志	黑龙江省三、四积温带玉米新品种抗倒伏性比较研究
A89	马培培	Resistance of Seed Internal Tissues to <i>Fusarium verticillioides</i> in Maize: Identification Methods, Resistant Lines, and Breeding Strategies
A90	马鹏	Integrate transcriptome and GWAS for functional characterization of long non-coding RNAs under deficient nitrogen in maize seedling
A91	马雪霞	Characterization of a new maize high-lysine mutant

A92	孟淑君	Fine mapping of the main QTL of ear length in maize
A93	母小焕	Gibberellins synthesis is involved in the reduction of cell flux and elemental growth rate in maize leaf under low nitrogen supply
A94	倪嘉成	A novel protein is essential for mitochondrial function and seed development in maize
A95	宁丽华	Maize endosperm-specific transcription factor <i>PBF1</i> dynamically regulates targeted genes balancing protein and carbohydrate storage in a nitrogen dependent manner
A96	宁强	A new role for ethylene as a developmental signal controlling ear length and kernel number in maize
A97	彭奕萱	Cloning and characterization of a maize kernel defective mutant encodes a PPR protein
A98	齐欣	玉米不同回交世代 DH 系遗传分析研究
A99	祁显涛	Systematic identification of endogenous RNA polymerase III promoters for efficient RNA guide based genome editing technologies in maize
A100	钱佳	Dynamic patterns of protein-coding and noncoding elements across maize development in a maize F₁ hybrid and its inbred parents
A101	秦文超	Dissecting the genetic architecture that controls the domestication of maize leaf morphology
A102	渠建洲	Evolutionary, structural and expression analysis of core genes involved in starch synthesis
A103	曲彦志	基于核磁共振的玉米不同籽粒类型单粒质量和含油率分析
A104	任如昌	<i>DEK43</i> Regulates Maize Kernel Development by Affecting <i>nad4</i> Splicing
A105	任珍静	Maize Empty pericarp602 encodes a P-type PPR protein that is essential for seed development
A106	时夏	Dissecting Heterosis During the Ear Inflorescence Development Stage in Maize via a Metabolomics-based Analysis
A107	宋楠楠	Overexpression of maize heat stress transcription factor gene <i>ZmHsf14</i> enhances thermo tolerance in transgenic <i>Arabidopsis</i>
A108	苏慧慧	Dual functions of <i>ZmNF-YA3</i> in photoperiod dependent flowering and abiotic stress responses in maize
A109	孙飞	Fine Mapping of <i>qLA3</i> Controlling Leaf Angle in Maize

<i>A110</i>	孙峰	<i>Empty pericar23</i> encoding a mitochondrial pentatricopeptide repeat protein, is involved in the cis-splicing of <i>nad1</i> intron 2 and seed development in maize
<i>A111</i>	孙瑞琪	细胞周期转换蛋白 APC/C ^{ZmCCS52B} 调节玉米生物量的研究
<i>A112</i>	孙霄鹏	A large-scale circular RNA profiling reveals universal molecular mechanisms responsive to drought stress in maize and <i>Arabidopsis</i>
<i>A113</i>	田有辉	Map-based cloning, phylogenetic and microsynteny analyses of <i>ZmMs20</i> gene regulating male fertility in maize
<i>A114</i>	万炯	顺反调控多态性调控玉米籽粒萌发杂种优势形成的网络解析
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<i>A119</i>	王国英	Characterization and fine mapping of a novel defective kernel mutant in maize
<i>A120</i>	王海海	A natural allele <i>vitreous endosperm1</i> regulates endosperm texture in maize seed
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<i>A122</i>	王鹤群	玉米 <i>PIN1a</i> 基因异源表达及介导百脉根-AM 真菌共生功能分析
<i>A123</i>	王宏泽	Fine mapping of <i>qLM</i> , controlling lesion mimic and multiple disease resistance in maize
<i>A124</i>	王洪秋	Identification of heterotic loci associated with grain yield and its components using two CSSL test populations in maize
<i>A125</i>	王璟璐	Research progress and prospect on Crop Phenomics database
<i>A126</i>	王群	Positional cloning of maize endosperm breakdown1 mutant
<i>A127</i>	王帅	Genome-wide identification and expression analysis of Dof (DNA binding with one finger) protein family in monocot and dicot species
<i>A128</i>	王夏青	Dynamic plant height QTL revealed in maize through remote sensing phenotyping using a highthroughput unmanned aerial vehicle (UAV)

<i>A129</i>	王怡凯	Fine mapping of <i>opaque endosperm 5(oe5)</i> , a gene controlling amino acid content in maize
<i>A130</i>	王永彦	Updating and interaction of ploycomb reprssive complex2 compenents in maize (<i>Zea mays</i>)
<i>A131</i>	王勇健	Three-dimensional shoot architecture evaluation of different maize varieties at early growth stage
<i>A132</i>	王振华	Analysis of cytology and expression of resistance genes in maize infected with <i>Sporisorium reilianum</i>
<i>A133</i>	魏一鸣	<i>APP1</i> Encoding a P-Loop NTPase Is Involved in Maize Kernel Development
<i>A134</i>	吴锋锴	玉米 <i>ZmLBD1</i> 响应缺磷胁迫和调节根系生长的功能研究
<i>A135</i>	吴刘记	Comparative proteomics combined with analyses of transgenic plants reveal <i>ZmREM1.3</i> mediates maize resistance to southern corn rust
<i>A136</i>	夏进良	Fine mapping of a quantitative trait locus for flowering time in maize
<i>A137</i>	夏宗良	硫代谢关键基因 <i>ZmSO</i> 调控玉米抗旱性的分子机制
<i>A138</i>	向楠	The effects of extreme temperatures on vitamins and their biosynthesis pathways in sweet corn (<i>Zea mays</i> L.) seedlings
<i>A139</i>	肖英杰	Genetic basis and prediction for 20 agronomic traits from inbred and hybrid populations in maize
<i>A140</i>	谢传晓	Genome Editing and Double Fluorescence Proteins Enable Robust Maternal Haploid Induction and Identification in Maize
<i>A141</i>	邢嘉韵	Proteomics Analysis of Two Maize Photo-Thermo-Sensitive Tasseless Lines
<i>A142</i>	熊静	<i>ZmLBD44</i> interacts with <i>ZmEXP7</i> to regulate root development and drought response in maize
<i>A143</i>	熊瑞	Gene expression and activity analyses of two terpene synthases responsible for biosynthesis of aroma compounds in <i>Osmanthus fragrans</i>
<i>A144</i>	熊子墨	Preliminarily analyzing the molecular mechanism of maize heterosis formation by high-throughput yeast two hybrid
<i>A145</i>	徐春晖	<i>EMB15</i> functions in plastid 30S ribosome assembly and embryogenesis in maize
<i>A146</i>	徐芳	The CLAVATA receptor <i>FASCIATED EAR2</i> functions through both heterotrimeric G-protein and β subunit in meristem development

<i>A147</i>	徐静	Genome-wide association study reveals the causes and consequences of natural variation in DNA methylation in maize
<i>A148</i>	徐莉萍	<i>Arabidopsis</i> BRASSINOSTEROID INACTIVATOR2 is a typical BAHD acyltransferase involved in brassinosteroid homeostasis
<i>A149</i>	许光辉	Evolutionary metabolomics identifies significant metabolic divergence between maize and its wild ancestor, teosinte
<i>A150</i>	严佳丽	Analysis of maize mutant library based on CRISPR/Cas9
<i>A151</i>	杨欢欢	A P-type pentatricopeptide repeat protein is involved in the splicing of multiple mitochondrial introns and seed development in maize
<i>A152</i>	杨继伟	Genetic Dissection of Haploid Male Fertility in Maize (<i>Zea mays</i> L.)
<i>A153</i>	杨俊	玉米草酸降解途径解析和籽粒品质遗传改良
<i>A154</i>	姚丽珊	整合关联分析和转录组分析挖掘玉米抗轮枝镰孢菌穗粒腐病基因
<i>A155</i>	姚天茏	The Genetic Architecture of Chlorophyll Content by Linkage and Association Analysis in Maize
<i>A156</i>	姚文	ECOGEMS: efficient compression and retrieve of SNP data of 2058 rice accessions with integer sparse matrices
<i>A157</i>	姚兴兰	富含维生素 E 和植酸酶的玉米新材料创制
<i>A158</i>	于延文	Ascorbic acid integrates the antagonistic modulation of ethylene and abscisic acid in the accumulation of reactive oxygen species
<i>A159</i>	余国武	Preparation of NAC78 antibody and its expression under salt stress
<i>A160</i>	余延辉	Fine mapping and functional analysis of <i>qDRI-2</i> for the dehydration rate of maize kernels
<i>A161</i>	袁跃	The Maize <i>Opaque 2 Regulated Kinase 1</i> (ORK1) Phosphorylates O2 and Reduces its Transcription Activation Activity
<i>A162</i>	张敖	Identification of maize brace-root quantitative trait loci in a recombinant inbred line population
<i>A163</i>	张博琦	The maize immune receptor ZmWAK recognizes secreted proteins from <i>Sporisorium reilianum</i>
<i>A164</i>	张红伟	Genomic prediction of the testcross performance across multiple populations

<i>A165</i>	张卉	PTPP, a nucleotidase encoding gene, is activated by the heat shock transcription factor <i>HSFA6a</i> and positively regulates ascorbic acid biosynthesis and drought tolerance
<i>A166</i>	张茂林	Engineering Quantitative Trait Variation in maize by genome editing
<i>A167</i>	张敏	Genome-wide transcriptome analysis of miRNAs and mRNAs in maize for high-density planting
<i>A168</i>	张明亮	Functional Study of An <i>O2</i> Downstream Target Gene
<i>A169</i>	张清华	两个野生玉米回交 RIL 群体的 QTL 定位
<i>A170</i>	张珊珊	Identification and characterization of three phytosterol synthesis related genes in Maize
<i>A171</i>	张体付	基于玉米基因组 PAV 结构变异及 5 个组织 RNA-Seq 数据的多性状全基因组预测
<i>A172</i>	张蜓	<i>ZmBRCA2</i> is dispensable for RAD51/DMC1-facilitated homologous recombination in maize meiosis
<i>A173</i>	张霞	Multivariate analyses of root phenotype and dynamic transcriptome underscore valuable root system architectures and drought responsive gene networks in maize
<i>A174</i>	张小敏	Genetic variations in <i>ZmTIP1</i> are critical for root hair Development and Drought Tolerance of Maize Seedlings
<i>A175</i>	张晓聪	Integrated transcriptome, small RNA, and degradome analysis reveals the complex network regulating starch biosynthesis in maize
<i>A176</i>	张啸	Characteristics of copy number variations and functional impact in maize population
<i>A177</i>	张战辉	The application of Short Tandem Target Mimic-mediated microRNAs inactivation in <i>Arabidopsis</i> and maize agronomic traits improvement
<i>A178</i>	张智超	玉米抗禾谷镰刀菌茎腐病 QTL 定位
<i>A179</i>	张中保	Distribution of four bioactive flavonoids in maize tissues of five varieties and correlation with expression of the biosynthetic genes
<i>A180</i>	张忠琴	Identification and Characterization of Maize ACD6-like Gene Reveal that <i>ZmACD6</i> Is The Maize Orthologue Conferring Resistance to Ustilago Maydis
<i>A181</i>	章琰	Acquisition of root-specific gene knockout transgenic plants by CRISPR/Cas9 gene editing approach in maize

<i>A182</i>	赵炳浩	Genetic architecture of maize stalk rind penetrometer resistance in linkage and association mapping populations
<i>A183</i>	赵秋芳	Characterization and the Expression Analysis of Nitrate Transporter (NRT) Gene Family in maize
<i>A184</i>	郑喜喜	Maize VP1-mediated ABA pathway is essential for proteome rebalancing between endosperm and embryo
<i>A185</i>	钟海旭	Exploring genetic basis of regulation in root to response low-phosphorus stress by <i>ZmPAT1</i> in maize
<i>A186</i>	钟涛	Identification and characterization of a major QTL- <i>qRgls1</i> against gray leaf spot in maize
<i>A187</i>	周巧	MicroRNA 对其靶基因的调控模式分析
<i>A188</i>	周清倩	A microtubule-associated protein was found to be involved in maize grain development
<i>A189</i>	周羽	Molecular characteristics of <i>segment 5</i> , a unique fragment encoding two partially overlapping ORFs in the genome of rice black-streaked dwarf virus
<i>A190</i>	周昱婕	优质蛋白玉米的研究进展
<i>A191</i>	周子键	Variations in an LRR-Receptor Like Kinase Gene, <i>SRRL</i> , Impart Different Quantitative Resistance to Seed and Ear Rot in Maize Heterotic Groups
<i>A192</i>	朱晨光	The PPR protein DEK41 affects cis-splicing of mitochondrial <i>nad4</i> intron 3 and seed development in maize
<i>A193</i>	朱万超	Gene Mapping and Cloning of Two Maize Kernel Mutants
<i>A194</i>	宗娜	玉米中介体亚基 ZmMED7 的初步功能分析
<i>A195</i>	左毅	<i>Dek42</i> encodes an RNA binding protein that affects alternative pre-mRNA splicing and maize kernel development

Abstracts

Plenary Talk

Plenary Talk 1

Maize kernel development: Still much to learn!

Brian A. Larkins

University of Nebraska-Lincoln, USA



Brian A Larkins's research program focused on the regulation of seed development and the synthesis of seed storage proteins. Storage proteins are the most abundant proteins in seeds, and as such they are the principal determinants of the protein quality of grains. Storage proteins are generally deficient in several amino acids that are required in human and livestock diets. Consequently, increasing the levels of these essential amino acids has long been a goal of plant breeders and cereal chemists. A major focus of his research was Quality Protein Maize, or QPM. The opaque2 mutation increases the content of essential amino acids in the maize kernel, but it also causes a soft

starchy endosperm that creates inferior grain quality. Genetic suppressors of opaque2 (o2 modifiers) were identified that ameliorate the negative phenotypic features of the o2 mutation, but the genes responsible for modification were not well characterized. His lab studied how the o2 mutation increases the lysine content of the grain and how o2 modifiers restore the normal hard, vitreous kernel phenotype. His lab also investigated cell cycle regulation and the role it plays in maize seed development, particularly endoreduplication in the endosperm.

Brian got his B.S. at the University of Nebraska-Lincoln in 1969, and his Ph.D. in Botany at the same university in 1974. Following one-year postdoc research at Purdue University, he became Assistant Professor of Biochemical Genetics there, in the Department of Botany and Plant Pathology. He was promoted to professor in 1984. In 1988 Brian left Purdue and went to the University of Arizona, where he served as head of the Department of Plant Sciences from 1988 to 1994. In 1995, he became the first holder of the Porterfield Chair, an endowed position in that department.

Brian served as an Associate Editor of Plant Physiology at the time when The Plant Cell was just established as a new journal. Following four years as an Associate Editor of Plant Cell he became Editor-in-Chief serving from 1993-1998. In 1998, Dr. Larkins was President of the ASPP. Among his numerous honors and recognition, he received the Charles A. Shull award from ASPP in 1983, for his work on maize seed storage proteins, and the Dennis Robert Hoagland award from ASPP in 1997. In 1996 Dr. Larkins was elected to the National Academy of Science.

In 2012 he moved to the University of Nebraska-Lincoln, where he started, and became associate vice chancellor for life sciences at UNL.

Plenary Talk 2

Endosperm cell differentiation requires efficient splicing of minor introns

A. Mark Settles

University of Florida



A. Mark Settles, Vasil-Monsanto Professor of Plant Cell and Molecular Biology, Horticultural Sciences Department, University of Florida. Mark obtained his Doctor of Philosophy in Genetics State University of New York (SUNY) at Stony Brook, New York in 1998. Work experience: Chair, Maize Genetics Steering Committee, 2015; Co-Chair, Maize Genetics Steering Committee, 2014; Visiting Professor, Federal University of Viçosa, Brazil, October, 2012; Director, Plant Molecular and Cellular Biology Program, University of Florida, 2011-present; Graduate Coordinator, Plant Molecular and Cellular Biology Program, University of Florida, 2009-2011; Vasil-Monsanto Associate Professor, University of Florida, 2007-2013; Vasil-Monsanto Assistant Professor, University of Florida, 2000-2007; Postdoctoral Associate, University of Florida, 1999-2000; Graduate Assistant, Cold Spring Harbor Laboratory, New York, 1994-1998; NIH Pre-doctoral Fellow, SUNY at Stony Brook, New York, 1993-1994. Honors and Awards: University of Florida Research Foundation Professor, 2016; HHMI Science for Life Distinguished Undergraduate Mentor Award, 2010; USDA Post-doctoral Fellowship, 2000. Areas of Research: Maize Genetics and Genomics, Seed Development, Seed Phenomics.

Plenary Talk 3

水稻复杂性状和杂种优势的遗传基础

韩斌

中国科学院上海植物生理生态研究所



韩斌，1985 年本科毕业于安徽师范大学；1988 年硕士毕业广西大学农学院（现广西大学）。1989 年 2 月，获得英国 Gatsby Charitable Foundation 奖学金，赴英国 John Innes Centre (约翰英纳斯研究中心) Sainsbury Laboratory (塞理斯伯里实验室) 攻读博士学位，从事植物病原菌分子遗传学研究，并于 1992 年 11 月获博士学位。1992 年 12 月~1998 年 8 月在英国剑桥大学植物科学系做博士后研究。1998 年 8 月，正值中国水稻基因组测序计划起步阶段，回到中科院国家基因研究中心工作。至今，一直从事水稻基因组学和水稻遗传学研究。现为中国科学院国家基因研究中心主任，研究员，中科院上海生命科学

研究院副院长，中科院上海植物生理生态研究所所长，中科院分子植物卓越创新中心主任。2004 年获得基金委“国家杰出青年基金”资助，2013 年当选为中科院院士，2014 年当选为发展中国家科学院院士。韩斌院士多年来专注于水稻基因组学和遗传学研究，在水稻基因组精确测序、水稻复杂性状的全基因组关联分析、栽培稻的起源驯化和水稻杂种优势分子遗传机制研究上，取得了既有重大理论意义又有重要应用价值的系统性原创性成果，在国际上产生了重要影响，为中国作物遗传学研究的发展做出了重要贡献。发表论文 108 篇，其中 70 篇为通讯和共同通讯作者（包括 3 篇 Nature 和 6 篇 Nature Genetics 论文），2014 年被汤森路透评为全球高被引科学家。2007 年获国家自然科学二等奖（排名第一）和 2017 年获一等奖（排名第二）及 2003 年上海市科技进步一等奖（排名第一）各 1 项；2013 年获“中国科学院杰出成就奖”（主要贡献者）；2017 年获“求是杰出科技成就集体奖”（排名第二）。国家基金委“创新群体学术带头人”，主持基础科学中心。先后担任 Molecular Plant、Plant Journal、eLife 等 9 个国际学术期刊的共同主编或编委，30 余次受邀在国际学术会议上做大会及专题报告，担任国际最著名的植物科学研究中心-英国 John Innes 研究中心科学顾问。

Plenary Talk 4

玉米品质的分子育种

李建生

中国农业大学



李建生，中国农业大学国家玉米改良中心教授。华中农业大学作物遗传育种专业研究生毕业，先后获硕士和博士。1989 年-1992 年，任美国普渡大学农学系客座助理教授。1997 年 6 月-9 月和 1998 年 3 月-8 月，任香港大学植物系客座研究员。1993-2000 年，历任华中农业大学作物遗传改良国家重点实验室，副教授，教授。国务院特殊津贴专家，现任中国作物学会理事、玉米专业化委员会主任。在 *Nature Genetics*、*Nature Communications*、*PNAS*、*Plant Physiology* 等期

刊发表 SCI 论文多篇。曾获国家技术发明二等奖、教育部技术发明一等奖、教育部科技进步一等奖等奖项。

Plenary Talk 5

Mechanism of miRNA Production in Plants

Xiuren Zhang

Texas A&M University, USA



Xiuren Zhang is a Professor at department of Biochemistry and Biophysics and Institute for Plant Genomics & Biotechnology at Texas A&M University. He earned his bachelor degree in Economic Botany from Southern Anhui Agricultural College in 1989, his master degrees in Horticulture from the China Agricultural University in 1994 and Auburn University in 1999 and his doctorate in Plant Biology from Cornell University in 2003. Dr. Zhang did his postdoc training at Dr. Nam-Hai Chua laboratory in Rockefeller University from 2003-2008. His research interests include RNA biology, epigenetic silencing, and abiotic stress. Dr. Zhang and his team members have made several paradigm-shifting discoveries in elucidating new functions and mechanisms of microRNA biogenesis machinery and RNA-induced silencing complexes in plants as well as host/virus interplay at the transcriptional and posttranscriptional levels. Dr. Zhang has published numerous significant publications in the top-tier journals including Cell, Nature and eLife. Schnable has also co-founded four start-up companies.

Plenary Talk 6

Mechanisms and functions of dynamic histone methylation in higher plants

曹晓风

中国科学院遗传与发育生物学研究所



曹晓风，植物表观遗传学家，中科院遗传与发育生物学研究所研究员，基因组生物研究中心主任。1988年毕业于北京大学生物系，1997年获北京大学生命科学学院博士学位。先后在英国 John Innes Centre、美国华盛顿州立大学、美国加州大学洛杉矶分校做博士后研究或工作。先后获得美国“杜邦青年科学家奖”、“中国青年女科学家奖”和中国科学院“十大女杰”等称号。入选国家级“新世纪百千万人才工程”和中组部“万人计划”第一批百千万工程领军人才，并于2015年当选中国科学院院士，2016年当选发展中国家科学院院士。先后担任科技部、农业部、基金委、中科院各类重大任务首席科学家，累计主持重大项目18项。她长期从事植物表观遗传学研究，在植物DNA甲基化、组蛋白共价修饰和小分子RNA等领域取得诸多系统性、原创性成果，累计发表论文100余篇，被SCI论文引用8000次以上，H因子40+。先后担任National Science Review, The Plant Cell, Frontier in Biology, Journal of Genetics and Genomics等国际学术期刊的副主编或编委。

Plenary Talk 7

Developing gene editing tools for functional genomics research and crop breeding

朱健康

中国科学院上海植物逆境生物研究中心



Jian-Kang Zhu is the Director of the Shanghai Center for Plant Stress Biology, Chinese Academy of Sciences, and a Distinguished Professor of Plant Biology, Departments of Horticulture and Landscape Architecture and Biochemistry at Purdue University. He earned his bachelor degree in soil and agricultural chemistry from Beijing Agricultural University in 1987; his master degree in botany from the University of California, Riverside in 1990, and his doctorate in plant physiology from Purdue University in 1993. In 2010, he was elected to be a member of National Academy of Sciences, USA. His research interests include abiotic stress signaling

and tolerance mechanisms in plants, epigenetic gene regulation, and gene targeting and genome engineering technologies.

朱健康，国际著名的植物生物学家、植物抗逆分子生物学领军人物。1987年毕业于北京农业大学土化系，1993年获得美国普渡大学植物生理学博士。2000年受聘美国亚利桑那大学植物科学系正教授。曾任加州大学河边分校整合基因组学研究所所长、Jane Johnson 讲座教授，沙特 KAUST 大学植物逆境基因组中心主任，现为美国普渡大学生物化学系和园艺及园林系杰出教授。2010年朱教授当选为美国科学院院士。2011年10月入选中央首批“千人计划”顶尖人才，2012年4月起负责新建中国科学院上海植物逆境生物研究中心并担任中心主任。朱健康教授主要从事植物逆境分子生物学和表观遗传学研究，另外，他的研究组也致力于通过开发和应用 CRISPR/CAS 和其他基因组工程技术来进行作物改良。曾获得亚利桑那大学生命科学与农学院年度最佳研究员奖和美国植物生物学会颁发的 Charles Albert Shull 奖。迄今为止，朱教授在《自然》、《科学》、《细胞》等世界高水平学术期刊上发表了 300 多篇研究论文，是世界植物科学领域发表文章引用率最高的科学家之一。

Invited Talk

Invited Talk 1

玉米 *Ga1* 和 *Ga2* 位点的分子遗传

陈化榜

中国科学院遗传与发育生物学研究所, 100101

(Submitted by 陈化榜 <hbchen@genetics.ac.cn>)

玉米是雌雄同株的异花授粉作物, 天然异交率达 95%以上。自然界中存在少数玉米材料, 它们只能自交或为其它玉米授粉, 却不能接受其它玉米花粉的单向杂交不亲和现象 (Unilateral Cross-Incompatibility, UCI)。玉米的 UCI 受控于配子体基因 (*Gametophyte factor*, *Ga*), 以近乎完全杂交不育的 *Ga1*、*Ga2* 以及 *Tcb1-S* (*Teosinte crossing barrier 1*) 最具代表性。我们就发现最早、研究最为广泛的 *Ga1* 和 *Ga2* 位点控制的玉米 UCI 现象, 开展了遗传分析、基因克隆与功能验证、不亲和机理的解析, 以及在生产实践中应用的科学研究。遗传分析证明, *Ga1* 和 *Ga2* 位点均由紧密连锁的显性单基因配子体遗传的花粉决定因子, 和隐性单基因孢子体遗传的花丝决定因子构成。通过关联分析和转录组分析以及图位克隆, 分别得到了花粉决定因子 (*ZmGa1P* 和 *ZmGa2P*) 和花丝决定因子 (*ZmGa2P* 和 *ZmGa2S*) 控制基因, 均编码果胶甲酯酶 (Pectin methylesterase, PME)。以 *Ga1* 为例, 转基因实验证明, 表达 *ZmGa1P* 基因的 *gal* 植株具备了突破 *Ga1-S* 植株花丝阻碍的能力, 而敲除 *ZmGa1S* 基因的 *Ga1-S* 植株则能够接受 *gal* 花粉。细胞学观察发现, 不亲和组合的花粉管在 *Ga1-S* 花丝中延伸受阻, 普遍出现末端膨大的表型, 且花粉管顶端的甲酯化程度较亲和组合明显升高。LC-MS/MS、互作分析以及酶活实验表明, *ZmGa1P* 与另一个在花粉中特异高表达的 PME 蛋白 *ZmPME10-1* 形成复合体, 维持 *ZmPME10-1* 在 *Ga1-S* 花丝中的酶活性。进一步研究表明, *ZmGa1P* 和 *ZmGa1S* 虽然不存在直接的相互作用关系, 但均与 *ZmPME10-1* 的 PME 结构域互作, 且 *ZmGa1P* 可与 *ZmPME10-1* 的全长蛋白相互作用, 暗示 *ZmGa1P* 与 *ZmPME10-1* 的 PME 结构域的互作效应较 *ZmGa1S* 更为强烈, 推测可能通过竞争解除源于 *ZmGa1S* 对 *ZmPME10-1* 的互作抑制作用而导致的酶活性降低。*ZmGa1P* 和 *ZmPME10-1* 分别与拟南芥花粉特异表达的 PMEs 聚类到相同的分枝, 敲除 *ZmPME10-1* 在拟南芥中的同源基因表现出角果长度变短以及育性变差的表型, 暗示单、双子叶 PMEs 功能的保守性。



陈化榜, 中国科学院遗传与发育生物学研究所研究员。1984年毕业于山东农业大学, 1999年获美国 Purdue University 农学博士学位。2011年12月入选中国科学院“百人计划”。主要从事玉米优异种质资源创制与利用、控制重要农艺性状基因的克隆与应用、育种和制种新理论及新方法的研究, 并致力于高产、稳产、高效、优质玉米新品种的规模化选育, 相关研究成果发表在《Nature Communications》, 《Plant Physiology》, 《Plant Journal》, 《Crop Science》, 《Theoretical & Applied Science》等期刊上。曾获中国科学院优秀“百人计划”奖, 中国农业植物新品种培育“育种之星”, “High Scholarship, Outstanding Achievement & Service”奖, The Honor Society of Agriculture, Gamma Sigma Delta, USA 和国家技术发明一等奖。

Invited Talk 2

Evolutionary divergence of the plant MCIA complex that is required for mitochondrial complex I assembly and seed development

Jiacheng Ni², Yongyan Wang¹, Hongyu Chen¹, Jihua Tang¹ and Guifeng Wang^{1*}

¹ Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China;

² Shanghai Key Laboratory of Bio-Energy Crops, School of Life Sciences, Shanghai University, Shanghai 200444, China.

*Corresponding author: guifeng.wang@henau.edu.cn

(submitted by Guifeng Wang < guifeng.wang@henau.edu.cn>)

Mitochondria, the powerhouse of the cell, are present in almost all eukaryotic cells where their primary role is the generation of ATP through oxidative phosphorylation. Mitochondria are double membrane bound organelles that evolve from endosymbiotic purple non-sulphur gram-negative bacteria (α -proteobacteria) about 2 billion years ago. The respiratory chain contains five respiratory complexes that allow for the establishment and utilization of a proton gradient across the mitochondrial inner membrane. Mitochondrial Complex I (NADH:ubiquinone oxidoreductase) has 44 subunits of which 7 are encoded by mitochondrial DNA (mtDNA), which utilizes a flavin mononucleotide (FMN) and its hydrophobic domain to oxidize NADH. Complex I assembly is an intricate process that are coordinately controlled by assembly factors in a step-wise fashion. In human, the mitochondrial complex I assembly (MCIA) complex, harboring five factors, is involved in the early assembly of the P_P-b subcomplex. The four core proteins in the MCIA are NDUFAB1 (NADH:ubiquinone oxidoreductase complex assembly factor 1), ECSIT (evolutionarily conserved signalling intermediate in toll pathway), ACAD9 (acyl-coA dehydrogenase family member 9), and TMEM126B (transmembrane protein 126B), as well as possible involvement of TIMMDC1 (translocase of inner mitochondrial membrane domain containing 1). However, most of these assembly factors have lost corresponding orthologs in plant. Here, we discuss the evolutionary divergence of the plant MCIA complex and its functions in mitochondrial complex I assembly and seed development.



王桂凤，河南农业大学农学院教授。2008 年于南京林业大学获遗传学博士学位；同年去上海大学生命科学学院工作，历任讲师、副教授；2014 年至 2016 年在瑞典农业大学从事博士后研究；2017 年至河南农业大学任特聘教授。主要从事：(1)作物种子储藏物质积累的分子机制；(2)植物种子发育的遗传与表观遗传调控。以第一或通讯作者在《Developmental Cell》、《Plant Cell》、《New Phytologist》等国际著名刊物发表论文 13 篇，以合作作者在《eLIFE》、《PloS Genetics》、《Plant Physiology》等国际主流期刊上发表研究论文 5 篇。获得国家自然科学基金（联合重点、面上和青年）4 项、上海市浦江人才计划和河南省科技创新

杰出青年。申请国家发明专利 9 项，已获批 3 项。

Invited Talk 3

Systematically dissection of molecular mechanisms underlying plant height in maize

Full Author List: Lin Li*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: Lin Li, hzaulilin@mail.hzau.edu.cn

(submitted by 李林<hzaulilin@mail.hzau.edu.cn>)

Ideal plant architecture is the basis of maize high-yield production, and plant height is one of the major factors constituting plant architecture in maize. However, the understanding of molecular mechanisms underlying plant height is still limited. Our transcriptome profiling suggested that protein coding genes, long noncoding RNA genes and circRNA genes are involved in the regulation of plant architecture through complicate regulatory networks. In addition, we conducted a genome-wide scan across 10 different RIL populations, and identified 83 plant height QTLs, most of which are rare variants only detected in one RIL population. To accelerate plant height QTL fine-mapping and cloning, we devised QTG-seq, by which we fine-mapped several plant height QTLs. Furthermore, we constructed a first generation interactome of maize, including the genome-wide interactions from genome 3D, co-expression networks at transcriptomic and translatomic levels, and protein-protein interactions. By collecting all the genetic, molecular, and omic datasets, we assembled a comprehensive biological big data for the understanding of plant height in maize. Using machine learning and subsequent molecular validations, hundreds of genes were identified and a complicate network with all different types of functional elements were uncovered to be associated with plant height variation in maize. Overall, our study provides a new perspective for the molecular mechanisms underlying plant height in maize in the era of “big data”.

Funding acknowledgement: the National Key Research and Development Program of China and Huazhong Agricultural University Scientific & Technological Self-innovation Foundation.



李林，华中农业大学植物科学技术学院教授。中国农业大学本硕博连读，2010年获农学博士学位，同年去美国明尼苏达大学从事博士后及研究助理工作，于2016年7月正式回国建立实验室，主要利用生物大数据进行玉米株型建成分子机制研究。以第一或通讯作者在《Genome Biology》，《Molecular Plant》，《PLoS Genetics》，《New Phytologist》，《Plant Physiology》等国际主流期刊上发表研究论文13篇。另外，以主要完成人身份获批准发明专利一项。

Invited Talk 4

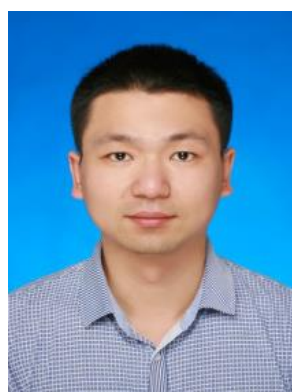
High-throughput greenhouse and field phenotyping tools to reveal the genetic architecture of phenotypic traits in maize

Wanneng Yang¹, Hui Feng¹, Chenglong Huang¹, Lingfeng Duan¹, Xiuying Liang¹, Jianxiao Liu¹, Guoxing Chen¹, Di Wu¹, Junli Ye¹, Lizhong Xiong¹, Mingqiu Dai¹, Jianbing Yan¹

¹National Key Laboratory of Crop Genetic Improvement, National Center of Plant Gene Research, Agricultural Bioinformatics Key Laboratory of Hubei Province, College of Plant Science & Technology, and College of Engineering, Huazhong Agricultural University, Wuhan 430070, PR China.

Corresponding author: Wanneng Yang (ywn@mail.hzau.edu.cn)

The functional analysis of the maize genome has entered into a high-throughput stage. However, traditional phenotyping is usually labor-intensive, time consuming, lower throughput, costly, and frequently destructive to plants, and is far behind the development of other -omic studies such as genomics. To relieve the bottleneck, multi-omics technologies are urgently needed, particularly in high-throughput phenotyping. In the recent years, we set up a high-throughput phenotyping platform from greenhouse to field level, and some interesting results were found that: (1) single leaf growth and whole plant growth of maize could be non-destructively monitored and quantified; (2) biomass accumulation and final yield were predicted using a combination of dissected traits in the early growth stage; (3) using the RGB images, hyperspectral, CT based traits (i-traits), we demonstrate that these i-traits can be used to monitor drought response and have high heritability; combined with the i-traits and genome-wide association studies (GWAS) or QTL, some new DR-related candidate gene could be detected; (4) to screen the maize plant in field level, a high-throughput field-plot phenotyping was developed combined with RGB, far-infrared, and hyperspectral imaging; and the maximum inspection speed can catch up to about 2 hours per 480 plots in field; (5) in addition, to measure the maize kernel traits and ear traits, a high-throughput yield traits scorer was developed based on linear scanning and PLC control. In future, combined with multi-scale phenotyping tools and other -omics tools on large panels of resequenced maize accessions, the strategies for the next green revolution can be accelerated and promoted.



杨万能，华中农业大学植物科学技术学院教授，作物遗传改良国家重点实验室独立 PI。2011 年毕业于华中科技大学武汉光电国家研究中心，从博士至今系统从事作物表型组学交叉学科研究，负责作物遗传改良国家重点实验室作物表型组学平台和团队建设，已成功研发一系列原创高通量表型仪器并推广。以（共同）第一作者、（共同）通讯作者在《Nature Communications》，《Molecular Plant》，《Plant Physiology》，《Journal of Experimental Botany》，《Plant Methods》，《Current Opinion in Plant Biology》等期刊发表 SCI 论文 14 篇；相关成果获得发明专利 12 项，其中第一

完成人 6 项，成果转化 1 项。

Invited Talk 5

玉米理想基因组智能设计育种

姜淑琴¹, 程前¹, 闫军¹, 王向峰^{1*}

¹ 中国农业大学, 国家玉米改良中心, 北京, 100091

*通讯作者: 王向峰, xwang@cau.edu.cn

随着人类社会步入人工智能的新纪元, 未来作物育种模式将发生历史性的变革。玉米育种科学将在组学大数据与生物信息学、基因编辑技术与合成生物学、人工智能与机器学习技术等多学科、多领域的共同支撑下, 推动玉米育种快速迈进 4.0 阶段。未来十年到二十年里, 我国玉米育种领域的核心发展目标之一是创建适合我国玉米育种产业国情的“玉米基因组智能设计育种”技术体系。所谓“基因组智能设计”的核心体现, 是应用人工智能模拟的方法, 为某一育种群体材料人工设计聚合了所有优势基因、具有“理想基因型”的虚拟基因组。再用机器学习模型预测虚拟亲本基因组与测试亲本基因组组配产生的杂交后代的“理想表型”。所谓“理想表型”则是该育种群体所能创造的杂种后代性状的理论上限, 是该育种群体的杂种优势利用的潜力极限。根据智能设计的理想基因型, 建立亲本自交系创制方案和杂交育种方案, 快速逼近杂交后代的理想表型。最终, 通过育种大数据与人工智能决策, 实现辅助育种家精准筛选玉米优良亲本组合, 快速创制玉米优良自交系, 有效地缩短育种周期、提高育种效率、降低育种成本。在未来 10 到 20 年内实现育种从“艺术”到“科学”到“智能”的革命性转变。

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王向峰, 中国农业大学农学院教授。2002 年获中国农业大学生物科学学士学位, 2007 年获北京大学生物信息学博士学位, 2007-2010 年先后在美国耶鲁大学与哈佛大学从事博士后研究工作, 2010-2014 年在美国亚利桑那大学任助理教授, 2014 年 6 月回国在中国农业大学农学院, 作物基因组与生物信息学系、国家玉米改良中心任职。课题组研究方向主要应用机器学习等大数据挖掘技术建立玉米全基因组选择辅助育种模型。

Invited Talk 6

Genetic Basis of Artificial Selection Response in High-Oil Maize

Gen Xu^{1,4}, Min Wang^{1,4}, Binghao Zhao^{1,4}, Yingjia Han^{1,4}, Hui Fang¹, Wenkang Chen¹, Weiya Li¹, Xiuyi Fu¹, Haijun Liu², Ning Yang², Huihui Li³, Dong Jin¹, Jiansheng Li¹, Jianbing Yan², Xiaohong Yang^{1*}

¹ State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center of China, MOA Key Lab of Maize Biology, China Agricultural University, Beijing 100193, China.

² National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China.

³ Institute of Crop Sciences and CIMMYT China Office, Chinese Academy of Agricultural Sciences, Beijing 100081, China.

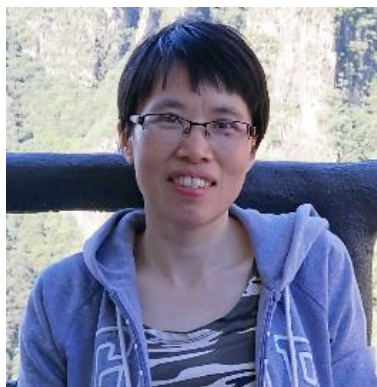
⁴ These authors contributed equally to this work.

*Corresponding author: yxiaohong@cau.edu.cn

(submitted by 杨小红 <yxiaohong@cau.edu.cn >)

High-oil maize is a product of artificial selection. To understand the genetic basis of artificial selection response in high-oil maize, we phenotyped and genotyped 400 individuals across three generations (C1, C4 and C7) of LvDaHongGu High-Oil (LDHGHO) population, which developed from nine elite inbred lines. Dynamic change was observed for oil content in maize kernels, as well as the tradeoffs for a few agronomic traits. Using 4,785,646 SNPs in 400 individuals in the LDHGHO population, we identified 55 selective sweeps with four selection patterns. Genome-wide association study identified a total of 5 to 23 loci significantly associated with each measured trait, with total explained phenotypic variation ranging from 21.65% to 88.66%. For each trait, one to eight significant association loci fell within the selected regions. 41.8% (23/55) of the selected regions were co-localized with loci associated with multiple traits, which were mainly caused by the genetic hitch-hiking effect and pleiotropy. Our results provide insights into the genetic basis of the accumulation of oil content and its selection tradeoff in a dynamic select population.

Funding acknowledgement: National Natural Foundation of China.



发表 SCI 论文 20 多篇。

杨小红, 中国农业大学, 农学院, 教授, 博士生导师。目前主要从事玉米高产优质的遗传基础和分子育种研究。2013 年入选教育部新世纪优秀人才支持计划; 2015 年入选北京市科技新星计划; 2017 年获国家优秀青年科学基金。2014 年获教育部技术发明一等奖 (第三完成人); 2016 年获国家技术发明二等奖 (第三完成人), 2017 年获神农中华农业科技奖 (第 12 完成人)。2008 年至今在 *Nature Genetics*、*PNAS*、*Nature Communications*、*Plant Cell* 等期刊发表 SCI 论文 60 多篇, 以 (共同) 第一或 (共同) 通讯作者

Invited Talk 7

Genome-wide Selection and Genetic Improvement During Modern Maize Breeding

Baobao Wang, Zechuan Lin, Xin Li, Jinsheng Lai, Matthew B. Hufford, Hang He and Haiyang Wang*

*presenter: whyang@scau.edu.cn

Modern crop breeding has made a profound impact on food production to sustain population growth during the past century; however, systematic analysis of the genomic basis underlying the dramatic increase in crop yields during modern breeding remains forthcoming. In this study, through assembling, resequencing and phenotyping of a maize diversity panel comprising 350 elite inbred lines from five discrete ‘breeding eras’ (2 in the USA and 3 in China), we identified 2046 genomic selective sweeps that represent the “breeding signatures” arising during modern maize breeding in the US and China. We additionally identified dozens of candidate genes related to stress tolerance and plant architecture for adaptation to modern agricultural management. Moreover, our results clearly demonstrate the emergent influence of simultaneous accumulation of favorable alleles in both the male and female parents during modern hybrid maize breeding. This work lays the foundation for a genomic selection platform for future maize breeding.

Keywords: Maize (*Zea mays* L.), high-density planting, selective sweeps, biotic and abiotic stress tolerance, plant architecture, genome-wide association study (GWAS)



王海洋，华南农业大学生命科学学院教授。1998年毕业于美国密执根大学获生物学专业博士学位；1998年至2002年在美国耶鲁大学分子、细胞和发育生物学系从事博士后研究。2002-2013年先后在美国康奈尔大学 Boyce Thompson 植物研究所和耶鲁大学分子、细胞和发育生物学系担任独立课题负责人和博士生导师；2013年起担任中国农业科学院生物技术研究所以研究员和玉米功能基因组创新团队首席科学家；2016年起担任华南农业大学生命科学学院二级教授。主要从事植物光信号转导和作物理想株型遗传调控网络和分子机理的研究。已发表 SCI 论文 110 余篇；一项工作被评为 2014 年度中国科学十大进展。

Invited Talk 8

自发免疫突变体-解析玉米抗病机理的有利工具

苟明月^{*1}, 李见坤¹, 徐莉萍¹, 母小焕¹, 于延文¹

¹河南农业大学农学院, 郑州, 450002

*通讯作者: mingyuegou@163.com

提交人: 苟明月 <mingyuegou@163.com>

在无病原侵染条件下即表现出叶片类似病斑表型的突变体被称为类病斑 (*disease lesion mimics, les*) 突变体。其中, 对病原物具有自发性、组成型免疫反应的突变体又被称为自发免疫突变体 (*autoimmune mutant*)。在拟南芥、水稻、小麦、玉米等模式植物和作物中存在大量自发免疫突变体, 它们是研究植物抗病防御反应及广谱抗性机制的有利工具。目前通过对拟南芥自发免疫突变体的研究, 已克隆近50个不同的抗病防御相关基因, 主要包括植物抗病 (*R*) 基因及多个抗病防御负调控因子。其中, 对 *snc1*、*cpr1/cpr30*、*bon1*、*siz1* 等突变体的研究表明, *R* 基因 *SNC1* 及其介导的拟南芥抗病防御反应受到多个基因的精细调控。据预测, 玉米中有超过200个 *les* 位点, 但目前被克隆的基因较少, 且这些位点是否参与植物抗病防御反应有待系统研究。本报告将以拟南芥自发免疫突变体的研究为参照, 讨论玉米 *les* 突变体的研究方法及其在抗病分子育种中的潜在用途。比如: 通过 *Mutator* 转座子介导的基因克隆及集群分离分析 (*BSA*) 法可快速克隆 *les* 突变基因; 通过对 *les* 突变体的多病害抗性监测及组学分析可系统解析各 *les* 位点的病理学效应。深入研究玉米 *les* 突变体的自发免疫调控机制将为玉米多抗分子育种奠定理论基础。



苟明月, 河南农业大学农学院教授, 博士生导师。2009年毕业于中国农业大学, 获理学博士学位; 2009-2012 年在美国康奈尔大学从事植物病理学博士后研究; 2012-2018 年在美国布鲁克海文国家实验室从事植物细胞壁合成机理研究。2018 年入选“河南省特聘教授”。目前从事植物抗病和细胞壁合成机理的交叉研究及作物分子育种工作。已在《Nature Plants》、《Plant Cell》、《Plant Physiology》、《Plant Journal》等期刊以第一作者身份发表论文 11 篇, 在《Nature Communications》等期刊合作发表论文 9 篇。研究成果多次被 Science Daily 等海外媒体报道, 谷歌学术论文统计被引用 772 次。曾担任《Plant Cell》、《Plant Biotechnology》、《Plant Physiology》等 10 余个植物学期刊审稿人。

Invited Talk 9

Dissection and adoption of quantitative disease resistance in maize

Qin Yang^{1*}, Peter Balint-Kurti², Mingliang Xu³

¹ College of Agronomy and State Key Laboratory of Crop Stress Biology for Arid Areas, Northwest A&F University, Yangling 712100, Shaanxi, China

² USDA-ARS Plant Sciences Research Unit, Department of Entomology and Plant Pathology, North Carolina State University, Raleigh, NC 27695, USA

³ National Maize Improvement Centre of China, China Agricultural University, Beijing 100193, China

*Corresponding author: qyang@nwfau.edu.cn

(submitted by 杨琴 (Qin Yang) < qyang@nwfau.edu.cn >)

Maize production is constantly threatened by the presence of devastating pathogens, especially fungal pathogens worldwide. Quantitative disease resistance (QDR) is the type of resistance most widely used by maize breeders. However, the molecular mechanisms underlying QDR remain poorly understood and exploited. In my talk, I will present some progress on how we've characterized key genetic components underlying quantitative resistance to different fungal pathogens in maize. Using a variety of forward and reverse genetic approaches including QTL mapping, genome-wide association studies, map-based cloning, insertional mutagenesis, and transgenic validation, we have identified and confirmed several genes associated with disease resistance to southern leaf blight, grey leaf spot, and stalk rot. These studies have provided evidence for the hypothesis that quantitative resistance loci might be a unique set of previously unidentified genes. We are beginning to investigate the molecular mechanisms underlying the quantitative resistance genes.

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杨琴，西北农林科技大学农学院教授，第十四批国家青年人才项目入选者。2005 年获得中国农业大学农学学士，2010 年获中国农业大学作物遗传育种学博士学位。先后在中国农业大学和美国北卡罗莱纳州立大学从事博士后及研究助理工作，于 2018 年 6 月正式回国组建实验室，主要开展重要玉米真菌病害的抗病机理研究。以第一或通讯作者在 Nature Genetics、PNAS、Molecular Plant、New Phytologist 等国际主流期刊上发表研究论文多篇。

Invited Talk 10

玉米减数分裂：知否，知否？

贺岩

国家玉米改良中心，中国农业大学，北京，100193

(Submitted by 贺岩<yh352@cau.edu.cn>)

减数分裂过程中的染色体重组是作物育种的遗传学基础。减数分裂分子途径起始于程序化 DNA 双链断裂（DSB）位点的产生，其形成和修复机制在不同物种间既呈现保守性，也具有一定程度的种属特异性。本团队通过在全球范围内收集玉米减数分裂突变体，并结合基因组学、细胞学等前沿手段，系统研究玉米减数分裂 DSB 产生和修复的分子机制，致力于解析 DNA 和 RNA 甲基化对减数分裂的表观遗传调控机制。我们期望通过深入研究植物减数分裂过程，调控其中的关键步骤，改变重组分布模式和增加遗传重组率，最终提高作物育种效率。

Meiosis recombination is the genetic basis of crop breeding. Meiosis is initiated from the programmed formation of DNA double strand breaks (DSBs). The molecular pathways involving DSB formation and repair exhibit both conserved and unique characters in different organisms. In the past decades, we have collected a broad range of meiotic mutants from all over the world, and we are now utilizing the cutting-edge approaches to decipher the molecular mechanisms underlying DSB formation and repair in maize. We are particularly charmed with the roles of epigenetic and epitranscriptome in regulating meiosis. We anticipate that our great efforts in studying plant meiosis will prominently facilitate crop breeding by enhancing recombination rates and/or altering recombination pattern along chromosomes.



贺岩，中国农业大学国家玉米改良中心教授。2010年毕业于美国佛罗里达大学并获得博士学位，2011-2013 在美国康奈尔大学做博士后研究。目前主要研究方向包括玉米减数分裂重组、表观遗传学及果穗脱水调控机理解析。近五年来，以通讯或第一作者在 Plant Cell, PNAS, Molecular & Cellular Proteomics, Plant Physiology, Plant Journal, Journal of Experimental Botany 等杂志发表论文 15 篇。此外，培育玉米新品种中农大 139 和中农大 189，并参加国家区试审定。

Short Talk

Short Talk 1

Maize *VKS1* is Essential for Early Endosperm Development by Regulating Mitosis and Cytokinesis

Yongcai Huang^{1, 2, 4}, Haihai Wang^{1, 4}, Xing Huang^{1, 2}, Qiong Wang¹, Jiechen Wang¹, Dong An³, Jiqin Li¹, Wenqin Wang³, Yongrui Wu^{1*}

¹National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology & Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200032, China

²University of the Chinese Academy of Sciences, Beijing 100049, China

³School of Agriculture and Biology, Shanghai Jiao Tong University, Shanghai 200240, China.

⁴These authors contributed equally to this work

*Corresponding Author: yrwu@sibs.ac.cn

(submitted by 黄永财 <ychuang@sibs.ac.cn>)

Cell number is a critical factor that determines maize kernel size. Rapid mitotic divisions in early endosperm development produce most cells comprising the starchy endosperm; however, the mechanisms underlying early endosperm development remain largely unknown. We isolated a previously undescribed maize mutant that shows a varied-kernel-size phenotype (*vks1*). *Vks1* encodes ZmKIN11, which belongs to the kinesin-14 subfamily and is predominantly expressed in early endosperm development. *VKS1* dynamically localizes to the nucleus and microtubules and plays key roles in free nuclei migration in the syncytium as well as in mitosis and cytokinesis in early mitotic divisions. Absence of *VKS1* has relatively minor effects on plants but causes deformities in spindle assembly, sister chromatid separation and phragmoplast formation in early endosperm development, thereby resulting in reduced cell proliferation. Severities of aberrant mitosis and cytokinesis within individual *vks1* endosperms differ, thereby resulting in varied kernel sizes. Our discovery highlights *VKS1* as a central regulator of mitosis in early maize endosperm development and provides a potential approach for future yield improvement.

Funding acknowledgement: National Natural Science Foundation of China (91635303, 31830063 to Y.W., and 31871625 to H.W.) and Chinese Academy of Sciences (XDPB0401 to Y. W.).

Short Talk 2

***ZmcytMdh4* encoding a cytosolic malate dehydrogenase is essential for storage reserve synthesis and seed development in maize**

Yongqiang Chen†(陈永强), Zhiyuan Fu*†(付志远), Jihua Tang*(汤继华)

National Key Laboratory of Wheat and Maize Crop Science / Collaborative Innovation Center of Henan Grain Crops / College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China. (河南农业大学国家小麦玉米作物学重点实验室/河南粮食作物协同创新中心/农学院, 郑州, 450002)

*Corresponding author: tangjihua1@163.com

(submitted by 陈永强<wuqiang646688@163.com>)

Genes involved in starch and storage protein biosynthesis have been revealed in maize (*Zea mays*). However, the underlying regulation mechanism between them is almost unknown. In this study, we isolated a cytosolic malate dehydrogenase (*cytMdh4*) gene using map-based cloning from a natural maize kernel mutant *cytmdh4*, which has significant changes in starch and protein content compared to wild type. Transgenic and allelism test demonstrated that *cytMdh4* was the gene controlling the mutant phenotype. A 3-bp in-frame deletion in this gene was identified as the causative variation which might change the conformation resulting into repressed cytMDH4 enzymatic activity. Targeted metabolomics analysis showed that reduced cytMDH4 activity influenced amino acid synthesis, especially lysine, via affecting α -ketoglutaric acid and malate/oxaloacetate accumulation levels. Transcriptional profile showed that differentially expressed genes of zein, lysine degradation (*LKR/SDH*) and starch biosynthesis (*Bt2*, *Sh2*, and *Wx*) positively or negatively regulated by maize endosperm-specific transcription factor *opaque2* (*O2*). Concurrently, the decreased oxidation state caused by malate accumulation repressed the activity of AGP (*Bt2* and *Sh2*), which may in turn restricted the starch biosynthesis. Taken together, *ZmcytMdh4*, by affecting malate accumulation and gene expression of starch, amino acids synthesis, play critical role in regulating starch and storage protein balance in maize endosperm.

Key words: Maize, cytosolic malate dehydrogenase 4, amino acid metabolism, starch synthesis, TCA cycle.

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Short Talk 3

Single gametophyte sequencing reveals that crossover events differ between sexes in maize

Cheng Luo¹, Xiang Li^{1*}, Qinghua Zhang¹, Jianbing Yan^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

* Corresponding author: lixiang1989@webmail.hzau.edu.cn (Xiang Li), yjianbing@mail.hzau.edu.cn (Jianbing Yan)

(submitted by 罗成<luochengxiansheng@webmail.hzau.edu.cn>)

Meiotic crossover (CO) plays a key role in producing gametophytes and generating genetic variation. The patterns of CO production differ inter- and intra-species, as well as between sexes. However, sex-specific patterns of CO production have not been accurately profiled independently of genetic backgrounds in maize. Here, we develop a method to isolate single female gametophyte for genomes sequencing in maize. We show that more COs are observed in male (19.3 per microspore) than in female (12.4 per embryo sac). Based on Beam-Film model, the more designated class I and II COs are identified in male than in female. In addition, CO maturation inefficiency (CMI) is detected in some genetic backgrounds, suggesting that maize may be an ideal model for dissecting CMI. This research provides insights toward understanding the molecular mechanism of CO production between sexes and may help to improve maize breeding efficiency through paternal selection.

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Short Talk 4

Evolutionary metabolomics identifies significant metabolic divergence between maize and its wild ancestor, teosinte

Guanghui Xu^{1,3}, Jingjing Cao^{1,2,3}, Xufeng Wang¹, Qiuyue Chen¹, Weiwei Jin¹, Zhen Li^{1,*} and Feng Tian^{1,*}

¹State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Key Laboratory of Biology and Genetic Improvement of Maize (MOA), Beijing Key Laboratory of Crop Genetic Improvement, Center for Crop Functional Genomics and Molecular Breeding, China Agricultural University, Beijing 100193, China

²Institute of Plant Protection, Jiangsu Academy of Agricultural Sciences, Nanjing 210014, China

³These authors contributed equally to this work

*Corresponding author: Feng Tian (ft55@cau.edu.cn) and Zhen Li (lizhenchem@cau.edu.cn)

(submitted by 许光辉<gh99@cau.edu.cn>)

Maize was domesticated from its wild ancestor, teosinte. Maize's new morphology and adaptation to diverse environments require coordinated changes in metabolic pathways that are essential for growth and development. However, how the metabolome was reshaped since domestication remains poorly understood. Here, we report a comprehensive assessment of divergence in the seedling metabolome between maize and teosinte. We demonstrate that teosinte, tropical maize and temperate maize experienced significant divergences in distinct sets of metabolites due to selection. To identify genetic factors controlling metabolic divergence, we assayed the seedling metabolome of a large maize-by-teosinte cross population. We show that the recent divergence between tropical and temperate maize was associated with more metabolite alterations and controlled by simpler genetic architecture. Using a statistical method that integrates transcriptome data, we identified candidate genes whose expression contributes to the metabolite variation in the maize-teosinte population and found that they are more likely under selection at nucleotide and transcript levels. Through overexpression or mutant analysis, we verified the roles of *FHT*, *Pr1*, and *ZmTPS1* in the divergence of their related biosynthesis pathways. Our findings not only provide important insights into domestication-associated changes in metabolism but also highlight the power of combining omics data for trait dissection.

Funding acknowledgement: the National Key Research and Development Program of China, the Recruitment Program of Global Experts and the Fundamental Research Funds for the Central Universities

Short Talk 5

High frequent DNA rearrangement at the 27-kD γ -zein locus creates a superior *o2* modifier for Quality Protein Maize breeding

Hongjun Liu^{1,5}, Yongcai Huang^{2,5}, Xiaohan Li^{1,5}, Haihai Wang², Yahui Ding¹, Congbin Kang¹, Mingfei Sun¹, Fangyuan Li¹, Jiechen Wang², Yiting Deng², Xuerong Yang¹, Xing Huang², Lingling Yuan⁴, Qiong Wang², Dong An³, Wenqin Wang³, David R. Holding⁴, Yongrui Wu^{2*}

¹ State Key Laboratory of Crop Biology, College of Life Sciences, Shandong Agricultural University, Tai'an 271018, China

² National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology and Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai China 200032

³ School of Agriculture and Biology, Shanghai Jiao Tong University, Shanghai, China 200240

⁴ Department of Agronomy and Horticulture, Center for Plant Science Innovation, Beadle Center for Biotechnology, University of Nebraska, Lincoln, NE, USA 68588-0665

⁵These authors contributed equally to this work.

*Correspondence should be addressed to Y. Wu (yrwu@sibs.ac.cn).

The duplication at the 27-kD γ -zein locus is a major *o2* modifier (*q γ 27*) for endosperm modification in Quality Protein Maize (QPM). *q γ 27* is unstable and frequently produces single copies. Due to the lack of effective phenotypic or molecular markers, it was previously not possible to determine the frequency of germinal DNA rearrangement at this locus. We designed a polymorphic PCR marker that could discriminate the duplicated copies and generated a mutant QPM line (K0326Y-*Del*) which entirely lacks the *q γ 27* locus. When different maize lines were crossed to the null K0326Y-*Del* line, the frequency of DNA rearrangement could be determined because the PCR products arose entirely from the parents contributing the *q γ 27* allele. The frequency with which *q γ 27* rearranges to single copies from one generation to another is on the order of 10^{-3} and varies dramatically among different lines, with the highest in A188 and lowest in Mo17. It occurs significantly higher in male than female gametogenesis in all lines. Due to a relatively higher frequency in W22, the triplication of 27-kD γ -zein gene was identified in a small number of different UniformMu stocks (W22 background). The greatly enhanced amount of 27-kD γ -zein protein by the triplication allele is sufficient to confer vitreous kernels when α -zeins are suppressed by RNAi. Our results highlight a novel approach to directly determine the frequency of DNA rearrangements, in this case resulting in copy number variation at the 27-kD γ -zein locus. Furthermore, this provides a highly effective way to test suitable parents in QPM breeding.

Keywords: Quality Protein Maize (QPM); *o2* modifier; Copy number variation; DNA rearrangement

Short Talk 6

A *helitron*-induced *RabGDIα* variant causes quantitative recessive resistance to maize rough dwarf disease

Suining Deng^{1,a}, Qingcai Liu^{1,a}, Baoshen Liu^{2,a}, Yongfu Tao^{1,a}, Haiyue Ai¹, Jianju Liu¹, Yongzhong Zhang², Yan Zhao², Mingliang Xu^{1,*}

¹ State Key Laboratory of Plant Physiology and Biochemistry/College of Agronomy and Biotechnology/National Maize Improvement Center/Center for Crop Functional Genomics and Molecular Breeding, China Agricultural University, 2 West Yuanmingyuan Road, Beijing 100193, P. R. China

² College of Agronomy/State Key Laboratory of Crop Biology, Shandong Agricultural University, Taian 271018, P. R. China

^a These authors contribute equally to this work

* Corresponding author: mxu@cau.edu.cn

(submitted by 邓穗宁<13391860723@163.com>)

Maize rough dwarf disease (MRDD), caused by various *Fijiviruses* in the family *Reoviridae*, poses a grave threat to maize production worldwide. We previously identified a major quantitative trait locus on chromosome 8, *qMrdd1*, that confers recessive resistance to one causal pathogen of *Rice black-streaked dwarf virus* (RBSDV). Through map-based cloning strategy, we demonstrate that Rab GDP dissociation inhibitor alpha (*RabGDIα*) is the host susceptibility factor for RBSDV. A *helitron* transposon insert into intron 10 of *RabGDIα* creates alternative splice sites and replaces the wild exon 10 of *RabGDIα* with a *helitron*-derived exon 10, resulting in recessive resistance to RBSDV. In the process of RBSDV infection, the *helitron*-induced *RabGDIα* splicing variant showed almost the same gene expression profiles as *RabGDIα* regardless of developmental stages and resistance performance. We also identified the viral P7-1 protein as the pathogenicity determinant that targets the wild-type *RabGDIα* by binding to the exon-10-encoded peptide and C-terminal region to initiate viral infection. However, P7-1 has difficulty recruiting the *helitron*-induced *RabGDIα* variant, this may impair viral intercellular movement and eventually leads to quantitative recessive resistance to RBSDV. Additionally, we confirm that all naturally recessive resistance genes probably arose from a single *helitron* insertion event. This resistance allele can be useful to improve the resistance of maize varieties to MRDD by marker-assisted selection, and potentially to increase the resistance of other crops to RBSDV.

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Short Talk 7

Variations in an LRR-Receptor Like Kinase Gene, *SRR1*, Impart Different Quantitative Resistance to Seed and Ear Rot in Maize Heterotic Groups

Zijian Zhou¹, Kewei He¹, Peipei Ma¹, Jianxin Wu², Wei Wang², Jingyang Gao¹, Ming Ju¹, Cong Mu¹, Yabin Wu¹, Chaopei Dong¹, Yunxia Song¹, Zongliang Xia², Jiafa Chen², Huiyong Zhang^{2,*}, Jianyu Wu^{1,2,*}

¹National Key Laboratory of Wheat and Maize Crop Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou, 450002, China

² College of Life Sciences, Synergetic Innovation Center of Henan Grain Crops and National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding Author: wujianyu40@126.com

(submitted by 周子键<zhouzijian19900601@163.com>)

Fusarium verticillioides is a widely-distributed soil-borne fungal species, which can be transmitted to seeds and causes seed rot, stalk rot, and ear rot by systemic infection in maize. The most significant single nucleotide polymorphism associated with seed rot in the *qFR1* region, which is a quantitative resistance locus in maize, has been identified in the *GRMZM2G009818* gene. Here, we demonstrated that *seed rot resistance 1* (*SRR1*, *GRMZM2G009818*) encodes a leucine-rich repeat-receptor like kinase that functions as a pattern recognition receptor and imparts quantitative resistance to seed rot caused by *F. verticillioides*. We demonstrated that *SRR1* is localized in the cytomembrane and can activate PAMP-triggered immunity, ETI, and salicylic acid signal after *F. verticillioides* inoculation. Three key functional variations in the coding region are responsible for five functional haplotypes of *SRR1*, which impart different resistance to seed and ear rot. An undesirable functional haplotype of *SRR1*, appears to have occurred after domestication of maize resulting in the loss of ear rot resistance; this haplotype accumulated in the TSPT heterotic group, which includes most of the male parents in Huang-Huai-Hai, the main maize producing area of China. Our results would help in better understanding of *SRR1*, an important quantitative resistance gene.

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Short Talk 8

Molecular dissection of maize seedling salt tolerance using a genome-wide association analysis method

Meijie Luo¹, Yunxia Zhang¹, Panpan Zhang¹, Kuan Chen¹, Wei Song¹, Xiaqing Wang¹, Baishan Lu¹, Yanxin Zhao^{1*}, Jiuran Zhao^{1*}

¹Beijing Key Laboratory of Maize DNA Fingerprinting and Molecular Breeding, Maize Research Center, Beijing Academy of Agriculture and Forestry Sciences (BAAFS), Beijing 100097, China

*Corresponding author: rentlang2003@163.com; maizezhao@126.com

(submitted by 骆美洁<mjluo108@163.com>)

Soil salinity is a major devastating abiotic factor that affects maize growth and productivity worldwide. However, knowledge of the molecular mechanisms of responses to salt stress in maize remains limited. To elucidate the genetic basis of salt tolerance traits, a genome-wide association study was performed on 348 maize inbred lines under normal and salt-stressed conditions. The phenotypic data for 27 traits revealed coefficients of variation > 25%. In total, 149 significant SNPs were identified, explaining 6.6%–11.2% of the phenotypic variation for each SNP. Of the 104 identified quantitative trait loci (QTLs), 83 were related to salt tolerance and 21 to normal traits. Additionally, 13 QTL were simultaneously identified by two to five traits. Ten and six QTLs controlling salt tolerance traits and root growth were co-localized with reported QTL intervals from linkage populations, respectively. Based on functional annotations, 16 candidate genes for salt tolerance were predicted. Two genes were identified as known maize salt response genes, one of which, *plasma membrane protein 3* (*PMP3*, *GRMZM2G477325*) was characterized to involve in ion homeostasis. One of the candidate genes, *GRMZM2G071119*, were located in a QTL harboring 11 peak SNPs and its *Arabidopsis* homolog is responsible for chloride transmembrane transport, making it a promising target for further functional investigations. These results aid in elucidating the genetic variation in salt tolerance and provide novel loci for the genetic improvement of maize with salt tolerance.

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Short Talk 9

A new role for ethylene as a developmental signal controlling ear length and kernel number in maize

Qiang Ning^{1, §}, Yinan Jian^{1, §}, Yanfang Du¹, Ran Zhao¹, Xiaomeng Shen¹, Haitao Jia¹, Lei Liu^{1,2}, Fang Yang¹, David Jackson^{1,2,*}, Zuxin Zhang^{1,*}

1 National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, PR China

2 Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, NY 11724, USA.

*Corresponding author: zuxinzhang@mail.hzau.edu.cn; jacksond@cshl.edu

(submitted by 宁强 ningqiang_404@163.com)

Maize (*Zea mays* L.), one of important cereal crop plants, produces two spatially separated inflorescences, the ear and the tassel. Axillary meristems produced on the ear inflorescence are initially indeterminate spikelet-pair meristems that form determinate spikelets which terminate with production of two pistillata florets through the arrest of the stamens. Therefore, meristematic activity of inflorescence meristems determines the number of florets on the ear, and fate of floral organs determines sex of floret. Here, we provide a direct evidence on ethylene level in the regulation of the meristem activity and then kernel number. We characterized that a gene encoding an ethylene biosynthesis enzyme the 1-aminocyclopropane-1-carboxylate oxidase2 (ACO2), is responsible for QTL *qEL7* for ear length and kernel number per ear by map-based cloning, gene expression, enzyme kinetic assay and transgenic validation. A 7_bp insertion/deletion closely nearby a FASCIATED EAR4 binding TGACG motif in *ZmACO2* promoter alters *ZmACO2* expression level and endogenous ethylene level. Silencing *ZmACO2* lines result in 14.6% to 24.3% of increase for ear length and 12.0% to 27.3% of increase for kernel number per row. The high ethylene level induces expression of BARREN INFLORESCENCE4 (BIF4) and AP2/EREBP transcription factors including INDETERMINATE SPIKELET1 and BRANCHED SHOOTLESS1 in the ear inflorescence. We propose a regulatory pathway of ethylene for activity maintenance of inflorescence meristem and provide a potential tool for improving grain yield by optioning endogenous ethylene level in maize.

Short Talk 10

Perturbation of the C terminus of *ZmSDW3* alters plant architecture in maize (*Zea mays* L.)

Hongchao Li¹, Lijing Wang², Meishan Liu¹, Zhaobin Dong¹, Qifang Li², Shulang Fei¹, Baoshen Liu^{2,*}, Weiwei Jin^{1,*}

¹National Maize Improvement Center, College of Agronomy and Biotechnology, China Agricultural University, Beijing 10093, China

²State Key Laboratory of Crop Biology, College of Agronomy, Shandong Agricultural University, Taian 271018, China

*Corresponding authors: weiweijin@cau.edu.cn; liubs@sdaa.edu.cn

(Submitted by 李洪超 <hongchaoli2008@163.com>)

Plant height and leaf angle are two crucial determinants of plant architecture in maize and are closely related to lodging resistance and canopy photosynthesis at high planting density. These two traits are primarily regulated by phytohormones, including gibberellins, brassinosteroids, and auxin. However, the role of ethylene in regulating plant architecture in maize, especially plant height and leaf angle, is unclear. Here, we characterized a semidominant maize mutant, *Semi-dwarf3* (*Sdw3*), which exhibits shorter stature and larger leaf angle than the wild type. Scanning electron microscopy observation showed that inhibition of longitudinal cell elongation in the internode and promotion in the auricle were mainly responsible for reduced plant height and enlarged leaf angle in *Sdw3*. Through map-based cloning, we identified a transposable element insertion in the candidate gene *ZmSDW3*, encoding an enzyme in ethylene biosynthesis. The transposon alters the C terminus of *ZmSDW3*. Transgenic analysis confirmed that the mutant *ZmSDW3* gene confers the phenotypes of *Sdw3*. Enzyme activity and protein degradation assays indicated that the altered C terminus of *ZmSDW3* increases this protein's stability but does not affect its catalytic activity. The ethylene content is significantly elevated in *Sdw3*, leading to reduced plant height and increased leaf angle. In addition, we demonstrated that *ZmSDW3* plays crucial roles in root development, flowering time, and leaf number, indicating that *ZmSDW3* is an important gene with pleiotropic effects during maize growth and development.

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Short Talk 11

The RALF-LRX-ANX/BUPs signaling pathway is conserved during pollen tube growth in maize

Lele Wang, Liangzi Zhou, Thomas Dresselhaus*

Cell Biology and Plant Biochemistry, University of Regensburg, Germany

*Corresponding author: Thomas.Dresselhaus@ur.de

(Submitted by 王乐乐 <lele.wang@ur.de>)

Small secreted peptides can be classified into two major groups, CRPs (cysteine-rich peptides) and non-CRPs [1]. Previous studies have shown that members of various CRP sub-classes are involved in different steps of the double fertilization process of flowering plants [2]. To investigate the roles of CRPs during maize reproduction, we performed RNA-seq analysis to identify CRPs with specific expression pattern during pollen tube growth and fertilization in maize. We identified four genes encoding rapid alkalization factor (RALF) CRPs, which are highly and exclusively expressed in germinated pollen tubes. To understand the function of these pollen-specific *RALFs* during reproduction, *RALF*-RNAi lines were generated. During *in vitro* germination tests, pollen tubes from down-regulated lines were less stable and burst much faster compared with wild type pollen tubes. Comparing phospho-proteomic data of pollen tubes from wild type and RNAi lines, we found that phosphorylation of PEX2, a maize homologue of Arabidopsis LRX8-11 is strongly induced in RNAi lines. Pull-down experiments also confirmed interaction of maize RALF2/3 with PEX2. In *Arabidopsis thaliana* it was further shown that pollen expressed RALF4/19 can interact with LRX and CrRLK1L receptor like kinases ANX1/2 and BUPs1/2 being involved in pollen tube integrity [3, 4]. Based on sequence alignment and expression pattern comparisons, several CrRLK1L receptor like kinases were identified in maize silks and pollen being capable to interact with maize RALF2/3. Moreover, maize and Arabidopsis RALFs and corresponding CrRLK1L receptor like kinases are interchangeable. Maize RALF2/3, for example, interact *in vitro* with Arabidopsis ANXUR1/2 and BUPs1/2 as well as LRX9/10/11 and *vice versa* indicating that the RALF-LRX-ANX/BUPs signaling pathway is conserved. Knocked-out CRISPR-Cas9 mutants of above pollen-specific *RALFs* and CrRLK1L receptor-like kinases have been generated and are currently being investigated.

Short Talk 12

Genome Editing and Double Fluorescence Proteins Enable Robust Maternal Haploid Induction and Identification in Maize

Le Dong¹, Lina Li¹, Changlin Liu¹, Chenxu Liu², Shuaifeng Geng¹, Xinhai Li¹, Changling Huang¹, Long Mao¹, Shaojiang Chen², Chuanxiao Xie^{1,*}

¹Institute of Crop Science, Chinese Academy of Agricultural Sciences, National Key Facility for Crop Gene Resources and Genetic Improvement, Beijing, 100081 China

²National Maize Improvement Center of China, China Agricultural University, No. 2 Yuan Ming Yuan West Road, Haidian District, Beijing 100193, China

(submitted by 谢传晓 <xiechuanxiao@caas.cn>)

Genome editing technologies has paved the way for exciting and novel applications in plant biotechnology. Doubled haploid (DH) technology has significant and valuable advantage over traditional approaches in crop breeding. It is pivotal to establish maternal haploid induction (HI) coupled with a stable and efficient haploid identification (HID) system that can be utilized to create maternal haploid inducer at any genetic background in maize and can also be extended to other important cereal crops. Here we showed that the targeted creation of HI lines by targeting its' *ZmMTL/ZmPLA1* gene using CRISPR/Cas9 and then they were then crossed to stacked with a HID tool that carried double-fluorescence-protein (DFP) markers to identify maternal parthenogenesis haploid seeds. Simultaneously, the DFP cassette was stably transformed into the maize variety ZC01 after it had been transiently verified the tissues-preferred expression patterns. The DFP-mediated haploid inducer lines (DHILs), which the DFP cassette transformant were successfully stacked with previous targeting *MTL* mutations, were developed to validate the concept of both maternal induction and robust haploid identification. We found that their HI rate ranged from 4.7% to 11.0% with an average of 7.47%. Furthermore, the DFP cassette was found to be transiently expressed in maturing grains of bread wheat (*Triticum aestivum* L.), rice (*Oryza sativa* L.) and barley (*Hordeum vulgare* L.) in the similar tissue-preferred patterns with maize. The results indicate that this developed HID should also work as essential components of DH for those important cereal crops. In conclusion, we have developed an approach to create haploid inducer with robust HID marker for DH breeding system in maize and, possibly, for other important cereals as well. The CRISPR/Cas9 mediated *ZmMTL* (*ZmPLA1*) target mutation enabled to create maternal haploid inducer. The embryo- and endosperm- specific DFP markers were useful as an effective selection marker for maternal haploid identification for both mature seeds and young embryos. In addition, the system should be robust in open pollination environment. It also offers the potential for other important cereals like wheat, rice and barley which has no efficient HID selection method. With its broad application, this system should significantly contribute to DH breeding and hence yield increase in major cereal crops.

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Short Talk 13

Development of a Haploid-Inducer Mediated Genome Editing (IMGE) System for Accelerating Maize Breeding

Baobao Wang^{1,3,*}, Lei Zhu^{1,3}, Binbin Zhao¹, Yongping Zhao¹, Yurong Xie¹, Zhigang Zheng², Yaoyao Li², Juan Sun² and Haiyang Wang^{2,*}

¹Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing 100081, China

²School of Life Sciences, State Key Laboratory for Conservation and Utilization of Subtropical Agro-Bioresources, South China Agricultural University, Guangzhou 510642, China

³These authors contributed equally

*To whom correspondence should be addressed: wangbaobao@caas.cn or whyang@scau.edu.cn

(submitted by 王宝宝 < wangbaobao@caas.cn >)

Crop breeding aims to generate pure inbred lines with multiple desired traits. Doubled haploid (DH) and genome editing using CRISPR/Cas9 are two powerful game-changing technologies in crop breeding. However, both of them still fall short in rapid generation of pure elite inbred lines with integrated favorable traits. We report here the development of a Haploid-Inducer Mediated Genome Editing approach (IMGE for short), which utilizes a maize haploid inducer (HI) line carrying a CRISPR/Cas9 cassette targeting for a desired agronomic trait to pollinate an elite maize inbred line, and to generate genome edited haploids in the elite maize background. Homozygous pure DH lines with the desired trait improvement could be generated within two generations, thus bypassing the lengthy procedure of repeated crossing and backcrossing used in conventional breeding for integrating a desirable trait into elite commercial backgrounds. We envisage that this technology could be widely used to accelerate crop breeding.

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Abstract

Abstract 1

Maize genotypes with contrasting root system architecture responded differently to drought stress

Tingting An¹, Sheng Qiao¹, Yan Fang^{1,2}, Yinglong Chen^{1,2,3*}

¹State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, Yangling 712100, China

²Institute of Soil and Water Conservation, Northwest A&F University, & Chinese Academy of Sciences, Yangling 712100, China

³The UWA Institute of Agriculture, & School of Agriculture and Environment, The University of Western Australia, Perth WA 6009, Australia

*Corresponding author: yinglongchen@hotmail.com

(submitted by 安婷婷<tingtingan2014@163.com>)

Abstract: Globally, the productivity of maize crops is seriously influenced by drought stress. Understanding of how to effectively utilize limited water resources by crops is critical in improving maize yield. This study used 18 maize genotypes with contrasting root system architecture selected from a phenotyping experiment for root trait characterization in 174 genotypes of maize from northern China and Serbia. Plants grew in a novel semi-hydroponic platform for 20 days after transplanting, and then treated with or without PEG-6000 in two doses: 10% (0.2 MPa) and 15% (0.4 MPa) for 10 days to simulate drought stress. Results showed that: (1) PEG induced drought stress decreased leaf number and plant height, but increased root-shoot ratio across the 18 genotypes; (2) the induced drought stress significantly reduced the number and diameter of lateral roots, and increased fine roots in the upper layer; (3) tissue peroxidase concentration and soluble sugar content were higher in the PEG treated plants than the control; Genotypes with contrasting root properties responded differently to drought stress. Some root morphological traits (such as root length, root biomass and node root angle) and root physiological traits, which are closely related to the roots absorptive capacity, can be used for identifying drought tolerance of maize genotypes. To conclude, this study identified large variation in root architecture traits and physiological traits, among the tested genotypes. It's necessary to search for genotypes with optimal root traits in maize breeding program aiming for improved adaptation to drought stress.

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Abstract 2

***ZmMs30* Encoding a Novel GDLS Lipase Is Essential for Male Fertility and Valuable for Hybrid Breeding in Maize**

Xueli An^{#1,2}, Zhenying Dong^{#1,2}, Youhui Tian^{#1,2}, Ke Xie^{#1,2}, Suowei Wu^{1,2}, Taotao Zhu¹, Danfeng Zhang², Yan Zhou², Canfang Niu^{1,2}, Biao Ma^{1,2}, Quancan Hou^{1,2}, Jianxi Bao^{1,2}, Simiao Zhang¹, Ziwen Li^{1,2}, Yanbo Wang¹, Tingwei Yan¹, Xiaojing Sun¹, Yuwen Zhang^{1,2}, Jinping Li², Xiangyuan Wan^{1,2*}

¹ Biology and Agriculture Research Center, University of Science and Technology Beijing, Beijing, 100024, China; ² Beijing Engineering Laboratory of Main Crop Bio-Tech Breeding, Beijing International Science and Technology Cooperation Base of Bio-Tech Breeding, Beijing Solidwill Sci-Tech Co. Ltd., Beijing 100192, China

Co-first authors; *Corresponding author: wanxiangyuan@ustb.edu.cn

(submitted by 安学丽 <xuelian@ustb.edu.cn >)

Abstract: Genic male sterility (GMS) is very useful for hybrid vigor utilization and hybrid seed production. Although a large number of GMS genes have been identified in plants, little is known about the roles of GDLS lipase members in anther and pollen development. Here, we report a maize GMS gene, *ZmMs30*, which encodes a novel type of GDLS lipase with diverged catalytic residues. Enzyme kinetics and activity assays show that *ZmMs30* has lipase activity and prefers to substrates with a short carbon chain. *ZmMs30* is specifically expressed in maize anthers during stages 7–9. Loss of *ZmMs30* function resulted in defective anther cuticle, irregular foot layer of pollen exine, and complete male sterility. Cytological and lipidomics analyses demonstrate that *ZmMs30* is crucial for the aliphatic metabolic pathway required for pollen exine formation and anther cuticle development. Furthermore, we found that male sterility caused by loss of *ZmMs30* function was stable in various inbred lines with different genetic background, and that it didn't show any negative effect on maize heterosis and production, suggesting that *ZmMs30* is valuable for crossbreeding and hybrid seed production. We then developed a new multi-control sterility system using *ZmMs30* and its mutant line, and demonstrated it is feasible for generating desirable GMS lines and valuable for hybrid maize seed production. Taken together, our study sheds new light on the mechanisms of anther and pollen development, and provides a valuable male-sterility system for hybrid breeding maize.

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Abstract 3

MicroRNAs Are Involved in Maize Immunity against *F. verticillioides*-associated Ear Rot

Zijian Zhou^{1#}, Yan Cao^{1#}, Tao Li¹, Xinghao Wang¹, Hang He², Wen Yao¹, Jianyu Wu^{1*}, Huiyong Zhang^{1*}

¹State Key Laboratory of Wheat and Maize Crop Science, Collaborative Innovation Center of Henan Grain Crops, College of Life Science, Henan Agricultural University, Zhengzhou 450002, China

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

[#]These authors contributed equally to this article.

*Correspondence: Huiyong Zhang (huiyong.zhang@henau.edu.cn) or Jianyu Wu

(wujianyu40@126.com)

(submitted by 曹岩 <caoyanyx163@163.com>)

Abstract: *Fusarium* ear rot (FER) is a common fungal disease caused by *Fusarium verticillioides* (*F. verticillioides*) in maize, which results in severe yield losses and grain contamination with health threatening mycotoxins. Although many studies have focused on the transcriptional regulation of defense responses against *F. verticillioides* infection in maize, less is known about the involvement of microRNAs (miRNAs) in this process. By deep sequencing small RNA libraries from maize kernels of susceptible (N6) or resistant (BT-1) inbred lines in normal condition and upon *F. verticillioides* infection, pathogen treatment was accompanied by dynamic alterations in the expression of a significant number of miRNAs, including new members of annotated miRNAs. The expression of some target genes was negatively correlated with the expression of miRNAs through integrated analysis of transcriptome, degradome and bioinformatics data. Functional categories revealed that pathogen-responsive miRNAs and their targets were preferentially enriched in phenylpropanoid metabolic process, plant-pathogen interaction, and plant phytohormone signal transduction pathways, suggesting that they might play different roles in maize defense against fungal pathogen infection. Moreover, transgenic maize plants overexpressing miR408b confers decreased resistance against *F. verticillioides* infection in the susceptible maize line. Taken together, our data provide new insight into the regulatory role of miRNA in maize immunity against FER and fungal disease-resistant breeding.

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Abstract 4

Effects of temperature, water content, pH and soil sterilization on the degradation of CP4-EPSPS protein released from herbicide-tolerant corn leaves in the soil

Xing Zeng^{#1}, Kang Wang^{#1}, Chenchen Feng¹, Xinyuan Song², Fengkun Qi¹, Lin Zhang¹, Ling Dong¹, Yu Zhou¹, Xianjun Liu¹, Zhenhua Wang^{1*}, Hong Di^{1*}

¹ College of Agronomy, Northeast Agricultural University, No. 600 ChangJiang Road, Xiang Fang District, Harbin 150030, Heilongjiang Province, China

²Jilin Academy of Agricultural Sciences, No. 1363 ShengTai Street, Changchun, Jilin 130124, China

*Corresponding author: zhenhuawang_2006@163.com, dihongdh@163.com

(submitted by 曾兴<zengxing980@hotmail.com>)

Abstract: Determining the influence of soil environmental factors on the degradation of CP4-EPSPS protein from herbicide-tolerant (HT) corn residue is crucial for assessing the ecological risks of commercialized transgenic crops. We investigated the effects of soil temperature (15 °C, 25 °C, 35 °C), soil water content (20%, 33%, 50%), pH (5.0, 7.0, 9.0) and soil treatment (not sterilized, sterilized) on the degradation of CP4-EPSPS protein released from the leaves of the HT corn variety CC-2. An exponential model (first order) was fitted to the degradation dynamics of CP4-EPSPS protein and used to estimate the time required for 50% degradation (DT50) and for 90% degradation (DT90). The results showed that CP4-EPSPS protein released from CC-2 leaves presented similar degradation features in all treatments: a sharp decrease in the early stage followed by a slow decline in the middle and late stages was observed. In the late stage (more than 120 d after the experiment started), CP4-EPSPS protein released from CC-2 leaves was not detected in the soil using enzyme-linked immunosorbent assay (ELISA) tests. The DT50 values of CP4-EPSPS protein released from CC-2 leaves ranged from 0.92 d to 11.32 d, and the DT90 values ranged from 3.11 d to 36.59 d. The results suggest that soil temperature and soil sterilization significantly affected the degradation of CP4-EPSPS protein. Faster degradation rates were observed at higher temperature under non-sterilized conditions, but soil water content and pH did not clearly affect the degradation of CP4-EPSPS protein. These findings suggest that under appropriate soil temperature conditions, CP4-EPSPS protein from HT corn residue will not persist or accumulate in the soil.

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Abstract 5

Inheritance of resistance to *Rhopalosiphum maidis* in maize

Han Chunyan¹, Li Xiaopeng², Chen Jiafa^{1*}, Wu Jianyu^{2*}

¹College of Life Sciences, Henan Agricultural University, Zhengzhou 450002, China.

²College of Agronomy, Henan Agricultural University, Zhengzhou, 450002, China.

*Corresponding author: chenjiafa2005@163.com; wujianyu40@126.com

(Submitted by <chenjiafa2005@163.com>)

Abstract: Maize leaf aphid (*Rhopalosiphum maidis* Fitch) is one of the most serious pests of maize worldwide, they not only threaten the growth and development of maize seriously, but also transmit many viruses, such as maize dwarf mosaic virus. Isolation of the resistance gene play an important role in understanding the resistance molecular mechanism and utilization in maize breeding programs. Through years of continuous observation and study, the best identification period is 10 days after insemination. Three QTLs on the chromosomes 4, 5, 7 which named *qRrm1*、*qRrm2* and *qRrm3* were identified by 250 RILs population derived from the resistant line and the susceptible line in three environments. At the same time, 22 stable SNPs related to the resistance to aphid were identified by GWAS in three environments, and two candidate genes were determined based on the nucleotide and protein sequences in two parental lines by linkage mapping and GWAS, two of them were verified by NILs (near-isogenic lines) population and their candidate genes were also predicated. The implementation of the task can help to reveal the resistance mechanism and provide the theoretical basis for resistance breeding.

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Abstract 6

玉米中 *STOMAGEN-Like* 基因的功能分析

陈青云, 覃丽芳, 周佳, 田贞, 樊宪伟*

亚热带农业生物资源保护与利用国家重点实验室; 生命科学与技术学院, 广西大学, 南宁, 530004

*通讯作者: fanxw@gxu.edu.cn

摘要: 气孔是植物与外界环境进行气体交换的主要通道, 气孔密度影响植物的光合作用效率和蒸腾速率。因此, 对气孔发育的研究一直是植物发育生物学和作物育种研究领域的热点和前沿。鉴于禾本科植物叶片中气孔构成和分布的特殊性, 对气孔发育的研究相对于模式植物拟南芥中气孔的研究较滞后。本研究在玉米中克隆了 *STOMAGEN* 基因同源基因--*ZmSTOMAGEN1* 和 *ZmSTOMAGEN2*, 并将 *ZmSTOMAGEN1* 进行异源表达, 发现转基因拟南芥 T1 的叶片气孔密度和指数分别是野生型的 4.01 和 1.75 倍。气孔分布模式也打破了“至少一个细胞间隔”规则, 在拟南芥中出现了成簇分布的气孔。为了明确 *ZmSTOMAGEN-like* 基因在玉米气孔发育过程中的功能, 构建了用于编辑 *ZmSTOMAGEN1* 及 *ZmSTOMAGEN2* 的 CRISPR/Cas9 系统, 通过农杆菌介导玉米幼胚遗传转化获得了 *ZmSTOMAGEN-like* 基因突变体 T1 代植株, 对叶片进行镜检分析, 发现与野生型相比玉米突变体叶片下表皮的气孔密度比野生型降低了 63.3%, 气孔指数也降低了 57.2%。通过对玉米 T1 代突变体叶片的光合特性分析, 发现突变体中的叶片气孔导度、光合作用效率、蒸腾速率与野生型相比分别下降了 45.2%、30.3%和 41.9%。并且突变体的最大净光合速率、暗呼吸速率、光补偿点、光饱和点和表观量子效率均低于野生型。这些结果表明, 突变体中 *ZmSTOMAGEN-like* 基因缺失引起玉米气孔发育的缺陷, 也显著影响了其光合作用的特性。

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Abstract 7

8 个玉米泛素结合酶家族基因对低氮胁迫的响应模式分析

陈曙^{1,2}, 赵秋芳^{1,2}, 陈宏良^{1,2}, 贾利强^{1,2*}

¹中国热带农业科学院亚热带作物研究所, 湛江, 524091

²农业部热带果树生物学重点实验室, 湛江, 524091

*通讯作者: 709778271@qq.com

摘要提交人: 陈曙<chenshu4321@163.com>

摘要: 泛素结合酶是泛素/蛋白酶体途径的重要组成部分, 在蛋白质的泛素化过程中具有重要作用。本文利用生物信息学对泛素 E2 家族成员进行系统进化及基因结构分析。系统进化分析显示, 泛素结合酶基因可分为 6 个亚家族, 各亚家族成员数目差异较大, 基因数目最多的亚家族是 UBC1, 为 22 个, 成员数最少的是 UBC3, 仅为 5 个。以亚家族 UBC2 为研究对象, 发现 UBC2 中一共存在 4 个旁系同源基因对, 分别是 ZmUBC3/ZmUBC70、ZmUBC8/ZmUBC34、ZmUBC31/ZmUBC53 以及 ZmUBC45/ZmUBC66。对 UBC2 中成员进行基因结构分析发现, 互为旁系同源的基因其基因结构相似, ZmUBC45/ZmUBC66 含有外显子数量最多, 为 9 个, ZmUBC31/ZmUBC53 外显子含量最小, 仅为 4 个。基因基序分析结果显示, ZmUBC3/ZmUBC70 含有基序数量最多, 为 8 个, ZmUBC8/ZmUBC34 与 ZmUBC31/ZmUBC53 基序含量最小, 均为 3 个。基因启动子作用元件分析结果显示, 泛素 E2 基因启动子元件类型可分为光响应元件、激素响应元件、胁迫响应元件、组织表达相关元件以及周期性调控相关元件 5 大类, 其中以前三种类型元件的数量最多。基因不同组织表达分析显示, 被检测基因在玉米雌花中均有很高的表达量, 在根和茎中表达量最低。低氮胁迫实验结果显示, 所有被检测基因在低浓度 NO_3^- (KNO_3) 处理下, 其基因相对表达量在 1h 降至最低, 随后缓步上升, 在 24h 时达到最大值, 但仍小于 0h 初始表达量; 在低浓度 NH_4^+ (NH_4Cl) 处理条件下, 各基因表达量逐渐降低, 在 24h 达到最低值。这些结果表明, 泛素结合酶基因可能参与了玉米应答低氮胁迫反应途径。

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Abstract 8

Translatome Profiling Shows Widespread Complete Dominance associated with Heterosis in Maize

Sijia Chen, Wanchao Zhu, Jia Qian, Lin Li*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: hzaulilin@mail.hzau.edu.cn

(submitted by Sijia Chen <1062921795@qq.com>)

Abstract: Heterosis refers to the phenomenon in which hybrid progeny shows better performance relative to their parents, and is widely utilized in maize production. However, the translatomic variation associated with heterosis is largely unknown. Here, the translatomeic landscapes of B73, Mo17 and their F1s were profiled for V4-stage leaves using the ribo-seq. A total of 15,485 genes were simultaneously detected to be translated in hybrid and its parental lines while 4,828 genes differentially expressed between the three genotypes at the translatomic level. We found that a translating ribosome protects an ~28-nucleotide in average region and moves in three-nucleotide periodicity, which were in accordance to that in yeast. Correspondingly, we got the transcriptomic data of V4-stage leaves and performed a comparison with translatomic data. Multiple modes of genes action were discovered across all three genotypes at translational and transcriptional level, respectively. In translatome, the complete dominance played a dominance accounted for 70.6% of the differentially expressed genes while comparable levels of the complete dominance (50.3%) and over-dominant (41.2%) were observed in transcriptome. In the process of genetic transmission from transcriptome to translatome, there were a substantial number of genes with effects switching from partial dominance to complete dominance (103), and from over-dominance to complete dominance (362). All these results suggest that widespread complete dominant effects may be associated with heterosis at the translatomic level in maize. Our comprehensive study provides a new insight into the molecular mechanisms associated with heterosis in maize.

Abstract 9

Phenotyping root system architecture in maize

Yinglong Chen^{1,2*}, Sheng Qiao¹, Zed Rengel², Kadambot Siddique²

¹State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, and Chinese Academy of Sciences, Yangling 712100, China

²The UWA Institute of Agriculture, & School of Agriculture and Environment, The University of Western Australia, Perth 6001, Australia

*Corresponding author: yinglong.chen@uwa.edu.au

(submitted by 陈应龙 <yinglong.chen@uwa.edu.au>)

Abstract: Crop root systems are often poorly adapted to soils with the major limiting factors being poor soil water holding capacity and nutrient deficiencies in many farmland. Changes in RSA may mediate plant adaptation to soils low in nutrient availability. Root system architecture (RSA) significantly influences crop foraging and capturing soil water and nutrients and thus determines crop productivity. Wide-scale use of root-related genetic information in crop breeding programs relies on accurate phenotyping of relatively large populations. However, phenotyping of root-related traits remain a challenge in translating physiological and genetic advances in understanding the role of root systems in improved adaptation to abiotic stress and enhanced productivity. Recently we developed a semi-hydroponic phenotyping system for high-throughput phenotyping of root trait variability in substantial collections of several important crops, including maize. This presentation will take maize and narrow-leaved lupin as examples to showcase the utility and advantages of this phenotyping system in characterizing root system architecture of early growth stages. The development of root phenotyping, imaging and modeling technologies in studying RSA under edaphic stress provide assistance in selecting future crop genotypes with efficient root systems for enhanced abiotic stress tolerance and improved crop adaptation.

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Abstract 10

***ZmcytMdh4* encoding a cytosolic malate dehydrogenase is essential for storage reserve synthesis and seed development in maize**

Yongqiang Chen†(陈永强), Zhiyuan Fu*†(付志远), Jihua Tang*(汤继华)

National Key Laboratory of Wheat and Maize Crop Science / Collaborative Innovation Center of Henan Grain Crops / College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China. (河南农业大学国家小麦玉米作物学重点实验室/河南粮食作物协同创新中心/农学院, 郑州, 450002)

*Corresponding author: tangjihua1@163.com

(submitted by 陈永强<wuqiang646688@163.com>)

Abstract: Genes involved in starch and storage protein biosynthesis have been revealed in maize (*Zea mays*). However, the underlying regulation mechanism between them is almost unknown. In this study, we isolated a cytosolic malate dehydrogenase (*cytMdh4*) gene using map-based cloning from a natural maize kernel mutant *cytmdh4*, which has significant changes in starch and protein content compared to wild type. Transgenic and allelism test demonstrated that *cytMdh4* was the gene controlling the mutant phenotype. A 3-bp in-frame deletion in this gene was identified as the causative variation which might change the conformation resulting into repressed cytMDH4 enzymatic activity. Targeted metabolomics analysis showed that reduced cytMDH4 activity influenced amino acid synthesis, especially lysine, via affecting α -ketoglutaric acid and malate/oxaloacetate accumulation levels. Transcriptional profile showed that differentially expressed genes of zein, lysine degradation (*LKR/SDH*) and starch biosynthesis (*Bt2*, *Sh2*, and *Wx*) positively or negatively regulated by maize endosperm-specific transcription factor *opaque2* (*O2*). Concurrently, the decreased oxidation state caused by malate accumulation repressed the activity of AGP (*Bt2* and *Sh2*), which may in turn restricted the starch biosynthesis. Taken together, *ZmcytMdh4*, by affecting malate accumulation and gene expression of starch, amino acids synthesis, play critical role in regulating starch and storage protein balance in maize endosperm.

Key words: Maize, cytosolic malate dehydrogenase 4, amino acid metabolism, starch synthesis, TCA cycle.

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Abstract 11

Genome wide association study revealing the genetic basis of ionic variations in maize

Yuanyuan Chen^a, Min Jin^a, Zhenfei Chao^b, Yingjie Xiao^a, Haijun Liu^a, Daiyin Chao^b, Jianbing Yan^{a*}

^aNational Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

^bShanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200031, China

*Corresponding author: yjianbing@gmail.com

(submitted by 陈园园 <yychentamu@163.com>)

Abstract: Maize (*Zea mays* L.) is an important food and feed crop, which is widely grown and largely consumed in China. In order to achieve the sustainable agriculture development and ensure the national food security, it is crucial to improve the utilization efficiency of mineral nutrients in the soil and reduce the accumulation of toxic heavy metal elements in maize. In this study, we used an improved multiple parents advanced intercross population derived from 24 elite maize inbred lines, and collected the ionic data of the leaves and mature kernel. Based on the genome-wide association analysis, around 700 significant QTLs were detected for exploring the causal genes that controlling the elements variation. In this project, we intend to use the ionic element variations of different tissues in the population under different environments, take advantages of the genome and transcriptome data, to deeply analyze the genetic factors affecting the absorption, transport and accumulation of elements, and construct gene regulatory network; further elucidate the molecular mechanism of genes controlling the efficient use of mineral nutrients in maize, and provide useful theoretical information and genetic resources for the resistance of maize to heavy metal pollution and efficient use of mineral nutrients.

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Abstract 12

Functional characterization of *ZmMAP3K1* and the association of its natural variations in maize drought tolerance

Hui Zhang^{1#}, Lili Qi^{1#}, Suwen Han¹, Yanli Xiang¹, Xiaopeng Sun¹, and Mingqiu Dai^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

These authors contributed equally to this article.

* Corresponding author: mingqiudai@mail.hzau.edu.cn

(Submitted by 代明球<mingqiudai@mail.hzau.edu.cn>)

Abstract: Drought is one of the main abiotic stresses that reduce maize growth and yield. The mitogen-activated protein kinases (MAPKs) play important roles in Arabidopsis abiotic stress responses. How maize *MAPKs* (*ZmMAPKs*) and their natural variations involved in drought tolerance remain largely unknown. In this study, we functionally characterized *ZmMAP3K1*, a maize *MAPKKK*. We observed that *ZmMAP3K1* was expressed in multiple tissues and up-regulated by ABA and drought treatments. *ZmMAP3K1* was localized in the membrane and cytoplasm. Transgenic Arabidopsis plants overexpressing *ZmMAP3K1* were hypersensitive to ABA but hyposensitive to drought stresses, indicating that *ZmMAP3K1* had a positive role in regulation of plant drought tolerance. Protein-protein interaction assays showed that *ZmMAP3K1* interacted with *ZmMEK1*, and some *ZmPP2C-A* and all *ZmSnRK2* proteins, indicating that it might have a role in regulation of ABA signaling. Re-sequencing and association analysis showed that there were natural variations of *ZmMAP3K1* in 368 maize inbreds that were significantly associated with drought tolerance. InDel720, one of these natural variations, was verified by two F_{2:3} populations, and Insertion720 was proved to be a favorable allele of maize drought tolerance. Our studies provide evidences demonstrating the important roles of MAPKs in maize drought tolerance, and the natural variations identified in *ZmMAP3K1* have a potential use in breeding drought-tolerant maize cultivars.

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Abstract 13

Maize *Dek33* encodes a pyrimidine reductase in riboflavin biosynthesis essential for oilbody formation and ABA biosynthesis during seed development

Dawei Dai^a, Hongyang Tong^a, Lijun Cheng^a, Fei Peng^a, Tingting Zhang^a, Weiwei Qi^a and Rentao Song^{a, b, *}

^aShanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China.

^bNational Maize Improvement Center of China, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

*Address for correspondence to rentaosong@cau.edu.cn.

(submitted by 戴大伟<david200901@126.com>)

Abstract: The maize (*Zea mays*) classic kernel mutant *defective kernel 33* (*dek33*) produced defective and occasionally viviparous kernel phenotype. In this study, we cloned *Dek33* by positional cloning and found that it encodes the pyrimidine reductase in riboflavin biosynthesis. In *dek33*, a single base mutation (G-to-A) in the C-terminal COG3236 domain caused a premature stop codon (TGA), producing a weak mutant allele with significant reduction of truncated DEK33 protein and riboflavin content. The *dek33* mutation significantly affected oilbody formation and suppressed endoreduplication. The *dek33* mutation also disrupted ABA biosynthesis, resulting in less ABA content, which might be responsible for the viviparous embryo. In addition, our results indicated that the COG3236 domain is important for the protein stability of DEK33. The yeast two-hybrid experiment identified several proteins that interact with DEK33, including RGLG2 and SnRK1, suggesting possible post-translational regulation to DEK33 stability. The interaction between DEK33 and these proteins was further confirmed by luciferase complementation image assay. This study provided a weak mutant allele to explore cellular responses due to impaired riboflavin biosynthesis during seed development. Our findings indicated that the COG3236 domain might be an essential regulatory structure for DEK33 stability in maize.

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Abstract 14

A *helitron*-induced RabGDI α variant causes quantitative recessive resistance to maize rough dwarf disease

Suining Deng^{1,a}, Qingcai Liu^{1,a}, Baoshen Liu^{2,a}, Yongfu Tao^{1,a}, Haiyue Ai¹, Jianju Liu¹, Yongzhong Zhang², Yan Zhao², Mingliang Xu^{1,*}

¹ State Key Laboratory of Plant Physiology and Biochemistry/College of Agronomy and Biotechnology/National Maize Improvement Center/Center for Crop Functional Genomics and Molecular Breeding, China Agricultural University, 2 West Yuanmingyuan Road, Beijing 100193, P. R. China

² College of Agronomy/State Key Laboratory of Crop Biology, Shandong Agricultural University, Taian 271018, P. R. China

^a These authors contribute equally to this work

* Corresponding author: mxu@cau.edu.cn

(submitted by 邓穗宁<13391860723@163.com>)

Abstract: Maize rough dwarf disease (MRDD), caused by various *Fijiviruses* in the family *Reoviridae*, poses a grave threat to maize production worldwide. We previously identified a major quantitative trait locus on chromosome 8, *qMrdd1*, that confers recessive resistance to one causal pathogen of *Rice black-streaked dwarf virus* (RBSDV). Through map-based cloning strategy, we demonstrate that Rab GDP dissociation inhibitor alpha (*RabGDI α*) is the host susceptibility factor for RBSDV. A *helitron* transposon insert into intron 10 of *RabGDI α* creates alternative splice sites and replaces the wild exon 10 of *RabGDI α* with a *helitron*-derived exon 10, resulting in recessive resistance to RBSDV. In the process of RBSDV infection, the *helitron*-induced *RabGDI α* splicing variant showed almost the same gene expression profiles as *RabGDI α* regardless of developmental stages and resistance performance. We also identified the viral P7-1 protein as the pathogenicity determinant that targets the wild-type *RabGDI α* by binding to the exon-10-encoded peptide and C-terminal region to initiate viral infection. However, P7-1 has difficulty recruiting the *helitron*-induced *RabGDI α* variant, this may impair viral intercellular movement and eventually leads to quantitative recessive resistance to RBSDV. Additionally, we confirm that all naturally recessive resistance genes probably arose from a single *helitron* insertion event. This resistance allele can be useful to improve the resistance of maize varieties to MRDD by marker-assisted selection, and potentially to increase the resistance of other crops to RBSDV.

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Abstract 15

Construction and application of a genome-wide maize overexpression mutant library

Dengxiang Du, Ruchang Jin, Jinjie Guo, Fangdong zhang*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: fdzhang@mail.hzau.edu.cn

(submitted by 杜邓襄 < ddx@mail.hzau.edu.cn >)

Abstract: Mutagenesis library provides an extensive resource for identifying the biological functions of genes in plants. As there is a direct causal relationship between mutants and relative mutant genes, the study of gene function is the most convincing. Building a superexpression mutant using activation tagging is another mutagenesis strategy in contrast to the loss-of-function variety.

In this study, we generated an overexpression mutant population through transposon mediated activation tagging system. The modified manual transposon Ds activation tagging was transferred into maize inbred line A188 using agrobacterium, a total of 54 transformations, which located on different position of B73 chromosomes, were obtained to construction Ds-tagging lines. After crossing with a material that carried stable transposable enzymes, the modified manual transposon was activated and inserted into 2, 2290 positions of maize genomic. , and 1, 490 phenotypic mutations were screened. The phenotypes mutated included plant height, height of ear position, growth potential, leaf shape, leaf color, leaf number, branch of male ear, resistance to disease, etc. Not only dominant mutation phenotype was detected in the mutant population, but also a stable recessive mutant phenotype was identified produced by the escalation expression of candidate genes.

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Abstract 16

Genetic architecture of natural variation in maize embryo size

Xiaoxia Du, Xiaowei Li, Jiansheng Li, Xiaohong Yang

State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center of China, MOA Key Lab of Maize Biology, China Agricultural University, Beijing 100193, China

*Corresponding author: yxiaohong@cau.edu.cn

(submitted by 杜晓夏<duxx@cau.edu.cn >)

Abstract: Mature embryo accounts for about 10% of maize kernel dry weight, whereas accumulates nearly 82% and 26% of kernel oil and protein, respectively. Previous genetic studies have uncovered numerous genes in embryogenesis and embryo development by mutant analysis while genetic basis of quantitative variation for embryo size in natural population remains poorly understood. Here we performed a genome-wide association analysis for 4 embryo size-related traits, embryo length, embryo width, embryo volume and hundred embryo weight, in a set of 513 inbred lines using around 0.56 million high-quality SNPs. The marker-trait association identified 49 SNPs were significantly associated with 4 embryo-related traits at $P < 1.79 \times 10^{-6}$, which were resolved to 29 candidate genes. For each trait, the number of detected significant loci ranged from 8 to 11, and the total phenotypic variation explained ranged from 9.7% to 14.4%. The eQTL analysis strongly suggests that most of the detected genes affect the phenotypic variation via transcriptional regulation. Our results provide insights into the genetic basis of natural variation in maize embryo size.

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Abstract 17

KRN4 controls kernel row number by long-distance regulation of UB3 expression in maize

Yanfang Du¹, Yong Peng¹, Lei Liu¹, Xiaomeng Shen¹, Yunfu Li¹, Dan Liu¹, Ran Zhao¹, Yinan Jian¹, Fang Yang¹, Xingwang Li¹, Zuxin Zhang^{1,*}

¹National Key Laboratory of Crop Genetic Improvement, College of Plant Science and Technology, Huazhong Agricultural University, Wuhan 430070, P.R. China

Abstract: *KERNEL ROW NUMBER4 (KRN4)* is an intergenic QTL controlling maize yield trait, and maps close to *UNBRANCHED3 (UB3)*, a negative regulator of KRN. However, the mechanism by which *KRN4* controls *UB3* expression remains unknown. In this study, we found that allelic variation in *KRN4* changes *UB3* expression and inflorescence meristem diameter and KRN in two sets of near isogenic lines, indicating that *KRN4* is agronomically important. To understand the molecular basis of the interaction between *UB3* and *KRN4*, we used chromatin immunoprecipitation followed by paired-end tag sequencing (ChIA-PET). We also used the short enhancer-like element from different *KRN4* alleles upstream of a minimal CaMV 35S or a *UB3* promoter to drive *luciferase* expression. Using ChIP-seq, we found that both *UB3* and *KRN4* are direct targets of *UB2*, a paralog of *UB3*, and *UB3* expression varies with *UB2*. Two enhancer-binding factors, OBF1 and OBF4, interact with *UB2*, and bind to *KRN4*. Therefore, we propose that *KRN4* regulates *UB3* expression by direct chromatin interactions, and *UB2* may mediate the establishment or maintenance of the appropriate chromatin configuration, and recruits OBF1 and OBF4 to form a transcriptional complex to fine tune *UB3* expression and, in turn, KRN. These results provide evidence for fine tuning of gene expression by intergenic QTLs in maize, and a new perspective for genetic control of a quantitative trait.

Keywords: Maize (*Zea mays L.*); Kernel row number; Intergenic region; Chromatin interaction; Enhancer.

Abstract 18

玉米 PYS1 蛋白调控叶绿体发育的分子机理研究

段志坤 王茂 李凯文 郭思义 李保珠 宋纯鹏*

河南大学, 植物逆境生物学重点实验室, 河南开封, 475004

*通讯作者: songcp@henu.edu.cn

(submitted by 段志坤<duanzhikun@henu.edu.cn>)

摘要: 叶绿体是植物所特有的半自主细胞器, 它不仅是植物进行光合作用为生命提供碳水化合物和能量的场所, 还参与合成氨基酸、脂肪酸、维生素、四吡咯等生长发育所必需的化合物。因此, 叶绿体的发育和功能状态对于植物生命活动极其重要。为了研究玉米叶绿体发育的调控机理, 我们从 maizeGDB (www.maizegdb.org) 网站订购一批具有高叶绿素荧光表型的突变体。经过筛选发现, 玉米 *pys1* (pale yellow seedling 1) 突变体幼苗严重黄化且在温室中土培 14 天后死亡。使用叶绿素荧光成像系统 CF Imager 检测发现, *pys1* 突变体最大光化学效率 Fv/Fm 显著低于野生型植株水平。进一步利用电镜观察发现 *pys1* 突变体叶绿体皱缩而不能维持正常的椭球形, 基质片层数量明显减少, 几乎没有基粒的垛叠。综上所述, *pys1* 是一个叶绿体发育缺陷的突变体。我们利用原生质体瞬时转化的方法确定 PYS1 定位于叶绿体。为了确定 PYS1 作用于叶绿体发育的具体时期, 我们利用转录组学 RNA-seq 的方法分析了突变体见光前、后基因表达的差异。结果显示, 突变体叶绿体发育相关基因在见光前期即与野生型存在差异, 且见光后差异更显著。进一步进行可变剪切的分析, 我们发现 PYS1 可能是作用于叶绿体发育相关基因的可变剪切过程。具体的作用位点有待于进一步验证。

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Abstract 19

玉米转录因子 *BES1/BZR1-7* 基因克隆及功能分析

冯文奇¹, 孙福艾², 路风中³, 刘炳良⁴, 李晚忱*, 付凤玲*, 于好强*,
四川农业大学玉米研究所, 成都, 61130

*通讯作者: yhq@sicau.tea.cn (submitted by 冯文奇<986960563@qq.com>)

摘要: BES1/BZR1 蛋白是油菜素内酯信号途径的唯一转录因子, 在植物生长发育及逆境应答中起重要作用。为研究玉米 *ZmBES1/BZR1-7* 基因的功能, 我们从玉米自交系 B73 中克隆到 *ZmBES1/BZR1-7* 基因, 对其功能进行初步分析。序列分析表明, *ZmBES1/BZR1-7* 基因无内含子, 开放阅读框 (Open reading frame, ORF) 长 975 bp, 编码 324 个氨基酸。*ZmBES1/BZR1-7* 蛋白 N 端高度保守, 包含 bHLH 结构域, 与水稻 OsBZR1-1 相似性达 75.95%。实时荧光定量 PCR 结果表明, *ZmBES1/BZR1-7* 在玉米叶和根中的表达显著受渗透与盐胁迫诱导; 处理 12h 后, 叶片中表达量达对照的 17.8 倍。酵母激活实验结果表明, *ZmBES1/BZR1-7* 具有转录激活活性。共相关分析结果表明, 玉米中有 72 个基因与 *ZmBES1/BZR1-7* 的表达相关性较高, 其启动子区均含有 E-box 或 BRRE 元件, 主要参与细胞、细胞组份、细胞代谢过程等。本研究为深入解析玉米 BES1/BZR1 的功能及其调控网络奠定了基础。

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Abstract 20

Variation of *ZmDRO1* gene to ABA response confers the difference of drought avoidance between modern maize and *Zea mays* ssp.

mexicana

Xuanjun Feng¹, Yunting Cai¹, Qinjun Wang¹, Zhengqiao Liao¹, Jia Li¹, Yanli Lu^{1*}

¹Maize Research Institute, Sichuan Agricultural University, Wenjiang 611130, Sichuan, China

*Corresponding author E-mail: luyanli@sicau.edu.cn

(submitted by 冯宣军 <xuanjunfeng@sicau.edu.cn>)

Abstract: Root Architecture Remodeling (RAR) is essential for plant to search moisture under water-limited soil. *DRO1* was reported, in *Oryza*, *Arabidopsis* and *Prunus*, to promote the formation of steeper root system, therefore, the plant maintained better vitality under drought conditions by acquiring water from deeper soil. Here, we reported that amino acid sequence of *ZmDRO1* is conservative between modern maize B73 and an ancestor *Zea mays* ssp. *mexicana* (thereafter, *mexicana*). However, the noncoding region, basal expression and ABA response were remarkably different between B73 and *mexicana*. *ZmDRO1* displayed lower basal expression but stronger response to ABA induction in *mexicana*. It was consistent with the phenotypic data that *mexicana* has smaller root growth angle under favorable environment but more remarkable root architecture remodeling toward deeper soil under drought stress. So, *mexicana* displayed less penalty of biomass than B73 under water limitation. Further, gene expression and RAR was analyzed in a recombinant inbred line (RIL) population and the positive function of *ZmDRO1* in drought avoidance was confirmed. Based on above results, ABA-induced high active artificial promoter was designed to control the expression of *ZmDRO1* in transgenic modern maize cultivar. Transgenic lines had higher *ZmDRO1* expression under water limitation and consequently promoted RAR toward deeper soil to avoid drought stress and maintain high yield performance. Collectively, these results demonstrated the application of *ZmDRO1* via ABA-inducible strategy to alter root architecture for drought avoidance.

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Abstract 21

Characterization and fine mapping of stay-green loci for leaf senescence in maize (*Zea mays* L.)

Xue Feng^{1,2}, Lili Liu^{1,2}, Zhigang Li^{1,2}, Huaiqing Hao¹, Dongyun Hao³, Haichun Jing^{1,2*}

¹ Key Laboratory of Plant Resources, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China

² University of Chinese Academy of Sciences, Beijing 100049, China

³ Key Laboratory for Molecular Enzymology and Engineering of the Ministry of Education, Jilin University, Changchun 130012, China

*Corresponding author: hcjing@ibcas.ac.cn

(submitted by 冯雪 <xuefeng@ibcas.ac.cn>)

Abstract: Senescence, one of the vital stages in leaf development, plays crucial role in crop yields. To explore the process of leaf senescence in maize (*Zea mays* L.), two inbred lines were used in this study. Leaves of 4-144 exhibited a stay-green phenotype 30 DAP (Days after pollination), while 4-287 was senescent. We developed a mapping population by crossing 4-287 (female parent) and 4-144 (male parent), identified two major QTLs (*stg3* and *stg7*) with F₂ population. QTL-seq (whole-genome sequencing-based QTL mapping) on the BC₃F₂ population grown in 2015, delimited the major locus to 15.35 Mb physical interval in chr3 (*stg3*) and 3.56 Mb physical interval in chr7 (*stg7*), respectively. To further narrowing down the QTL regions, SNP and Indel markers were developed. Locus of *stg3* was narrowed down to a 5.5 Mb interval between markers A000681 and A006030. Meanwhile, *stg7* was narrowed down to a 361.5 Kb interval between markers G2 and G5. Compared with the senescent parent 4-287, NILs performed the phenotype of delaying senescence obviously. Besides, Ear weight of F₁ hybrid (*stg7*-NIL × 4-144) was obviously better than JD27 (4-287 × 4-144). In total, this study identified two major loci for leaf senescence in maize which provides new insight for high quality crop breeding.

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Abstract 22

Maize *Dek45* encodes a non-zein PB protein that interacts with 22kD zein

Yang Feng¹, Yafei Ma², Fan Feng², Weiwei Qi², Rentao Song^{1,*}

1 State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100094, China

2 Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

*Corresponding author: rentaosong@cau.edu.cn

Abstract: Protein body (PB) is the storage protein organelle in maize endosperm cells. Different type of zeins contribute the main structure of PB. However, there are also other non-zein PB proteins detected in PB. Few non-zein PB proteins, such as O1, FL1 and O10 were identified and found to play important roles in PB morphology and zein deposition. In this study, a new maize defective kernel mutant *dek45* was characterized by small, collapsed kernels and lethal seedlings. The *Dek45* was cloned by *Mu*-tag isolation and confirmed by allelism test. Subcellular fraction assay and immune-electron microscopic analysis indicated that DEK45 mainly presented in the PBs. Yeast two hybrid assay and BiLUC assay revealed that DEK45 interacts with 22kDa α zein. These results suggested a potential mechanism of non-zein proteins transport to PB through its interaction with major zein proteins. In addition, further studies revealed that, *Dek45* is also important to mitochondria function. Transcriptome analysis revealed enhanced expression of genes in alternative respiratory pathway and mitochondria function. Morphologic analysis indicated that the structure of mitochondria was badly damaged in *dek45*. *Dek45* offers a new perspective of the research of non-zein PB proteins transport mechanism and the function of non-zein PB protein in other organelles.

Abstract 23

A new subclass of the mitochondrial-localized RNA-binding proteins required for seed development in maize and Arabidopsis

Yu Feng², Qiuzhen Tian¹, Hongyu Chen¹, Jihua Tang¹ and Guifeng Wang^{1*}

¹ Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China;

² Shanghai Key Laboratory of Bio-Energy Crops, School of Life Sciences, Shanghai University, Shanghai 200444, China.

*Corresponding author: guifeng.wang@henau.edu.cn

(submitted by Yu Feng <243940065@qq.com>)

Abstract: Maize provides food, feed and industrial raw materials of worldwide importance. Maize kernel-related mutants are important resources for elucidating genetic control of the deposition of storage compounds and seed development. The maize mutant 1759 is a single-gene controlled recessive mutation via EMS mutagenesis. The mutant kernel is small, pale and shrunken, as well as empty pericarp. The causal gene was in a region of about 1300kb covering 15 candidate genes, by analyzing an F2 population of 1424 individuals. DNA sequencing indicated that a C-to-T transition appears in the first exon of gene A, thus causing an amino acid substitution from proline to serine. Maize association population analysis revealed that the SNP is unique to mutant 1759. Strikingly, disruption of each homologs of gene A in the same subclass uniformly leads to seed abortion in Arabidopsis. Gene A encodes an RNA-binding protein targeted into mitochondria. Unexpectedly, biochemical analysis indicated that no significant difference was detected in mitochondrial function of the mutant in contrast to the wild type. New data suggested that this subclass RNA-binding protein are mostly likely to implicate in mitoribosomal biogenesis.

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Abstract 24

玉米棕色叶脉突变体基因鉴定及功能分析

付春祥^{1*}, 吴振映¹, 熊王丹¹, 任浩², Roje Sanja³, 齐天雄¹, 白泽涛¹, 刘雨辰¹, 苏昆龙¹

¹中国科学院青岛生物能源与过程研究所, 青岛, 266101

²南京林业大学, 南京, 210037

³美国华盛顿州立大学, 99164

*通讯作者: fucx@qibebt.ac.cn

(Submitted by 付春祥<fucx@qibebt.ac.cn>)

摘要: 玉米棕色叶脉突变体 (*brown midrib mutant*) 最初发现于 1924 年, 至今已有 90 多年的历史, 是研究单子叶植物细胞壁木质素合成调控的重要资源。目前共发现 6 种不同座位的 *bm* 突变体 (*bm1-6*), 其中, *bm2* 和 *bm3* 已经被应用于商业化育种, 并产生了高细胞壁利用效率的青贮玉米新品种。目前本实验室已经完成 *bm1-6* 玉米棕色叶脉突变体的基因鉴定, 发现上述 *bm* 基因分别属于木质素单体合成途径和植物一碳代谢途径。进一步对 *bm2* 基因——*MTHFR* 进行了深入研究, 发现 *MTHFR* 是一碳代谢途径的关键酶, 联接四氢叶酸和甲硫氨酸循环, 潜在影响木质素甲基化单体合成的甲基供体—*S*-腺苷甲硫氨酸 (*SAM*) 的合成。一碳代谢途径中间体分析表明, *MTHFR* 突变后导致其编码蛋白催化的底物 5, 10-亚甲基四氢叶酸显著积累, 而其产物 5-甲基四氢叶酸的水平则显著降低。令人意外的是, *MTHFR* 所催化反应下游的 *SAM* 水平却没有发生改变。进一步研究表明, *SAM* 经甲基化反应后生成的去甲基化产物—*S*-腺苷高半胱氨酸 (*SAH*) 在突变体中显著积累, 最终导致较高的 *SAH/SAM* 比例。体外玉米叶脉粗蛋白提取物和木质素氧甲基化酶体外重组蛋白对木质素中间体的甲基化催化活性分析表明, *SAH* 能够显著抑制木质素中间体的甲基化反应。由于玉米遗传转化效率的制约, 我们利用本实验室同属禾本科 C4 植物的柳枝稷为遗传转化材料, 系统研究了包括 *SAM* 合成酶 (*SAMS*) 和 *SAH* 水解酶 (*SAHH*) 在内的涉及 *SAM* 和 *SAH* 代谢的多个关键酶基因对木质素合成的影响, 发现无论单独降低 *SAM* 水平还是单独提高 *SAH* 水平, 均能够抑制木质素甲基化单体的合成。我们研究说明, 调控植物一碳代谢能够显著改变木质素氧甲基化酶的底物 *SAM* 和抑制剂 *SAH* 的代谢平衡, 从而影响木质素甲基化单体的合成。因此今后对一碳代谢途径调控机制的深入解析, 有望为重要粮食和经济作物的细胞壁高效利用和抗倒伏抗逆性的提升, 提供更多更新的靶标。

基金项目: The National Key Technologies Research & Development Program-Seven major crops breeding project (2016YFD0101803).

Abstract 25

Genetic basis of selection for kernel nutritional traits during maize domestication and improvement

Hui Fang^{1,2,4}, Xiuyi Fu^{1,2,4}, Yuebin Wang^{3,4}, Jing Xu^{1,2}, Haiying Feng^{1,2}, Weiya Li^{1,2}, Orawan Jittham^{1,2}, Lili Zhang^{1,2}, Ning Yang³, Gen Xu^{1,2}, Min Wang^{1,2}, Xiaowei Li^{1,2}, Jiansheng Li^{1,2}, Jianbing Yan³, Xiaohong Yang^{1,2*}

¹State Key Laboratory of Plant Physiology and Biochemistry, China Agricultural University, Beijing 100193, China.

²National Maize Improvement Center of China, MOA Key Lab of Maize Biology, Beijing Key Laboratory of Crop Genetic Improvement, China Agricultural University, Beijing 100193, China.

³National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

⁴These authors contributed equally to this study.

*Corresponding author: yxiaohong@cau.edu.cn.

(submitted by 付修义 <fuxiuyicau@163.com>)

Abstract: Maize kernel nutritional traits are important in terms of human and animal nutrition, and experienced a selection due to human diverse needs. The knowledge about the genetic basis of selection for kernel nutritional traits during maize domestication and improvement is limited. Here we performed a QTL mapping for 19 oil and 14 carotenoid related traits in a maize-teosinte population derived from maize inbred line Mo17 and teosinte X26-4. We found both single and epistatic QTL contribute to the oil and carotenoid variation between maize and teosinte, and over half of teosinte alleles of single additive-effect QTL contribute to the increase of the values of the detected traits. The constructed trait-QTL network revealed the traits in the same metabolic pathway shared a great number of common loci, while less for the traits in different metabolic pathway, consistent with the metabolic nature of oil and carotenoid. The selection features and evolutionary trajectories of the genes or loci underlying oil and carotenoid variation during maize domestication and improvement are complex. We identified a mutator distance relative (MuDR) transposable element (TE) in the first intron of *DXS2* encoding a limited enzyme in the MEP pathway, which increases carotenoid accumulation via enhancing *DXS2* expression level. TE insertion arose after domestication and underwent divergent selection in yellow and white maize. These results provide new insights into the genetic basis of selection for oil and carotenoid related traits during maize domestication and improvement.

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Abstract 26

Linkage mapping and GWAS reveal candidate genes conferring thermo-tolerance of seed-set in maize

Jingyang Gao¹, Songfeng Wang², Zijian Zhou¹, Shiwei Wang¹,Chaopei Dong¹,Cong Mu¹,Yunxia Song¹, Peipei Ma¹, Chengcheng Li²,Zhao Wang¹,Kewei He¹,Chunyan Han²,Jiafa Chen², Haidong Yu^{2*}, Jianyu Wu^{1*}.

¹National Key Laboratory of Wheat and Maize Crop Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou, 450002, China

² College of Life Sciences, Synergetic Innovation Center of Henan Grain Crops and National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding Author: wujianyu40@126.com

(submitted by 高景阳<swgc2013@163.com>)

Abstract: High-temperature stress (HS) will increasingly affect crop yield worldwide. In order to determine the genetic basis of thermo-tolerance of seed-set in maize in field conditions, a QTL mapping in a recombinant inbred line (RIL) population was performed using a collection of 8329 high-density single nucleotide polymorphisms (SNP) markers developed in this study, combined with a genome-wide association study (GWAS) of 261 diverse maize lines using 259,973 SNPs. In total, 4 quantitative trait loci (QTLs) and 17 genes associated with 42 SNPs related to thermo-tolerance of seed-set were identified by linkage mapping and GWAS, respectively. Four candidate genes among them were found in both linkage mapping and GWAS. Thermo-tolerance on seed-set were increased significantly in the near-isogenic lines (NILs) incorporating the four genes in a susceptible parent background. Moreover, the expression profiles of two of the four candidate genes showed that they were induced by high temperature in maize tassel in the tolerant parent background. These genetic analyses indicated that thermo-tolerance of maize seed-set is regulated by multiple genes with minor effect, in which calcium signaling plays a core role. The pyramiding breeding with beneficial alleles and candidate genes could improve seed-set and yield of maize under HS.

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甜玉米自然老化其生理生化机制分析

宫捷，张丽萍，范竞升，谢富贵，冯发强

华南农业大学 农学院/广东省植物分子育种重点实验室，广东 广州 510642

摘要：【目的】为探究甜玉米种子自然老化耐性差异，以及老化对种子生理生化的影响，揭示甜玉米种子活力的调控机理。【方法】本研究选用亲缘关系较远、遗传多样性丰富的 21 份甜玉米自交系，分别自然老化处理 2、4 和 6 个月。发芽实验检测种子活力相关指标，包含发芽势（GP）、发芽率（GR）、发芽指数（GI）、活力指数（VI）；采用酶联免疫法、蒽酮比色法、BCA 蛋白浓度测定自然老化过程中的生理生化指标，包括超氧化物歧化酶（SOD）、过氧化氢酶（CAT）、过氧化物酶（POD）、淀粉酶（AMY）、赤霉素（GA）、脱落酸（ABA）、总淀粉（TS）、总蛋白（TP）、丙二醛（MDA）。对老化处理 6 个月的发芽率差异较大的两份自交系进行了扫描电镜观察。【结果】随着老化时间增加，各自交系的 GP、GR、GI、VI 逐渐降低，表明自然老化降低种子活力。相关性分析发现，老化不同时期种子活力各指标间达显著正相关。对种子活力各指标和生理生化指标间进行方差分析发现，除 GP 外，其他指标在材料间均差异显著；相关分析发现，SOD 与 GR、GI 呈极显著正相关，CAT 与 VI 呈显著负相关，而与 TP 极显著正相关，AMY 与 TP 极显著正相关。扫描电镜观察发现高活力自交系种子淀粉粒排列紧密，较少发生分解，而低活力种子淀粉粒有明显的分解表象。【结论】自然老化会影响种子内部的生理生化反应且不同自交系间差异显著，表明遗传因子是种子的耐老化的重要因素，不同耐老化自交系间的淀粉粒分解存在差异。

关键词：甜玉米 自然老化 种子活力 生理生化分析

Abstract 28

The host-virus protein interaction network in maize resistance to sugarcane mosaic virus

Yangqing Gong¹, Qingqing Liu¹, Pengfei Leng¹, Qianya Xu¹, Mingliang Xu^{1*}

¹National Maize Improvement Center of China, China Agricultural University, Beijing 100193, China

*Corresponding author: mxu@cau.edu.cn

(submitted by 龚央庆<gyq15201433474@126.com>)

Abstract: *Sugarcane mosaic virus* (SCMV), belonging to genus Potyvirus, family Potyviridae, is the causal agent to cause a devastating maize viral disease. In our previous study, we demonstrated that *ZmTrxh* and *ZmABP1* are two genes conferring maize resistance to SCMV. *ZmTrxh* is an atypical h-type thioredoxin protein that acts as molecular chaperone to impart early resistance to SCMV, while *ZmABP1* was considered as auxin binding receptor to function mainly at later infection stage. However, the molecular mechanism underlying maize resistance to SCMV remains unclear. In the current study, we used *ZmTrxh* and *ZmABP1* as bait proteins to perform yeast-two-hybrid to screen their target proteins. Consequently, we obtained two *ZmTrxh*-interacting host proteins, SLK and SO, and one *ZmABP1*-interacting protein Bsd. Intriguingly, SLK was also demonstrated to interact with the viral 6K1, which was involved in virus replication and movement. Similarly, Bsd was found to interact with the virial protein CI, an important component of the virus wheel inclusion body to help virus movement between cells. Taken together, these results suggest that *ZmTrxh* and *ZmABP1* can protect plant proteins from virus hijacking, thereby enhance maize defense response upon SCMV infection.

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Abstract 29

New insight into lignin biosynthesis: P450 complex formation and electron shuttle

Mingyue Gou^{1, 2*}, Xiaoman Yang², Yunjun Zhao², Xiuzhi Ran², Yanzhai Song², and Chang-Jun Liu²

¹ State Key Laboratory of Wheat and Maize Crop Science, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China

² Biology Department, Brookhaven National Laboratory, Upton, NY 11973

*Corresponding author: mingyuegou@163.com

(submitted by 苟明月 <mingyuegou@163.com>)

Abstract: Lignin is a complex and irregular biopolymer that support secondary cell wall for water conductance and plant strength. Its biosynthesis requires three endoplasmic reticulum (ER)-resident cytochrome P450 monooxygenases, C4H, C3' H and F5H, to establish the structural characteristics of its monomeric precursors p-hydroxyphenyl (H), guaiacyl (G) and syringyl (S). These P450 enzymes were reported to associate with each other or potentially with other soluble monolignol biosynthetic enzymes to form an enzyme complex. However, the molecular basis governing such enzyme organization and functioning remains elusive. We conducted immunoprecipitation (IP) coupled with Liquid chromatography-mass spectrometry (LC-MS) to identify the potential protein complex associated with these P450 enzymes in Arabidopsis; by which, we collectively discovered multiple potential P450 interacting proteins. Among which, we found Arabidopsis membrane steroid-binding proteins (MSBPs) serve as a scaffold to physically organize monolignol P450 monooxygenases, thereby regulating the lignin biosynthetic process; while cytochrome b5 isoform D (CB5D) is an indispensable electron shuttle protein specific for S-lignin biosynthesis. Downregulation of MSBP genes substantially impairs the stability and activity of the MSBP-interacting P450 enzymes and, consequently, total lignin deposition. Disrupting *CB5D* results in >60% reduction of S lignin subunits but no impairment on G-lignin formation. Our study demonstrates that MSBP proteins are essential structural components that physically organize and stabilize the monolignol biosynthetic P450 enzyme complex, and CB5D functions as an obligate electron shuttle intermediate specifically augmenting F5H catalyzed reaction thereby controlling S-lignin biosynthesis.

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Abstract 30

Identification of Quantitative trait loci associated with drought tolerance at reproductive stage in maize (*Zea mays*. L) across multi-environments

Xinmin Hu^{1,2}, Guihua Wang¹, Zhenxiang Xu¹, Hongwei Zhang², Jianhua Wang¹, Riliang Gu^{1*}, Guoying Wang^{2*}

1 College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

2 Institute of Crop Sciences, Chinese Academy of Agricultural Sciences, Beijing 100081, China

* Corresponding authors: rilianggu@cau.edu.cn and wangguoying@caas.cn.

Abstract: As global climatic changes, maize production affected by drought stress more and more. Application of haploid technology and marker-assisted selection will accelerate identify quantitative trait loci (QTL) and screen materials for drought tolerance. A double haploid (DH) population created from the cross han21(drought tolerant) × ye478(drought sensitive), including 217 lines was utilized to identify stable QTL (sQTL) in field trials by multiple locations in many years. Significant phenotypic variation was observed for grain yield (GY), anthesis-silking interval (ASI) and ear setting percentage (ESP) between contrasting water regimes. A total of 105 QTL was detected with single-environment QTL analysis, spreading on all ten chromosomes. Fourteen sQTL were identified across at least two environments or traits. Fifty-nine BC₃F₇ families derived from the same parent certified that introgression of some sQTL could improve plant drought tolerance. This work provides new chromosomal regions for fine mapping and molecular marker-assisted breeding.

Abstract 31

Two Transcripts of *ZmHsf12* Have Distinct Functions in Arabidopsis Under Heat Stress

Yinrui Ge¹, Mingyue Guo¹, Jing Wang¹, Nannan Song¹, Qianqian Qin¹, Qizhi Liang¹, Weina Si¹, Haiyang Jiang^{1*}

¹National Engineering Laboratory of Crop Stress Resistance Breeding, Anhui Agricultural University, Hefei 230036, China

*Corresponding author: hyjiang@ahau.edu.cn

(submitted by 郭明月 <GuoMingyue@ahau.edu.cn>)

Abstract: Heat stress has adverse effects on plant growth and crop yield. Heat shock transcription factors (HSFs) are highly conserved in plants, involving in a variety of abiotic stress responses. We identified a potential stress-related maize HSF gene, *ZmHsf12*. Two transcripts of *ZmHsf12*, *ZmHsf12I* and *ZmHsf12II*, were obtained by RT-PCR. Compared with *ZmHsf12I*, 15 amino acids were absent in the DNA binding domain of *ZmHsf12II*. Subcellular localization analysis suggested that both of them were located in the nucleus. Analysis of Transcriptional activity confirmed that *ZmHsf12II* has transcription activity in yeast cells, while *ZmHsf12I* has no transcription activity, which indicated that the extra 15 amino acids might be response for the loss of transcription activity in *ZmHsf12I*. Finally, the two transcripts were overexpressed in Arabidopsis, respectively. We found that *ZmHsf12II*-overexpressing transgenic line has significantly higher survival rates under heat stress and sensitivity to ABA than wild type, while no visible phenotypic differences could be observed between *ZmHsf12I*-overexpressing transgenic lines and wild type. Altogether, these results indicate that the two transcripts of *ZmHsf12* have distinct functions and *ZmHsf12II* gene could enhance the thermotolerance of Arabidopsis by a ABA-mediated heat shock pathway.

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Abstract 32

Cloning and expression of *ZmXTH23* in maize (*Zea mays*) and its response to salt and drought stress

CHEN Dong-Bin^{1,3}, LI Jun^{2,3}, GUO Xin-Mei^{2,3*}, SONG Xi-Yun^{2,3*}

1 College of Life Science, Qingdao Agricultural University, Qingdao 266109, Shandong, China; 2 College of Agronomy, Qingdao Agricultural University, Qingdao 266109, Shandong, China; 3 Key Laboratory of Major Crop Germplasm Innovation and Application in Qingdao, Qingdao 266109, Shandong, China

* Corresponding author, xmguo2009@126.com; songxy@qau.edu.cn.

(submitted by 郭新梅<xmguo2009@126.com>)

Abstract: Xyloglucan endotransglucosylase/hydrolase(XTH) is a key enzyme in the process of plant cell wall remodeling which participates in the regulation of plant growth and development. To elucidate and explore the function of XTH in the process of abiotic stress response, *ZmXTH23*(LOC100191584) in XTH family was cloned from maize inbred line Chang7-2 and its biological function was studied. Bioinformatics analysis showed that *ZmXTH23* contained a complete open reading frame of 897 bp encoded 298 amino acids and was a member of LamG superfamily. Besides, it performed xyloglucan endotransglycosidase (XET) activity and its amino acid sequence had the closest affinity with that of *PmXTH25* (RLM56175.1) of *Panicum miliaceum*(Similarity reached 90%).The expression pattern of *ZmXTH23* was analyzed by qRT-PCR. The results showed that the expression of *ZmXTH23* was tissue-specific, it had the highest expression level in young stem($P<0.01$), and was induced by ABA, NaCl and PEG6000. Prokaryotic expression analysis of SDS-PAGE and western blot confirmed that *ZmXTH23* protein could be expressed in *E. coli* BL21 recombinant host strain. The growth of host bacteria pET28a-*ZmXTH23* under different concentrations of salt and mannitol stress was better than that of host bacteria pET28a, which indicated that the *ZmXTH23* protein enhanced the salt tolerance and drought resistance of pET28a-*ZmXTH23* recombinant host strain. In conclusion, *ZmXTH23* plays an important role in response to abiotic stress and the functional study of *ZmXTH23* gene provides basic information for the creation of new maize germplasm resistant to stress.

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Abstract 33

Cloning and characterization of *ZmELS2* gene that regulates leaf pre-senescence in maize

Zhanhui Zhang, Hui Zhang, Jihua Tang, Zhanyong Guo*

Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding author: gu0zhy@163.com

(Submitted by 郭战勇 < gu0zhy@163.com >)

Abstract: The growth and maize grain yield depends on the precise regulation of photosynthesis of leaves. Green-stay plays an important role in achieving high yield, however, pre-senescence will decrease the maize grain yield. Therefore, dissecting the molecular mechanisms underlying in leaf senescence is of crucial importance for breeding elite maize varieties. In our previous study, a single gene inherited from a maize mutant with pre-senescence phenotype was identified, named as maize *early leaf senescence 2* (*ZmELS2*). The leaves of the mutant start aging at V6 stage. And the mutants have irregular ears, with few kernels on the ears. These phenotypes implied that the mutant gene is associated with the onset of leaf senescence. An F₂ segregation population was constructed for candidate gene mapping, by crossing *ZmELS2* mutants with an inbred line L119-8. Combining BSA strategy and subsequent fine mapping procedure, the candidate gene was mapped on Chr.1, with a physical distance of 91.7 kb. This region contains two protein coding genes. Incorporating functional predication and variants analysis, the candidate gene was identified encodings a member of Inositol monophosphatase 3 family. This candidate gene has a base transversion (C to T) at the 64th base of the first exon. Inositol monophosphatase is an important component in the phosphatidylinositol signaling pathway that is proved to regulate environmental factor responding. Hereby, *ZmELS2* is possible to determine the onset of leaf pre-senescence through regulating the levels of phosphatidylinositol. The present work will benefit further gene cloning, and provide technical support for breeding elite inbred lines and hybrids.

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Abstract 34

Phylogenetic analysis and syntenic network characterize the conserved and diverse evolutionary patterns of *NAC* gene families in plants

Zelong Li^{1,2}, Chengzhi Ren^{1,2}, Junfeng Tan^{1,2}, Ting Zhang^{1,2}, Xiaohong Chen^{1,2}, Zhaoxue Han^{1,2*}

¹State Key Laboratory of Crop Stress Biology for Arid Areas, College of Life Sciences, Northwest A&F University, Shaanxi, Yangling 712100, China

²College of Life Sciences, Northwest A&F University, Shaanxi, Yangling 712100, China

*Corresponding author: han_zx@126.com

(submitted by 韩兆雪 < han_zx@126.com >)

Abstract: The *NAC* proteins are plant-specific transcription factors and play crucial roles in diverse biological processes, such as growth, development, and adaption to adverse factors. In this study, we performed a systematic bioinformatics analysis of the *NAC* gene families across 51 complete plant genomes. We identified 5399 putative *NAC* transcription factor genes that were clustered into 11 distinct clades. The *NACs* in angiosperms are widely distributed in all clades, however, *NACs* in green algae, mosses, ferns and gymnosperms exhibit clade-specific distribution. For the purpose of investigating the evolutionary history of the *NAC* genes in angiosperms, we analyzed the *NACs* in *Arabidopsis thaliana* and *Zea mays*, and fifteen common ancestral genes were identified between them. Meanwhile, we observed that *NACs* in *Zea mays* and *Arabidopsis thaliana* have different expansion patterns: The expansion efficiency of *NAC* genes in *Arabidopsis thaliana* decreased significantly after Eudicots and Monocots diverged. To further observe the evolution and diversification patterns of *NAC* genes, we performed a syntenic analysis of *NAC* genes in 51 species and identified conserved and diverse evolution patterns from the results of syntenic network clusters. Taken together, these results provide new hypotheses for the expansion and functional diversification of the *NAC* gene families.

Abstract 35

Maize *Dek15* encodes the cohesin-loading complex subunit SCC4 and is essential for chromosome segregation and kernel development

Yonghui He¹, Jinguang Wang², Weiwei Qi², and Rentao Song^{1*}

¹ State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

² Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

*Correspondence Author: rentaosong@cau.edu.cn

(submitted by 何永辉<heyonghui@cau.edu.cn>)

Abstract: Cohesin complexes maintain sister chromatid cohesion to ensure proper chromosome segregation during mitosis and meiosis. In plants, the exact components and functions of the cohesin complex remain poorly understood. Here, we positionally cloned the classic maize (*Zea mays*) mutant *defective kernel 15* (*dek15*), revealing that it encodes a homolog of SISTER CHROMATID COHESION PROTEIN 4 (SCC4), a loader subunit of the cohesin ring. Developing *dek15* kernels contained fewer cells than the wild type, but had a highly variable cell size. The *dek15* mutation was found to disrupt the mitotic cell cycle and endoreduplication, resulting in a reduced endosperm and embryo lethality. The cells in the *dek15* endosperm and embryo exhibited precocious sister chromatid separation and other chromosome segregation errors, including misaligned chromosomes, lagging chromosomes, and micronuclei, resulting in a high percentage of aneuploid cells. The loss of *Dek15/Scc4* function upregulated the expression of genes involved in cell cycle progression and stress responses, and downregulated key genes involved in organic synthesis during maize endosperm development. Our yeast two-hybrid screen identified the chromatin remodeling proteins chromatin remodeling factor 4 (CHR4), chromatin remodeling complex subunit B (CHB)102, CHB105, and CHB106 as SCC4-interacting proteins, suggesting a possible mechanism by which the cohesin ring is loaded onto chromatin in plant cells. This study revealed biological functions for DEK15/SCC4 in mitotic chromosome segregation and kernel development in maize.

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Abstract 36

玉米苞叶层数主效 QTL-*qHN1* 的精细定位

于停¹, 王树楷¹, 韩艳斌¹, 张国斌¹, 陈翠霞^{1*}

¹山东农业大学农学院, 泰安, 271018

*通讯作者: cxchen@sdaa.edu.cn

(submitted by 陈翠霞<cxchen@sdaa.edu.cn>)

摘要: 玉米收获期籽粒脱水速率慢、含水率较高不仅阻碍了玉米生产全程机械化步伐, 而且严重影响玉米收获质量。研究表明苞叶层数是影响玉米成熟后期籽粒脱水速率快慢的主要性状之一。为了研究通过合理减少苞叶层数, 选育出籽粒脱水速率快、适宜机收玉米品种, 我们对560份来自中国 and 美国的玉米骨干自交系进行多年多点的苞叶性状调查, 并从中筛选出4份苞叶层数表型稳定且极端的玉米材料, 将其作为研究对象组配作图群体。利用CM23和CC76构建的178个重组自交系 (Recombinant Inbred Lines, RILs) 家系群体, 结合234对SSR标记构建连锁图谱和2年表型数据, 运用完备复合区间作图法对玉米苞叶层数进行QTL定位, 共检测到5个主效QTLs, 分别位于5条染色体上。同时利用PG3和L11构建RILs家系群体, 结合10K SNP芯片结果和2年表型数据进行QTL定位验证, 检测到6个主效QTL, 分别位于6条染色体上。两次初定位结果相似, 说明实验结果可信, 可进行后续实验。通过连续自交及分析CM23和CC76双亲重测序数据开发CAPS和InDel标记构建了控制苞叶层数主效QTL-*qHN1* 杂合自交家系 (Heterogeneous Inbred Family, HIF), 对遗传效应进行了验证, 并将qHN-6-1进一步定位于1.3 MB区间之内。下步工作将进一步缩小定位区间, 找出定位区间内的差异表达基因, 并对基因功能进行预测注释, 找出可能参与苞叶发育相关的基因, 作为候选基因做进一步研究, 为玉米苞叶层数主效基因的克隆及分子育种奠定基础。

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Abstract 37

玉米抗灰斑病主效 QTL *qRglsB11* 的定位

胡 龙¹, 戴智康¹, 赖志兵^{1*}

¹作物遗传改良国家重点实验室, 华中农业大学, 武汉, 430070

*通讯作者: zhibing@mail.hzau.edu.cn

(submitted by 胡龙<hulong12138@163.com>)

摘要: 玉米灰斑病是由尾孢菌引起的一种真菌性叶部病害, 会对玉米的品质和产量造成损失。玉米感灰斑病后, 光合作用面积减少, 从而导致光合效率下降, 碳水化合物合成速率降低, 减产可达 10-40%。本研究中, 我们利用高抗自交系 B11 和高感自交系 B206 构建的 183 个 BC₁F₁ 家系群体进行玉米抗灰斑病表型鉴定; 同时, 对这 183 份材料进行简化基因组测序 (GBS) 获得相应的基因型, 结合表型和基因型数据进行 QTL 分析, 在 bin1.06 位置上鉴定到一个主效 QTL, 能解释 20-30% 的表型变异率。另外在 4 号染色体上还鉴定到一个微效 QTL, 能解释 5% 的表型变异率。利用 BC₂F₁ 群体鉴定到 QTL 区段 30 种不同重组单株, 通过自交获得相应的 BC₂F₂ 群体。计划 2019 年春播通过子代检测明确不同重组基因型的抗灰斑病表型, 以进一步缩小 QTL 区间。本研究为抗灰斑病 QTL 的克隆奠定了基础。

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Abstract 38

Cloning and function analysis of *ZmCTL* gene associated with seed development in maize

Mingjian Hu¹, Shuang Yang¹, Xiangbo Zhang¹, Bo Yang¹, Weibin Song, Haiming Zhao, Jinsheng Lai^{1*}

¹State Key Laboratory of Agrobiotechnology, National Maize Improvement Center of China, China Agricultural University, Beijing, 100089, China

Corresponding author: jlai@cau.edu.cn

(Submitted by 胡明建<humingjian0724@126.com>)

Abstract: Kernel development is very important for food safety. Improvement of grain weight is one of the main targets in maize breeding. However, the mechanisms of yield formation remain largely elusive. Here, we report the characterization of a defective kernel-3322 (dek-3322) mutant in maize. Map-based cloning of one mutant revealed an intron mutation in the choline transporter-like protein (CTL) which is highly conserved exist in eukaryotes, formation six types of alternative splicing. *ZmCTL* mutations cause developmental arrest in the embryo and endosperm, especially in the basal endosperm transfer layer (BETL). Loss function of *ZmCTL1* resulted in protein and soluble sugar excess phenotype. Our results indicated that *ZmCTL1* mutations alter the lipid component and reduce the number of plasmodesmata (PD), and also disrupt ionic homeostasis. A conserved serine phosphosites that required for the protein transport function was identified in maize. We also shown that *ZmCTL1* is constitutively expressed in a variety of tissues, but phosphorylation was found primary in seed, ear, and stem. The growth of *Escherichia coli*, *Agrobacterium tumefaciens* and Yeast were significantly repressed in the presence of *ZmCTL1*, which indicated that this protein is toxic to host cell.

Key words: maize, Kernel development, *ZmCTL1*, plasmodesmata, BETL

Funding acknowledgement: Chinese Postdoctoral Science Foundation

Abstract 39

Genome-wide association study deciphers the genetic architecture of carbohydrate partitioning in maize kernels

Shuting Hu^{1,2}, Yingni Xiao^{1,2}, Yingjia Han^{1,2}, Jiansheng Li^{1,2}, Xiaohong Yang^{1,2*}

¹State Key Laboratory of Plant Physiology and Biochemistry, China Agricultural University, Beijing 100193, China.

²National Maize Improvement Center of China, MOA Key Lab of Maize Biology, Beijing Key Laboratory of Crop Genetic Improvement, China Agricultural University, Beijing 100193, China.

*Corresponding author: yxiaohong@cau.edu.cn

(submitted by 胡书婷<hushutingcau@163.com>)

Abstract: Starch, protein, and oil are three major chemical compositions in maize kernels which have critical roles as the sources of food, feed, and industrial products. The carbohydrate in terms of sucrose is transported from photosynthetic tissues through the phloem to ear as the raw material to branch into starch, protein, and oil biosynthesis. To better understand the genetic basis of carbohydrate partitioning in maize kernels, genome-wide association studies on starch, protein, oil and three derivative traits were performed using 558,629 SNPs characterized in 508 diverse inbred lines. A total of 110 unique loci were significantly associated with six detected traits at $P < 1.08E-05$, with 53.15% loci co-localized with previously identified QTLs. The number of loci for each trait ranged from 6 to 71 and totally explained 25.49% to 90.83% of phenotypic variations. A loci-trait network showed that 54 loci were unique for one trait, 56 loci had pleiotropic effects on at least 2 traits. These results implied that kernel compositions might be genetically co-regulated. Expression QTL (eQTL) analysis defined 33 cis-eQTL and 8 trans-eQTL, suggesting that transcriptional regulation is one of the major molecular mechanisms for regulating kernel component variations. A gene ontology analysis revealed that 33 genes are linked to biological pathways involved in kernel compositions metabolism. These results will consequently provide the theoretical and technical basis for synergistically or directionally improving starch, protein and oil content.

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Abstract 40

Genome-wide proteomic profiling reveals the role of dominance protein expression in heterosis in immature maize ears

Xiaojiao Hu¹, Hongwu Wang¹, Kun Li, Yujin Wu, Zhifang Liu^{*} & Changling Huang^{*}
Institute of Crop Science, Chinese Academy of Agricultural Sciences, National Engineering Laboratory for Crop Molecular Breeding, Beijing, 100081, China.

^{*}Corresponding authors: huangchangling@caas.cn, liuzhifang@caas.cn

(submitted by 胡小娇<huxiaojiao@caas.cn>)

Abstract: Heterosis refers to the phenomenon in which hybrid progeny show superior performance relative to their parents. Early maize ear development shows strong heterosis in ear architecture traits and greatly affects grain yield. To explore the underlying molecular mechanisms, genome-wide proteomics of immature ears of maize hybrid ZD909 and its parents were analyzed using tandem mass tag (TMT) technology. A total of 9,713 proteins were identified in all three genotypes. Among them, 3,752 (38.6%) proteins were differentially expressed between ZD909 and its parents. Multiple modes of protein action were discovered in the hybrid, while dominance expression patterns accounted for 63.6% of the total differentially expressed proteins (DEPs). Protein pathway enrichment analysis revealed that high parent dominance proteins mainly participated in carbon metabolism and nitrogen assimilation processes. Our results suggested that the dominant expression of favorable alleles related to C/N metabolism in the hybrid may be essential for ZD909 ear growth and heterosis formation. Integrated analysis of proteomic and quantitative trait locus (QTL) data further support our DEP identification and provide useful information for the discovery of genes associated with ear development. Our study provides comprehensive insight into the molecular mechanisms underlying heterosis in immature maize ears from a proteomic perspective.

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Abstract 41

Assessment of phytochemical biosynthesis and accumulation in black sweet corn during kernel development

Xiaodan Hu¹, Xinbo Guo^{1,2*}, Rui-Hai Liu^{1,2*}

¹School of Food Science and Engineering, South China University of Technology, Guangzhou 510640, China

²Department of Food Science, Stocking Hall, Cornell University, Ithaca, NY 14853, USA

*Corresponding author: xbg720@gmail.com

(submitted by Xiaodan Hu<xiaodanhu1827@163.com>)

Abstract: Black sweet corn, a crop with abundant nutritional value and high processing value is widely studied in food industries. In this study, phytochemical profiles including phenolics, flavonoids, vitamin E and carotenoids along with their biosynthesis were analyzed in black sweet corn during kernel development.

Amongst phenolic acids, gallic acid and ferulic acid are the main components that increased with ripening. Cyanidin, pelargonidin and paeoniflorin were identified as the main anthocyanins with contents of around 4.3, 22.8 and 1.7 mg/100g FW, respectively when the corns matured. The total contents of phenolics and flavonoids at the mature stage is around three and four times more than the contents at pollination stage, which results in a grow of Oxygen Radical Absorbance Capacity (ORAC) and cell antioxidant activity (CAA) results. Through the dynamic accumulation process of vitamin E content during the development of sweet corn kernels, results showed an obvious increasing trend in α -T, α -T3, and γ -T3, in which γ -T3 has the highest accumulation of around 9.0 μ g/g FW. The gene expression levels of vitamin E biosynthesis showed that HPT and TC increased during grain development, while the expression level of γ -TMT gradually decreased and reached a stable level on 15 DAP (day after pollination) after pollination. The dominated components in carotenoids of black corn are lutein and zeaxanthin with proportions of 18.04% and 61.17%, respectively. 20 to 25 DAP was considered as the best stage from carotenoids accumulation, with an increase from 15.9 to 27.3 μ g/g FW. Expression of carotenoids genes showed that HYD, LCY and PSY make major influence on the accumulation of total carotenoid during kernel development.

玉米根系响应干旱胁迫的研究进展

黄帝^{1,2}，刘保兴^{1,2}，陈应龙^{2,3,4*}

¹ 西北农林科技大学林学院，陕西省杨凌，712100

² 西北农林科技大学黄土高原土壤侵蚀与旱地农业国家重点实验室，陕西省杨凌，712100

³ 中国科学院水土保持研究所，陕西杨凌，712100

⁴ 西澳大学农业研究所、农业与环境学院，西澳州珀斯，6009

*通讯作者：yinglongchen@hotmail.com

(submitted by 黄帝<2353187851@qq.com>)

摘要：根系是植物吸收土壤水分和养分的重要器官；根系的生长发育、构型及活力直接影响着作物的生长发育、营养状况和产量。玉米是世界上第三大农作物，我国玉米种植面积超过了水稻和小麦，成为第一大农作物，但玉米也是需水量最大和对水分胁迫较为敏感的作物，干旱是限制玉米产量的关键因子之一。目前，大多数学者从干旱程度、干旱时间、灌水方式以及种植方式等方面，开展了玉米根系对干旱响应的研究，通过分析作物地上部分的形态、生理、生物量和产量指标，以及根系形态学性状等，探讨玉米根系对干旱胁迫的适应性及响应策略。不同水分胁迫影响玉米根系长度、根系生物量、根表面积、根冠比等表型性状，使根系结构发生改变。干旱胁迫降低了玉米地上生物量、根直径、根条数和根总表面积，但促进了土壤上层根系的生长，促进了根冠比和根系活力。同时，水分胁迫会提高保护酶的活性，不同种类的保护性酶对干旱胁迫的敏感程度不同；根系可溶性蛋白含量随胁迫程度的增加而降低，可溶性糖含量逐渐上升。干旱胁迫下ABA作为根源信号参与调控气孔关闭，减少水分散失。玉米根系水流导度以及根系解剖学结构中通气组织的大小与密度与水分运输及抗旱能力有关。因此，玉米根系通过调整根系形态、解剖学结构及生理生化反应来协同减少水分胁迫造成的危害。本文通过对国内外玉米根系响应干旱的研究现状进行了总结，通过对玉米根系对干旱胁迫响应机制的系统研究，为农业生产提供理论依据。

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Abstract 43

The creation of high amylose maize material by CRISPR/Cas9 gene editing approach

Shengchan Huang^a Chongzhi Gong^a, Weiwei Qi^a and Rentao Song^{a,b*}

^aShanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China.

^bNational Maize Improvement Center of China, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

*Address for correspondence to rentaosong@cau.edu.cn.

(submitted by 黄圣禅<1633010867@qq.com>)

Abstract: Maize (*Zea mays*) is one of the most important food and feed crops in the world. Starch and storage proteins are the primary nutrient components of maize endosperm, with starch accounting for ~75% of the dry weight of maize kernels. The maize endosperm starch is composed of about 30% amylose and 70% amylopectin, and amylose is an important industrial raw material. In this study, CRISPR/Cas9 gene editing approach was used to knock-out the key gene, *Amylose extender1* (*Ae1*), which is involved in starch synthesis to increase the amylose content in maize endosperm. Six transgenic events have been obtained, among which two events have mutation in *Ae1*. Four editing targets in *Ae1* were selected, and the T0 generation showed that one transgenic event has a 22-bp deletion between the first target and the second target and a 1-bp insert at the fourth target, and another transgenic event has a 1-bp deletion at the fourth target. We then crossed T0 population to W22 background to generate T1 ears. We will then focus on phenotypic analysis and amylose content determination of T1 mutant kernels, and hopefully to obtain transgenic kernels containing high amylose starch content.

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Abstract 44

Maize *VKS1* is Essential for Early Endosperm Development by Regulating Mitosis and Cytokinesis

Yongcai Huang^{1, 2, 4}, Haihai Wang^{1, 4}, Xing Huang^{1, 2}, Qiong Wang¹, Jiechen Wang¹, Dong An³, Jiqin Li¹, Wenqin Wang³, Yongrui Wu^{1*}

¹National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology & Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200032, China

²University of the Chinese Academy of Sciences, Beijing 100049, China

³School of Agriculture and Biology, Shanghai Jiao Tong University, Shanghai 200240, China.

⁴These authors contributed equally to this work

*Corresponding Author: yrwu@sibs.ac.cn

(submitted by 黄永财 <ychuang@sibs.ac.cn>)

Abstract: Cell number is a critical factor that determines maize kernel size. Rapid mitotic divisions in early endosperm development produce most cells comprising the starchy endosperm; however, the mechanisms underlying early endosperm development remain largely unknown. We isolated a previously undescribed maize mutant that shows a varied-kernel-size phenotype (*vks1*). *Vks1* encodes ZmKIN11, which belongs to the kinesin-14 subfamily and is predominantly expressed in early endosperm development. VKS1 dynamically localizes to the nucleus and microtubules and plays key roles in free nuclei migration in the syncytium as well as in mitosis and cytokinesis in early mitotic divisions. Absence of VKS1 has relatively minor effects on plants but causes deformities in spindle assembly, sister chromatid separation and phragmoplast formation in early endosperm development, thereby resulting in reduced cell proliferation. Severities of aberrant mitosis and cytokinesis within individual *vks1* endosperms differ, thereby resulting in varied kernel sizes. Our discovery highlights VKS1 as a central regulator of mitosis in early maize endosperm development and provides a potential approach for future yield improvement.

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Abstract 45

The molecular mechanism of the maize PLATZ transcription factor family involved in maize endosperm development and storage filling

Jiechen Wang^{1#}, Chen Ji^{1,2#}, Yongrui Wu^{1*}

¹National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology & Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200032, China.

²University of the Chinese Academy of Sciences, Beijing 100049, China.

[#]These authors contributed equally to this work.

*Corresponding Author: yrwu@sibs.ac.cn

(submitted by 冀晨<jichen@sibs.ac.cn>)

Abstract: In our previous work, we had cloned a classical maize endosperm mutant *fl3*, which encodes a PLATZ family protein and exhibits severe defects in endosperm filling (2017, Plant Cell). PLATZ proteins are a novel class of plant-specific zinc-dependent DNA-binding proteins that are classified as transcription factors (TFs). Here, we identified and cloned 17 *PLATZ* genes in the maize (*Zea mays*) genome. According to phylogenetic analysis, *PLATZ4* were identified to be a homologous gene of *Fl3*, which has a more ubiquitous expression pattern than *Fl3* and also has high expression in the developing maize kernel. Another three *PLATZ*s, *PLATZ2*, *PLATZ10* and *PLATZ14*, were found to be expressed at the early stage of maize endosperm by RT-PCR and RNA-seq. These five *PLATZ*s potentially have functional redundancy and we created null mutants by CRISPR/Cas9 to each of them. Our recent work shows that *FL3* (*ZmPLATZ12*) interacts with *RPC53* and *TFC1*, two critical factors in the RNA polymerase III (RNAPIII) transcription complex. Using the yeast two-hybrid assay, we determined that seven other *PLATZ*s interacted with both *RPC53* and *TFC1*, whereas three had no protein-protein interaction with these two factors. The other six *PLATZ*s interacted with either *RPC53* or *TFC1*. These findings indicate that *ZmPLATZ* proteins are generally involved in the modulation of RNAPIII-mediated small non-coding RNA transcription and regulated the maize endosperm development and storage filling.

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Abstract 46

Positional cloning of *qDTA6* controlling flowering time in maize

Hong Jia¹, Dan Li¹, Cong Li¹, Guanghui Xu¹, Jinge Tian¹, Li Guo¹, Chenglong Wang¹, Lishuan Wu¹, Yameng Liang¹, Xuehan Wang¹, Weihao Wu¹, Jinliang Xia¹, Xu Han¹, Wenchao Qin¹, Feng Tian^{1,*}

¹National Maize Improvement Center of China, Key Laboratory of Biology and Genetic Improvement of Maize(MOA), Beijing Key Laboratory of Crop Genetic Improvement, China Agricultural University, Beijing 100193, China

*Corresponding author: ft55@cau.edu.cn

(Submitted by 贾宏<hongjia@cau.edu.cn>)

Abstract: Maize (*Zea mays* ssp. *mays*) was domesticated from its wild progenitor, teosinte (*Zea mays* ssp. *parviglumis*), a species that is restricted to tropical environments in southwestern Mexico. From its center of origin, maize adapted to a wide range of environments from latitude 58° north to 40° south and became one of the most widely planted crops in the world. Flowering time plays an important role in the local adaptation of plants. Although numerous loci affecting flowering time have been mapped in maize, their underlying molecular mechanisms and roles remain largely unknown. Using a large population of 866 maize-teosinte BC₂S₃ recombinant inbred lines (RILs), we previously performed quantitative trait locus (QTL) mapping for days to anthesis (DTA) and detected a QTL (*qDTA6*). The maize allele at this locus can accelerate flowering by ~1.2 days. To fine-map *qDTA6*, a heterogeneous inbred family (HIF) was selected and was used to generate a large near-isogenic line (NIL) population (n=1000). Through a recombinant-derived progeny testing strategy, *qDTA6* was delimited to a 150-kb region. In the fine mapping region, there were 10 annotated genes. Our findings provide important insights into the regulation of flowering time.

Funding acknowledgement: the National Key Research and Development Program of China (2016YFD0100404 and 2016YFD0100303); the National Natural Science Foundation of China (31771806 and 91535108)

Abstract 47

An evolutionary and expressional pattern of the bZIP family in maize

Liqliang Jia, Qiangfang Zhao, Shu Chen

Key laboratory of Tropical Fruit Biology (Ministry of Agriculture), South Subtropical Crop Research Institute, Chinese Academy of Tropical Agricultural science, Zhanjiang, 524091, China.

Abstract: Basic leucine zipper (bZIP) transcription factor gene family is one of the largest and most diverse families in plants. The bZIP TF family plays an important role in growth, development, and response to abiotic and biotic stresses. Nonetheless, knowledge concerning the specific expression patterns in maize and in response to various abiotic stress is limited. In this study, a total of 143 putative maize *ZmbZIP* genes were identified and renamed on the basis of their respective chromosome distribution. Phylogenetic analysis showed that *ZmbZIPs* were classified into 11 groups. The majority of *ZmbZIP* genes in the same subfamily shared similar gene structures and conserved motifs. The chromosome distribution and duplication analysis suggest that expansion of the maize bZIP transcription factor family was greatly contributed by the segmental/chromosomal duplications rather than tandem duplication. Expression data further revealed constitutive or organ-specific expression patterns of *ZmbZIP* genes in tissues. We also detected several key maize *bZIP* genes involved in abiotic or nitrogen stress responses suggested the possible multiple functions of these genes. Our results provide a valuable foundation for functional dissection of the different *ZmbZIP* homologs in maize and for molecular breeding studies of *bZIP* genes in maize.

Abstract 48

Detailed characterization of non-crossover recombination event spectrum by single-microspore sequencing in maize and rice

Ning Yang¹, Xiang Li¹, Chenglin Jiang¹, Lu Chen¹, Qinghua Zhang¹, Jianbing Yan^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: yjianbing@gmail.com

(submitted by 姜程淋<jiangcl@webmail.hzau.edu.cn>)

Abstract: Homologous recombination, resulting from meiotic crossovers (CO) and non-crossovers (NCO), is a fundamental biological phenomenon. Studies pay more attention to CO than NCO events due to technological limitations. Here, we isolated and deeply sequenced single microspore genomes of maize and rice from meiotic tetrads to profile NCO accurately. Under very stringent conditions, 176 CO and 1,127 NCO events were identified in four maize tetrads; 78 CO and 146 NCO events were identified in three rice tetrads. The 80% of maize (12/15) and 100% of rice (7/7) NCO events were validated through sequencing of PCR amplicons. The length of NCO tracts varied between 286 – 6,957bp with a median of 3,107bp in maize and 1,291 – 11,623bp with a median of 3,801bp in rice. The distribution of NCOs was not random and enriched in genic regions. A CCN repeat motif common to maize and rice, and a rice-specific A-rich motif were enriched in NCO tracts. CG and CHG methylation levels decreased around the CCN repeated motif in maize. GC-biased gene conversion (gBGC) probably contributed to the GC content bimodality at third codon positions (GC3) and to the 5'-3' gradient of GC content. These results provide evidence that NCO events, whose patterns are quite different from CO events, are widespread in the genome and may play an important role in gene and genome evolution.

Abstract 49

Characterization of proteome variation during modern maize breeding

Luguang Jiang^{#1}, Bo Li^{#1}, Shengxue Liu², Hongwei Wang³, Cuiping Li⁴, Shuhui Song⁴, Mary Beatty⁵, Gina Zastrow-Hayes⁵, Xiaohong Yang¹, Feng Qin^{*2} and Yan He^{*1}

¹MOE Key Laboratory of Crop Heterosis and Utilization, National Maize Improvement Center of China, China Agricultural University, Beijing 100094, China;

²College of Biological Sciences, China Agricultural University, Beijing 100094, China;

³Agricultural College, Hubei Collaborative Innovation Center for Grain Industry, Yangtze University, Hubei 434000, China;

⁴BIG Data Center, Beijing Institute of Genomics, Chinese Academy of Sciences, Beijing 100101, China;

⁵DuPont Pioneer, Johnston, IA 50131, USA

[#]These authors contributed equally to this work.

^{*}Corresponding authors: yh352@cau.edu.cn; qinfeng@cau.edu.cn

(submitted by 姜露光<lgjiang@cau.edu.cn>)

Abstract: The success of modern maize breeding has been demonstrated by remarkable increases in productivity with tremendous modification of agricultural phenotypes over the last century. Although the underlying genetic changes of the maize adaptation from tropical to temperate regions have been extensively studied, our knowledge is limited regarding the accordance of protein and mRNA expression levels accompanying such adaptation. Here we conducted an integrative analysis of proteomic and transcriptomic changes in a maize association panel. The minimum extent of correlation between protein and RNA levels suggests that variation in mRNA expression is often not indicative of protein expression at a population scale. This is corroborated by the observation that mRNA- and protein-based co-expression networks are relatively independent of each other, and many pQTLs arise without the presence of corresponding eQTLs. Importantly, compared to transcriptome, the subtypes categorized by the proteome show a markedly high accuracy to resemble the genomic subpopulation. These findings suggest that proteome evolved under a greater evolutionary constraint than transcriptome during maize adaptation from tropical to temperate regions. Overall, the integrated multi-omics analysis provides a functional context to interpret gene expression variation during modern maize breeding.

Funding acknowledgement: National Key Research and Development Program of China (2017YFD0101104); National Program on Key Basic Research Project of China (2014CB147300).

Abstract 50

Editing of 19-kD and 22-kD α -zein gene families by CRISPR/Cas9 gene editing approach in maize

Lifang Jin^a, Fei Peng^a, Weiwei Qi^a and Rentao Song^{a, b, *}

^aShanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China.

^bNational Maize Improvement Center of China, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

*Address for correspondence to rentaosong@cau.edu.cn.

(submitted by 金立方<1606953337@qq.com>)

Abstract: Zeins are the major storage protein in maize (*Zea mays*) kernel, in which, α -zeins are the major component which are responsible for the formation of protein body. Due to the presence of zeins, maize kernels display poor protein quality with a lack of essential amino acid. In this study, we generated knockout lines of maize α -zein gene families using Clustered regularly interspaced short palindromic repeats (CRISPR) /CRISPR-associated protein 9 (Cas9) gene editing approach. Two gRNAs were designed to introduce mutation in a conserved region targeting 19-kD α -zein genes or 22-kD α -zein genes, respectively. T1 population with W22 background were obtained, and were then self-crossed to generate T2 ears, which displayed variable opaque kernel phenotype. Using SDS-PAGE, HPLC and Western blot analysis, six events with significantly reduced level of α -zeins were observed. In addition, SDS-PAGE analysis indicated notably increase in non-zein content compared with W22 kernels. Our data showed that the lysine content in mature kernels was increased in the four selected events compared with W22 kernels. Our study provided a new approach for improvement of maize protein quality by CRISPR/Cas9.

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Abstract 51

Genome wide association study underlying the genetic basis of RFOs metabolic pathway in maize

Yuanyuan Chen¹, Min Jin¹, Shijuan Yan², Jianbing Yan^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

²Guangdong Academy of Agricultural Sciences, Guangzhou 510630, China

*Corresponding author: yjianbing@gmail.com

(submitted by 金敏<834245464@qq.com>)

Abstract: Raffinose family oligosaccharides (RFOs) that widely exist in plants are of great importance in plant abiotic stress defense, seed germination and seed longevity in storage. However, the regulation network of RFOs metabolism is still ambiguous. In the current study, we use the maize CUBIC (Complete-diallel plus Unbalanced Breeding-like Intercross) population, which consists of 1404 progenies descended from 24 Chinese elite inbred lines, to conduct genome wide association analysis for a set of primary metabolites in maize leaf and mature kernel. A total of 1095 loci were found to be significant associated with 57 metabolic variations in leaf and 539 significant associations detected for 55 metabolites in kernel. Interestingly, two major QTLs are constantly identified in maize leaf and kernel, controlling the variations of galactinol and raffinose. Within these two QTL regions, we find two reported genes (*ZmRS* and *ZmGols2*) encoding raffinose synthase and galactinol synthase respectively. Taking advantages of high resolution and strong mapping power of CUBIC population, we also obtained several novel QTLs across the whole genome, which provides new evidence and opportunity to elucidate the regulation network and catabolic process of RFOs in maize.

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Abstract 52

Specific expression of *GS5* gene in endosperm increases maize yield

Ruchang Jin, Dengxiang Du, Jinjie Guo, Xuxin Li, Fangdong zhang*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: fdzhang@mail.hzau.edu.cn

(submitted by 金如昌 <jrc@webmail.hzau.edu.cn >)

Abstract: *GS5* gene is located at rice chromosome 5 and encodes a serine carboxypeptidase. It is a positive regulator of grain width, grain weight and grain filling rate. The higher expression level is correlated with increasing rice size and yield. Under driven by maize endosperm specific promoter *ZmMRP-1*, 23 *GS5* transgenic maize lines were obtained by using *Agrobacterium tumefaciens*-mediated callus transformation and heat shock-induced marker elimination system. The analysis of PCR and Southern hybridization showed that the *GS5* gene was integrated into the transformed lines by single copy and inherited according to the Mendelian rule. After several successive generations of transgenic *GS5* lines, qRT-PCR analysis, cytological observation and starch content determination in maize kernel of T₃, T₄, T₅ materials were carried out. The results showed that *GS5* gene was specifically expressed in maize endosperm and starch content increased in grains. There was no significant difference of agronomic characters between transgenic plant lines and wild type, but the grain length and grain width were 5.80% and 9.59% higher than wild type, respectively. 100-grain weight increased by 7.88%-14.10%. By introducing a rice *GS5* gene specifically expressed in maize endosperm, the grain yield of maize was significantly increased, and these transgenic lines may be used in maize production for breeding high-yielding maize varieties. The introduction of exogenous genes into maize can effectively broaden the genetic germplasm of maize.

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Abstract 53

Stable-isotope labeling for quantitative peptidomics of maize treated by *Rhizoctoniasolani* stress and *Bipolaris maydis* stress

Gaorui Jin, Xiaojing Liu, Zhenzhen Zheng, Beijiu Cheng, Xin Zhang*

National Engineering Laboratory of Crop Stress Resistance Breeding, Anhui Agricultural University,
Hefei 230036, China

*Corresponding author: xinzhang@anau.edu.cn

(Submitted by 晋高锐<359262616@qq.com>)

Abstract: In the past few years, increasing interest has been directed to bioactive peptides of plant. Recent evidences of plant peptides functioning in the responses of plants to diverse environmental cues such as stress and infection by pathogens, suggests their potentially wide applicability in agrochemical and pharmaceutical industries. In the present study, a quantitative plant peptidomics strategy was established and used in the research of *Rhizoctoniasolani* and *Bipolaris maydis* infections of maize seedlings. After 5 days of infection, the maize plants showed obvious plaque. Peptides were extracted, labeled with TMT stable isotopic tags and identified by nanoLC/MS/MS. The results showed that 361 and 585 peptides were found in *Rhizoctoniasolani* stress and *Bipolaris maydis* stress, respectively, in which 120 and 176 peptides showed differential abundance compared to their corresponding no-treatment group. Among these peptides, 74 peptides up-regulated and 46 peptides down-regulated in *Rhizoctoniasolani* treatment, whereas 136 peptides up-regulated and 41 peptides down-regulated in *Bipolaris maydis* treatment. The biological functions of these varied peptides are being studied further. As a combination library of bioactive peptides, plant peptidomics provide a suitable starting point for future bioactivity studies.

Abstract 54

Different mechanisms regulate the expression of *TPS6* to improve the disease resistance and drought resistance in maize

Shengyan Li, Mengtao Li, DanWang, Hai Wang, Zhihong Lang^{*}

Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing 100081, China

^{*}Corresponding author: langzhihong@caas.cn

(submitted by 李圣彦<lishengyan@caas.cn>)

Abstract: Maize plants emit a complex blend of secondary metabolites (mainly terpenoids) to defend the biotic stress and abiotic stress. *TPS6* gene encoded a sesquiterpene synthase, which catalyzed β -macrocarpene and β -bisabolene. In our studies, *TPS6* gene expressed in B73 leaf induced by *Ustilago maydis* and in B73 root induced by ABA hormone which showed the expression regulation mechanisms of *TPS6* were different in leaf and root. The transgenic maize plants with *TPS6* gene overexpression and CRISPR-Cas9 knock-out were obtained. GC-MS showed the release of β -macrocarpene and β -bisabolene and accumulation of its derivatives Zealexins in *TPS6*-OX transgenic maize leaf and root, but all of them could not be detected in *TPS6*-KO transgenic maize. Transgenic maize plants were tested for disease resistance, insect resistance and drought resistance. Compared with control and *TPS6*-KO maize plants, *TPS6*-OX transgenic maize plants had significantly improved drought resistance and disease resistance, but had no significant inhibitory effect on the growth of Armyworm larvae. It suggested that *TPS6* and its secondary metabolites might perform different functions in the aerial and underground part of maize. The expression of *TPS6* was induced by pathogen in the aerial part of maize to improve the disease resistance. Meanwhile, the expression of *TPS6* was induced by ABA in underground part of maize to improve the drought resistance. We also found ABA contents in leaf and root of *TPS6*-OX transgenic maize were significantly higher than those of control and *TPS6*-KO plants, which indicated that *TPS6*, its secondary metabolites and ABA might work together to involve in maize defense response to abiotic stress, but the feedback regulation mechanism was not clear.

In order to explain the molecular mechanism of *TPS6* involved in maize defense response, the further works focus on the interaction mechanism between *TPS6* and ABA, the mechanism of pathogen-induced *TPS6* expression.

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Abstract 55

Genetic analysis of *cis*-regulatory imprint control at the *Floury3* locus

Qi Li^{1,2}, Xixi Zheng^{1,2}, Changsheng Li¹, Xiaomei Dong³, Jinsheng Lai³, Yongrui Wu^{1*}

¹National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology and Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200032, China

²University of the Chinese Academy of Sciences, Beijing 100049, China.

³State Key Laboratory of Agrobiotechnology and National Maize Improvement Center, Department of Plant Genetics and Breeding, China Agricultural University, Beijing 100094, China.

*Corresponding author: yrwu@sibs.ac.cn

(submitted by 黎颀<liqi01@sibs.ac.cn>)

Abstract: Endosperm development requires the female central cell to initiate whole-genome DNA demethylation for high expression of endosperm-specific genes, such as *zein* genes, *Opaque2* (*O2*), and *Floury3* (*Fl3*). In these processes, some genes exhibit differential expression between paternal and maternal alleles which represents the parent-of-origin effect of imprinted genes. Here, we analyzed how imprinted gene-neighboring region shaped their expression pattern. Utilizing ~500 inbred lines and the semi-dominant mutant *fl3-ref*, caused by a single base substitution in the imprinted *ZmPLATZ12* gene, we found that the introgression of Qi205 genetic background could alleviate dominant-negative effect caused by *fl3-ref* in the endosperm. Qi205 also modulates the differential gene expression and methylation patterns between the paternal *Fl3-Qi205* and maternal *fl3-ref* alleles. Genetic mapping revealed that the regulatory factor in the Qi205 genome is a single dominant locus and closely linked to *Fl3*. However, no sequence variation was detected between *Fl3-Qi205* and *Fl3-805A/B73/Mo17*, in the promoter, coding region, and terminal sequences. Our results suggest that a distal *cis*-regulatory element releases the epigenetic suppression of *Fl3-Qi205* allele in the developing endosperm when it is transmitted through the male. Further analysis of *Fl3-Qi205* will potentially uncover mechanisms of how non-coding sequences controls the imprinted gene expression in maize endosperm.

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Abstract 56

Flavonols affects drought tolerance through regulating stomatal movement and alleviating oxidative damage in maize seedlings

Baozhu Li, Guiling Sun, Ting Sun, Ruonan Fan, Shenglong Bai, Yanting Fan, Siyi Guo, Shiquan Huang, Xiaohong Zhu and Chun-Peng Song*

Key Laboratory of Plant Stress Biology, State Key Laboratory of Cotton Biology, School of Life Science, Henan University, Kaifeng 475004, China.

*Corresponding Author Email: songcp@henu.edu.cn

(submitted by Baozhu Li <lbzt01@henu.edu.cn >)

Abstract: The growth and yield of maize (*Zea mays L.*) are frequently threatened by drought stress and as a result, drought tolerance mechanism is a key problem in improving maize drought tolerance. In this study, *dos57* with lower leaf temperature, faster water loss rate, improved tolerance during drought stress, was screened out from our EMS B73 mutant library to study maize drought response mechanism. We generated the transcriptome data for *dos57* and wild type plants under drought stress. Functional GO categories analysis of differentially expressed genes (DEGs) obtained from transcriptome data showed that only a few genes related to stress response but key genes involved in flavonol biosynthesis was significantly enriched in *dos57*. The accumulation of flavonols in maize leaves and flavonols in guard cells was promoted by drought stress especially in *dos57*. The scavenging ability of *dos57* extracts to oxygen free radical was more effective than that of B73 after drought treatment. With the decrease of soil moisture content, guard cells in *dos57* accumulated more flavonols and less hydrogen peroxide than that in B73. Photosynthetic rate, stomatal conductance, transpiration rate and water use efficiency of *dos57* were significantly higher than the corresponding B73 during drought stress. In addition, *dos57* exhibited more biomass and root/shoot rate than B73 after drought. Taken together, *dos57* provides important materials to study drought response mechanism in maize seedlings, and flavonols regulate maize seedling drought tolerance by affecting stomatal movement and drought-induced oxidative damage.

Keywords: Flavonols, Drought stress, H₂O₂, Maize, Transcriptome, Guard cells

Funding acknowledgement: National Natural Science Foundation of China (31430061, U1604233).

Abstract 57

The *ZmbZIP22* transcription factor regulates 27-kD γ -zein gene transcription during maize endosperm development

Chaobin Li¹; Yihong Yue²; Hanjun Chen²; Weiwei Qi²; Rentao Song^{1,3 *}

¹ National Maize Improvement Center of China, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

² Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

³ Center for Crop Functional Genomics and Molecular Breeding, China Agricultural University, Beijing 100193, China

*Corresponding author: rentaosong@cau.edu.cn

(submitted by 李朝斌<chaobinli@126.com>)

Abstract: Zeins are the most abundant storage proteins in maize (*Zea mays*) kernels, thereby affecting the nutritional quality and texture of this crop. 27-kD γ -zein is highly expressed and plays a crucial role in protein body formation. Several transcription factors (TFs) (O2, PBF1, OHP1, and OHP2) regulate the expression of the 27-kD γ -zein gene, but the complexity of its transcriptional regulation is not fully understood. Here, using probe affinity purification and mass spectrometry analysis, we identified *ZmbZIP22*, a TF that binds to the 27-kD γ -zein promoter. *ZmbZIP22* is a bZIP-type TF that is specifically expressed in endosperm. *ZmbZIP22* bound directly to the ACAGCTCA box in the 27-kD γ -zein promoter and activated its expression in wild tobacco (*Nicotiana benthamiana*) cells. 27-kD γ -zein gene expression was significantly reduced in CRISPR/Cas9-generated *zmbzip22* mutants. ChIP-seq (chromatin immunoprecipitation coupled to high-throughput sequencing) confirmed that *ZmbZIP22* binds to the 27-kD γ -zein promoter in vivo and identified additional direct targets of *ZmbZIP22*. *ZmbZIP22* can interact with PBF1, OHP1, and OHP2, but not O2. Transactivation assays using various combinations of these TFs revealed multiple interaction modes for the transcriptional activity of the 27-kD γ -zein promoter. Therefore, *ZmbZIP22* regulates 27-kD γ -zein gene expression together with other known TFs.

Funding acknowledgement: National Natural Science Foundation of China (NSFC), National Key Research and Development Program of China, National Postdoctoral Program for Innovative Talents.

Abstract 58

Perturbation of the C terminus of *ZmSDW3* alters plant architecture in maize (*Zea mays* L.)

Hongchao Li¹, Lijing Wang², Meishan Liu¹, Zhaobin Dong¹, Qifang Li², Shulang Fei¹, Baoshen Liu^{2*}, Weiwei Jin^{1*}

¹National Maize Improvement Center, College of Agronomy and Biotechnology, China Agricultural University, Beijing 10093, China

²State Key Laboratory of Crop Biology, College of Agronomy, Shandong Agricultural University, Taian 271018, China

*Corresponding authors: weiweijin@cau.edu.cn; liubs@sdau.edu.cn

(Submitted by 李洪超 <hongchaoli2008@163.com>)

Abstract: Plant height and leaf angle are two crucial determinants of plant architecture in maize and are closely related to lodging resistance and canopy photosynthesis at high planting density. These two traits are primarily regulated by phytohormones, including gibberellins, brassinosteroids, and auxin. However, the role of ethylene in regulating plant architecture in maize, especially plant height and leaf angle, is unclear. Here, we characterized a semidominant maize mutant, *Semi-dwarf3* (*Sdw3*), which exhibits shorter stature and larger leaf angle than the wild type. Scanning electron microscopy observation showed that inhibition of longitudinal cell elongation in the internode and promotion in the auricle were mainly responsible for reduced plant height and enlarged leaf angle in *Sdw3*. Through map-based cloning, we identified a transposable element insertion in the candidate gene *ZmSDW3*, encoding an enzyme in ethylene biosynthesis. The transposon alters the C terminus of *ZmSDW3*. Transgenic analysis confirmed that the mutant *ZmSDW3* gene confers the phenotypes of *Sdw3*. Enzyme activity and protein degradation assays indicated that the altered C terminus of *ZmSDW3* increases this protein's stability but does not affect its catalytic activity. The ethylene content is significantly elevated in *Sdw3*, leading to reduced plant height and increased leaf angle. In addition, we demonstrated that *ZmSDW3* plays crucial roles in root development, flowering time, and leaf number, indicating that *ZmSDW3* is an important gene with pleiotropic effects during maize growth and development.

Funding acknowledgement: The National Natural Science Foundation of China (NSFC)

Abstract 59

***Mutator* 转座子介导的玉米籽粒发育突变体的基因克隆**

李见坤¹, 陈梦瑶¹, 苟明月^{1*}

¹ 河南农业大学农学院, 郑州, 450002

*通讯作者: 苟明月 <mingyuegou@163.com>

(submitted by 李见坤 <lijiankun198676@163.com>)

摘要: 玉米籽粒作为玉米产量的直接体现形式, 研究其形成和发育的分子机理对我们进一步解析、利用和改良玉米的籽粒性状具有重要的意义。尽管目前已经克隆出一些控制种子发育的基因, 但总体上来看, 人们对玉米籽粒发育的遗传基础缺乏深入和全面的认识。利用玉米特有的 *Mutator*(*Mu*)转座子, 我们已经收集鉴定了 50 余份由 *Mutator* 诱导、表型稳定、表现为单基因隐性遗传的籽粒发育突变体。表型主要包括: 小粒突变体, 空果皮突变体, 皱缩类突变体以及胚乳粉质突变体等。目前, 利用针对少数突变体的 *Mu* 标签法, 我们已经克隆了 *dek123* 突变体的候选基因, 下一步将利用针对 *Mutator* 的特异的测序技术, 完成其他突变体的基因克隆以及部分基因的功能分析。这些突变体的解析将为我们理解玉米籽粒发育的遗传基础和玉米分子设计育种提供强有力的理论支撑。

Abstract 60

***ZmRAP2.7*, an AP2 transcription factor, is involved in maize brace roots development via *miR156-miR172* pathway**

Jieping Li^{1,2}, Fanjun Chen¹, Yanqing Li¹, Pengcheng Li¹, Yuanqing Wang¹, Guohua Mi¹, Lixing Yuan^{1,*}

¹ Key Lab of Plant-Soil Interaction, MOE, College Resources and Environmental Sciences, China Agricultural University, Beijing, China, 100193

²Key Laboratory of Plant Stress Biology, State Key Laboratory of Cotton Biology, School of Life Sciences, Henan University, Kaifeng, Henan Province, China, 475001

* To whom correspondence should be addressed. E-mail: yuanlixing@cau.edu.cn

(submitted by 李结平 <ljp905@126.com>)

Abstract: The number of maize brace roots is most important root architecture trait influencing lodging, water and nutrient uptake. Understanding the genetic basis for brace roots development may facilitate efforts to improve important agronomic traits. In this study, we demonstrated that *ZmRAP2.7*, an AP2 transcription factor, is involved in the development of brace roots in maize. The express level of *ZmRAP2.7* was increased in the *Corngrass1*, which initialized more brace roots than wild type, and when the *ZmRAP2.7* was knocked out in *RAP2.7-Mu* mutant, the plant developed less brace roots than wild type. Meanwhile the *SNP1499*, located at the fifth exon of *ZmRAP2.7* gene among maize panel AM508 was association with the brace roots number. This study linked the *miR156-miR172* pathway to the development of brace roots in maize. *ZmRAP2.7* would be used to create molecular marker for root breeding in the future to improve the root system architecture and enhance the ability of avoiding the biological and non-biological stress in maize.

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Abstract 61

玉米快速叶绿素荧光参数与叶片表型相关性状的 QTL 分析

李露露, 李志鹏, 刘欢欢, 邓德祥*, 印志同*

扬州大学农学院, 扬州, 225009

*通讯作者: ztyin@yzu.edu.cn; dxdeng@yzu.edu.cn

Submitted by 李露露 lululi_yz@163.com

摘要: 叶片是玉米进行光合作用的主要器官, 叶片的形状、大小、夹角等表型可能会影响玉米的光合作用。快速叶绿素荧光参数可以灵敏反应植物光合作用时光能的转化利用率。本研究以 210 份 RA×郑 58 重组自交系为试验材料, 在三个大田种植条件下, 定位玉米快速叶绿素荧光参数 (ABS/CS₀, DI₀/CS₀, TR₀/CS₀, ET₀/CS₀, RC/CS₀, ET₀/TR₀, PI_{cs})、叶长、叶宽、叶夹角等性状的 QTL, 以期为玉米高光效育种和相关基因挖掘提供参考。三个环境共检测到 55 个和快速叶绿素荧光参数相关的 QTL, LOD 值为 2.52–6.72; 检测到 39 个控制叶片表型的 QTL, LOD 值为 2.54–10.85。上述 QTL 解释的表型变异为 4.62%–62.8%, 其中有 13 个叶绿素荧光参数相关 QTL 解释的遗传变异超过 10%, 12 个叶片表型相关性状的 QTL 解释的遗传变异超过 10%。另外发现, 叶绿素荧光参数相关 QTL 和叶片表型性状相关 QTL 在玉米 1、8 号染色体上富集, 且有多重叠的 QTL 区间。

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Abstract 62

***KERNEL NUMBER PER ROW6* encodes a serine/threonine protein kinase to regulate grain yield by phosphorylating an ARF-GAPase activating protein in maize**

Haitao Jia ^{a,§}, Manfei Li ^{a,§}, Weiya Li ^b, Lei Liu ^a, Yinan Jian ^a, Qiuli Zhu ^a, Wei Sun ^a, Zhixing Yang ^b, Xiaomeng Shen ^a, Qiang Ning ^a, Yanfang Du ^a, Jianbing Yan ^a, David Jackson ^{a,c}, XiaohongYang ^b, Zuxin Zhang ^{a,1}

^a National Key Laboratory of Crop Genetic Improvement, College of Plant Science and Technology, Huazhong Agricultural University, Wuhan 430070, P.R. China

^b State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center of China, MOA Key Lab of Maize Biology, Beijing Key Lab of Crop Genetic Improvement, China Agricultural University, Beijing 100193, P.R. China

^c Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, NY 11724, USA.

[§] These authors have equal contribution to the article

¹ Corresponding authors: Zuxin Zhang, zuxinzhang@mail.hzau.edu.cn

(submitted by 李曼菲<limanfei@webmail.hzau.edu.cn>)

Abstract: We establish that *KERNEL NUMBER PER ROW6* (*KNR6*), encodes a serine/threonine protein kinase which controls maize grain yield by determining pistillate floret number, ear length and kernel number per row through fine-mapping, association mapping, expression analysis and transgenic validation. Further investigation identified a Harbinger-like element (*TE*) presence/absence variation (TE-PAV) in the 5'-untranslated region of *KNR6* as the causal variant, with a strong effect on grain yield. The Harbinger-like *TE* regulated *KNR6* expression by functioning as repressor and/or altering epigenetic status of *KNR6* upstream region. *KNR6* showed protein kinase activity, and phosphorylated the ARF-GTPase activating protein (AGAP) by interacting with AGAP. The *AGAP* knockout lines showed reduced ear length and kernel number. Therefore, we propose that *KNR6* regulates floral number and grain yield through influencing AGAP phosphorylation to modulate auxin dependent ear inflorescence development, and provides a potential application to enhance grain yield of maize hybrids.

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Abstract 63

Different mechanisms regulate the expression of *TPS6* to improve the disease resistance and drought resistance in maize

Shengyan Li, Mengtao Li, DanWang, Hai Wang, Zhihong Lang^{*}

Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing 100081, China

^{*}Corresponding author: langzhihong@caas.cn

(submitted by 李圣彦<lishengyan@caas.cn>)

Abstract: Maize plants emit a complex blend of secondary metabolites (mainly terpenoids) to defend the biotic stress and abiotic stress. *TPS6* gene encoded a sesquiterpene synthase, which catalyzed β -macrocarpene and β -bisabolene. In our studies, *TPS6* gene expressed in B73 leaf induced by *Ustilago maydis* and in B73 root induced by ABA hormone which showed the expression regulation mechanisms of *TPS6* were different in leaf and root. The transgenic maize plants with *TPS6* gene overexpression and CRISPR-Cas9 knock-out were obtained. GC-MS showed the release of β -macrocarpene and β -bisabolene and accumulation of its derivatives Zealexins in *TPS6*-OX transgenic maize leaf and root, but all of them could not be detected in *TPS6*-KO transgenic maize. Transgenic maize plants were tested for disease resistance, insect resistance and drought resistance. Compared with control and *TPS6*-KO maize plants, *TPS6*-OX transgenic maize plants had significantly improved drought resistance and disease resistance, but had no significant inhibitory effect on the growth of Armyworm larvae. It suggested that *TPS6* and its secondary metabolites might perform different functions in the aerial and underground part of maize. The expression of *TPS6* was induced by pathogen in the aerial part of maize to improve the disease resistance. Meanwhile, the expression of *TPS6* was induced by ABA in underground part of maize to improve the drought resistance. We also found ABA contents in leaf and root of *TPS6*-OX transgenic maize were significantly higher than those of control and *TPS6*-KO plants, which indicated that *TPS6*, its secondary metabolites and ABA might work together to involve in maize defense response to abiotic stress, but the feedback regulation mechanism was not clear.

In order to explain the molecular mechanism of *TPS6* involved in maize defense response, the further works focus on the interaction mechanism between *TPS6* and ABA, the mechanism of pathogen-induced *TPS6* expression.

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Abstract 64

Combining genome-wide selected region sweep and association mapping to identify ear appearance genes in maize

Ting Li^{12#}, Jianzhou Qu^{12#}, Xiaokang Tian¹², Ningning Wei¹², Yonghui Lao¹², Yahui Wang¹², Yinchuan Hao¹², Xinghua Zhang¹², Jiquan Xue^{12*}, Shutu Xu^{12*}

¹The key Laboratory of Biology and Genetics Improvement of Maize in Arid Area of Northwest Region, Ministry of Agriculture, College of Agronomy, Northwest A&F University, Yangling 712100, Shaanxi, China

²Maize Engineering Technology Research Centre of Shaanxi Province, Yangling 712100, Shaanxi, China

*Corresponding author: xjq2934@163.com; shutuxu@nwfau.edu.cn

#These authors contributed equally to this work.

(submitted by 李婷<ltstime@163.com>)

Abstract: In maize, which was hybrid in actual production, how to improve the parents was the key cycle during breeding. And human selection made an important role during species improvement. So it is important to elucidate the impact of human selection on breeding. Herein, 221 inbred lines selected from two heterosis group named Shaan A and Shaan B were sequencing by Genome based sequencing for dissecting genome change under different selection pressure. Meanwhile, using within 121 inbred lines, genome wide association analysis was carried on for ear appearance related traits which were significant differential between the inbred lines from two heterosis groups. In a genome-wide scan, the inbred lines from Shaan A group and Shaan B group have obvious population divergences and different selection pressure distributed in 309 regions with 636 genes. Furthermore, co-expression network analysis and functional enrichment analysis show that these selected genes focus on regulating growth and development. Finally, combining the genome wide analysis of the differential ear related traits, four associated genes co-located in the related selected regions was high linkage disequilibrium with each other. Moreover, the atlas of spatiotemporal patterns of four key genes in support of future efforts for understanding underlying mechanisms that control ear length and fruit length. These results showed these four gene might be working for ear appearance by joint action. In conclusion, this study expounds a novel insight into dissecting complex traits using breeding materials and the finding about ear length, fruit length, barren tip length and setting rate will accelerate future efforts aimed at crop improvement.

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Abstract 65

在玉米籽粒中合成长链多不饱和脂肪酸EPA和DHA的研究进展

李文宗¹, 王磊^{1*}

¹中国农业科学院生物技术研究所, 北京, 10081

*通讯作者: wanglei01@caas.cn

(submitted by 李文宗<lwzm1010@163.com>)

摘要: 长链多不饱和脂肪酸(long chain polyunsaturated fatty acid, LCPUFA)对维持人体健康具有重要作用, 对其需求逐年增加, 但是由于环境污染与渔业资源的下滑, 有限的鱼油资源越来越不能满足人们需求。运用现代生物技术人们已相继分离了多个 LCPUFA 合成相关基因, 并阐明了多条 LCPUFA 合成代谢途径。玉米作为重要的粮食作物, 玉米油中的不饱和脂肪酸含量高达 80%~85%, 本研究利用玉米胚乳特异性启动子并筛选克隆六个不同的 LCPUFA 合成通路基因, 利用基因工程技术在玉米籽粒中合成 LCPUFA, 其中 EPA 的含量最高能达到 5.92%, DHA 含量最高能达到 4.14%。

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Abstract 66

一份新的玉米矮秆突变体的遗传分析与图位克隆

李 杨¹, 周 阳¹, 李晓峰¹, 何如钰¹, 张 萍¹, 唐祈林^{1*}

¹ 四川农业大学玉米研究所, 成都, 611130

*通讯作者: tangqilin71@163.com

(submitted by 李 杨<Yang_Li89@yeah.net>)

摘要: 矮秆突变体对于阐明植物生长与发育机制非常重要。矮秆突变体 d024 是经远缘杂交诱导得到的一个矮秆突变体。对突变体 d024 的研究结果表明: 该突变体 d024 株高 90.05cm, 穗位高 35.75cm、平均节长 6.98cm, 植株整体协调; 将 d024 与株高不同的 3 个自交系分别构建 F1、BC 和 F2 群体, 遗传分析表明该矮秆性状受 1 对隐性核基因控制; 以 d024/SW1611-F2 为定位群体, 采用集团分离分析法(BSA), 利用分子标记将矮秆基因初步定位于三号染色体短臂 3.02; 对突变体 d024 外施激素, 经成组数据分析表明, 该突变体对 (GA₃、IAA、BR) 均不敏感, 其与在三号染色体上已克隆的矮秆基因不同; 进一步将矮秆基因 d024 定位于三号染色体分子标记 Chr3-558 和 Chr3-584 之间, 遗传距离分别为 1.87cM 和 3.52cM, 物理距离为 266.62kb; 生物信息学分析显示该区段包含 9 个候选基因。进一步对候选基因进行克隆将为作物育种改良, 培育理想株型提供新的种质资源, 也为矮秆突变体的创制提供新的途径。

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Abstract 67

Serendipitous treasures: the maize transposable elements *Dotted*, *Mrh* and more

Jingsheng Tan¹, Guantao Zhao¹, Chunsheng Cong¹, Chuxi Li, Dooner Hugo K.², Yubin Li^{1,*}

¹ Biotechnology Research Institute, Chinese Academy of Agricultural Sciences

² Waksman Institute of Microbiology, Rutgers, the State University of New Jersey

*Corresponding author: liyubin@caas.cn

(Submitted by 李玉斌<liyubin@caas.cn>)

Abstract: Transposable elements (TEs) are the major components of most sequenced genomes and influential players in genome structure variation and evolution. Over 90% of the maize genomic DNA sequences are TEs and TE-derivatives. Among them, various families of class II transposons (DNA transposons) play a major role in shaping the dynamic genome. Most of them are nonautonomous driven by autonomous transposons whose presence and interactions are usually revealed genetically prior to their molecular structure characterization.

Dotted (*Dt*) was the first maize element characterized genetically as causing mutations at another locus, long before its recognition as a TE. We are attempting to isolate it because of its enormous historic importance in the development of the concept of controlling elements. From a segregating population of the *Dt/al-rDt* two-element system, we have cloned sequences amplified with primers based on the terminal/sub-terminal regions of *rDt*. Sequence analysis revealed the presence of candidate *Dt* elements encoding a conserved *hAT* family dimerization domain-containing protein. We are in the process of identifying which of these corresponds to Rhoades' classic *Dt* element and revealing their distinctive genetic behavior.

The autonomous transposon *Mrh* is known genetically to regulate its receptor element *rMrh* at the *Al* locus. The 80-bp terminal inverted repeats of *rMrh* are 70% identical with *Jittery*'s over the first 50 bp. We have amplified *Mrh*-related sequences using primers based on the *rMrh* TIRs. Sequence analysis indicates a high conservation of the encoded *Mrh* transposase to the known JITA transposase of *Jittery*, the second cloned autonomous element of Mutator superfamily. We have examined the possible genetic interaction between *Jittery* and *Mrh* and have shown that *Jittery* is able to trans-activate the *rMrh* element at the *Al* locus, as *Mrh* does. In addition, of serendipity, we have found out the second autonomous TE that seems able to trans-activate the transposition. We have named this novel element as *SMT* (Siblings of *Mrh* Transposon). Furthermore, we have also identified another closely related putative autonomous TE and named as *CMT* (cousins of *Mrh* Transposon), which is apparently another novel family of very-low-copy-number transposons in the maize genome. Our on-going efforts are to investigate how transposon specificities originate within a superfamily, an important issue in transposon evolution.

Funding acknowledgement: National Natural Science Foundation of China Grants (31871642 to Y.L.), CAAS Elite Youth Program Grant to Y.L.

Abstract 68

Genome-wide association study to dissect genetic basis of stay-green in maize under drought stress

Zhigang Li^{1,2,3}, Lili Liu^{1,2}, Xue Feng^{1,2}, Fang Sun^{1,2}, Huaiqing Hao^{1,3}, Haichun Jing^{1,2,3*}

¹Institute of Botany, Chinese Academy of Science, Beijing, 100093, China

²University of Chinese Academy of Sciences, Beijing, 100090, China

³Engineering Laboratory for Grass-based Livestock Husbandry, Chinese Academy of Science, Hulun Buir, 021000, China

*Corresponding author: hcjing@ibcas.ac.cn

(Submitted by 李志刚<zhigangli@ibcas.ac.cn>)

Abstract: Stay-green refers to leaf senescence delayed or rate reduced in the late stage of development, so as to maintain green features. Compared with the senescent varieties, the stay-green varieties had stronger resistance to drought stress. The research about it in maize is relatively lagging behind. Now agricultural production is facing more and more serious drought stress, so it is of great significance to study drought-tolerant and stay-green maize. In this work, we used 370 maize inbred lines to investigate the stay-green phenotype on 15, 25, 35, 45 days after drought treat. The data of drought and control group were analyzed by GWAS with 4.38 million high quality SNP markers, and the genetic loci affecting the target traits were mined to analyze the genetic basis of drought-tolerant and stay-green traits in maize. We identified 37 loci significantly associated at $p < 1.58E-8$ by FarmCPU model. Among them 6, 10, 12 and 9 were detected for SGLN, SGD, LWC and SWC. They can explain the phenotypic variance from 17.31% for SGLN to 61.53% for SGD. The results will provide valuable theoretical guidance to develop corresponding molecular markers for MAS breeding.

Funding acknowledgement: National Key Research and Development Program of China, Key Strategic Priority Research Program of the Chinese Academy of Sciences, National Transgenic Major Program of China

Abstract 69

Genetic determinant controlling maize Rubisco activase gene expression and a comparison with rice counterpart

Yu Zhang¹, Yong Zhou¹, Qian Sun¹, Dexiang Deng¹, Huanhuan Liu¹, Saihua Chen¹, Zhitong Yin^{1*}

¹Jiangsu Key Laboratory of Crop Genetics and Physiology/Co-Innovation Center for Modern Production Technology of Grain Crops/Key Laboratory of Plant Functional Genomics of the Ministry of Education/Jiangsu Key Laboratory of Crop Genomics and Molecular Breeding, Yangzhou University, Yangzhou 225009, China

*Corresponding author: ztyin@yzu.edu.cn.

Abstract: Rubisco activase (RCA) regulates the activity of Rubisco and is a key enzyme of photosynthesis. In this study, correlation analysis in approximately 200 maize inbred lines showed that the expression level of the maize RCA gene *ZmRCAβ* was correlated positively with grain yield. A genome-wide association study revealed both *cis*-expression quantitative trait loci (*cis*-eQTLs) and *trans*-eQTLs underlying the expression of *ZmRCAβ*, with the latter playing a more important role. Further allele mining and genetic transformation analysis showed that a 2-bp insertion and a 14-bp insertion in the promoter of *ZmRCAβ* conferred increased gene expression. Because rice is reported to have higher RCA gene expression than does maize, we subsequently compared the genetic factors underlying RCA gene expression between maize and rice. The promoter activity of the rice RCA gene was shown to be stronger than that of the maize RCA gene, suggesting that replacing the maize RCA gene promoter with that of the rice RCA gene would improve the expression of RCA in maize. Collectively, the above results increase understanding of the genetic mechanism that underlies RCA gene expression and identify new targets for both genetic engineering and selection for maize yield improvement.

Key words: Rubisco activase (RCA), photosynthesis, maize, rice, Quantitative trait locus (eQTL), promoter

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Abstract 70

The ceRNA-miRNA-target gene regulatory networks contribute to anther development in maize

Ziwen Li^{1,2#}, Xueli An^{1,2#}, Taotao Zhu¹, Tingwei Yan¹, Suowei Wu^{1,2}, Youhui Tian¹, Jinping Li², Xiangyuan Wan^{1,2*}

¹Biology and Agriculture Research Center, University of Science and Technology Beijing, Beijing 100024, China; ²Beijing Engineering Laboratory of Main Crop Bio-Tech Breeding, Beijing International Science and Technology Cooperation Base of Biotechnology Breeding, Beijing Solidwill Sci-Tech Co. Ltd., Beijing 100192, China. # Ziwen Li and Xueli An have contributed equally to this work.

* Corresponding author: wanxiangyuan@ustb.edu.cn

(submitted by 李紫文<liziwen@ustb.edu.cn>)

Abstract: The “competing endogenous RNA (ceRNA) hypothesis” has been recently proposed for a new type of gene regulatory model. Anther development is a crucial biological process in plant reproduction, and its gene regulatory network has been gradually revealed during the past two decades. However, it is still unknown whether ceRNAs contribute to anther development. We performed RNA and small RNA sequencing of anther tissues sampled at three developmental stages in two maize lines. A total of 28,233 stably transcribed loci, 61 known and 51 potentially novel microRNAs (miRNAs) were identified from the transcriptomes. We then reconstructed 79 ceRNA-miRNA-target gene regulatory networks consisting of 51 known miRNAs, 28 potentially novel miRNAs, 619 ceRNA-miRNA pairs, and 869 miRNA-target gene pairs. Several well studied miRNA-target gene pairs and six target genes associated with plant flower development were located in some networks. Our results provide an insight that the ceRNA-miRNA-target gene regulatory networks contribute to anther development in maize. Further functional studies on a number of ceRNAs, miRNAs, and target genes will facilitate our deep understanding on mechanism of anther development and sexual plant reproduction.

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Abstract 71

A sequence-indexed *Mutator* insertional library as an important resource for maize functional genomics study

Lei Liang¹, Ling Zhou², Yuanping Tang³, Teng Song¹, Niankui Li¹, Fan Feng³, Yafei Ma³, Dongsheng Yao³, Yang Feng¹, Han Zhao^{2*}, Rentao Song^{1*}

¹State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

²Provincial Key Laboratory of Agrobiolgy, Jiangsu Academy of Agricultural Sciences, Nanjing, China

³Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

* Corresponding Author: rentaosong@cau.edu.cn, zhaohan@jaas.ac.cn

(submitted by 梁磊 <lianglei@cau.edu.cn>)

Abstract: Sequence-indexed insertional libraries are critical resources for functional gene study in model plants, such as *Arabidopsis* and rice. In maize, the release of its reference genome greatly facilitated the study of functional genes, however the available sequence-indexed insertional library, such as the UniformMu, only covers relative small fraction (36%) of annotated genes in maize genome. In this study, a new sequence-indexed maize *Mutator* insertional library was generated through high-through-put sequencing of enriched *Mu*-tags from 1,000 *Mu* F₂ lines. Total 30,982 high quality insertion sites were obtained genome widely. These *Mu* insertions distributed on 14,486 genes, representing 32.7% of all annotated genes in the maize genome. About 49% (7,043 genes) of them harbored two or more insertions per gene. The distribution patterns of *Mu* insertions on maize genome and relative to gene structure were highly similar to the UniformMu library. Together with the UniformMu library, about 46% (20,316) maize genes were targeted, and 69% (14,011) of them contained two or more insertions per gene. The dataset and mutant seeds generated in this study will be available to the maize research community and will facilitate functional genomics studies.

Abstract 72

玉米/大豆间作下磷高效吸收的根系生物学机制研究进展

刘保兴^{1,2} 黄帝^{1,2} 陈应龙^{2,3,4,*}

¹西北农林科技大学林学院 陕西省杨凌 712100

²西北农林科技大学黄土高原土壤侵蚀与旱地农业国家重点实验室 陕西省杨凌 712100

³中国科学院水土保持研究所 陕西省杨凌 712100

⁴西澳大学农业研究所、农业与环境学院 西澳州珀斯 6009

*通讯作者: yinglongchen@hotmail.com

(submitted by 刘保兴<1916696310@qq.com>)

摘要: 大豆和玉米间作下, 两种作物根系所处的生态位不同, 根系交互分布在土壤中, 增加了作物对土壤磷的有效吸收面积; 根干重、根冠比、体积、表面积、根长和吸收面积都有不同程度的增加, 从而有利于作物对磷的吸收利用。间作条件下豆科植物根系会分泌大量的有机酸和磷酸酶类, 这些物质能够水解土壤中的有机磷, 促进豆科植物自身以及玉米对磷的吸收利用。大豆和玉米间作能够改变根际微生物群落的分布, 增加了丛枝菌根真菌的丰富度和多样性, 促进了土壤中难溶磷的活化, 提高了间作群体对磷的吸收利用。本文对玉米和大豆间作条件下磷高效吸收利用的根系生物学机制进行了综述, 以期为下一步的研究工作及生产实践提供理论依据。

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Abstract 73

Genome-wide analysis of *LAZ1* gene family from maize

Bing Liang Liu¹, Hao Qiang Yu¹, Qian Wen, Feng Ling Fu*, Wan Chen Li*

Maize Research Institute, Sichuan Agricultural University, Chengdu, Sichuan 611130, PR China

* Corresponding authors: ffl@sicau.edu.cn (F.L. Fu), aumdys@sicau.edu.cn (W. C. Li).

¹ Contributed equally

(submitted by 刘柄良<lbfree@foxmail.com>)

Abstract: Lazarus 1 (LAZ1) is a six transmembrane protein with DUF300 domain, and functions as organic solute transporter in vertebrates. No literature has been available on genome-wide identification of the LAZ1 gene family in plant. In this study, nine members of the *ZmLAZ1* gene family were identified from the maize genome, and characterized for their chromosomal locations, gene duplication, gene structure, *cis*-acting elements, and expression pattern in different organs and developmental stages. Except motif 1, which forms the DUF300 domain and functions as transmembrane organic solute transporter, was conserved among all the nine members, other six motifs were shared by the members within the same phylogenetic clade. Except *ZmLAZ1-6*, eight open reading frames were experimentally cloned and sequenced. A 75 bp alternative splicing was found from the open reading frame of *ZmLAZ1-9*. In bioinformatics prediction and laboratorial transient expression, the subcellular localization of the nine members included nucleus, cytoplasm, cytoplasm plasmalemma, as well as different organelles. The expression of the eight *ZmLAZ1* genes was detected in root and leaf by real-time quantitative PCR. Their relative expression levels showed abundant diversity among different organs and development stages, and in response to abscisic acid, simulative drought and NaCl treatments. This might be partially explained by the abundant diversity of *cis*-acting elements during their promoter regions. All these results demonstrated that the nine members of the *ZmLAZ1* protein family function as transmembrane organic solute transporters at different steps of different pathways in different organs, tissues or organelles at different development stages.

Funding acknowledgement: National Key Science and Technology Special Project (2016ZX08003-004) and Sichuan Science and Technology Program (2018JY0470).

Abstract 74

The study of the molecular mechanism of drought stress affecting the anthesis and silking interval

Boxin Liu^{1,2,4}, Bin Zhang^{1,4}, Feng Qin^{3*}

¹Key Laboratory of Plant Molecular Physiology, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China.

²Graduate University of the Chinese Academy of Sciences, Beijing 100093, China.

³State Key Laboratory of Plant Physiology and Biochemistry, College of Biological Sciences, China Agricultural University, Beijing 100193, China.

⁴These authors contributed equally to this work.

*Corresponding author: qinfeng@cau.edu.cn

(submitted by 刘博欣 <carrot881216@163.com>)

Abstract: Drought is a major environmental stress on maize production worldwide. The yield is greatly reduced especially when drought happens on flowering stage. As a kind of typical monoecious plant, maize produces male flowers (tassel) on the top of the plant and female flowers as a terminal branch (ear). However, environmental adversity often significantly extends development of the two unisexual flowers, known as anthesis and silking interval (ASI), which greatly hampers successful pollination and withdraw the yield. In this research, we investigated the maize B73 inbred line under four water regimes during the flowering stages in field conditions. The water deficit stress reduced plant height and decreased yield, while increased ASI, to different levels in relation to the extent of the stress. Ear development was clearly delayed by drought, whereas tassels were less affected. In order to understand how ears were impacted by the stress, we built up a transcriptome atlas of B73 ears from 5 to 50 mm under four water regimes. Genes specifically expressed in four different ear developmental stages and constitutively highly expressed genes throughout four stages were identified under well-water condition. To eliminate the distinctions implicated by the delayed development under the stresses, we compared the transcriptomes of ears with the same developmental stage, but not of the same growth date, under different water regimes. As a result, in the 50 mm ears a large number of genes were unraveled to be continuously down-regulated in accordance to the extent of stress. Especially the genes encoding expansins, XETs and aquaporins which are involved in cell expansion were significantly down-regulated in the 50 mm ears, which was probably implicated in a reduction in silk elongation under water deficit. Through candidate gene association study, an expansin gene, *GRMZM2G368886*, was identified as the most significantly correlative with ASI phenotype under drought stress ($p = 3.58e-04$). This study provides insights into understanding the mechanism how ASI occurs in maize under drought stress.

Funding acknowledgement: This research was supported by the National Natural Science Foundation of China (31625022) and National Key Research and Development Plan of China (2016YFD0100605).

Abstract 75

High frequent DNA rearrangement at the 27-kD γ -zein locus creates a superior *o2* modifier for Quality Protein Maize breeding

Hongjun Liu^{1,5}, Yongcai Huang^{2,5}, Xiaohan Li^{1,5}, Haihai Wang², Yahui Ding¹, Congbin Kang¹, Mingfei Sun¹, Fangyuan Li¹, Jiechen Wang², Yiting Deng², Xuerong Yang¹, Xing Huang², Lingling Yuan⁴, Qiong Wang², Dong An³, Wenqin Wang³, David R. Holding⁴, Yongrui Wu^{2*}

¹ State Key Laboratory of Crop Biology, College of Life Sciences, Shandong Agricultural University, Tai'an 271018, China

² National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology and Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai China 200032

³ School of Agriculture and Biology, Shanghai Jiao Tong University, Shanghai, China 200240

⁴ Department of Agronomy and Horticulture, Center for Plant Science Innovation, Beadle Center for Biotechnology, University of Nebraska, Lincoln, NE, USA 68588-0665

⁵ These authors contributed equally to this work.

*Correspondence should be addressed to Y. Wu (yrwu@sibs.ac.cn).

Abstract: The duplication at the 27-kD γ -zein locus is a major *o2* modifier (*q γ 27*) for endosperm modification in Quality Protein Maize (QPM). *q γ 27* is unstable and frequently produces single copies. Due to the lack of effective phenotypic or molecular markers, it was previously not possible to determine the frequency of germinal DNA rearrangement at this locus. We designed a polymorphic PCR marker that could discriminate the duplicated copies and generated a mutant QPM line (K0326Y-*Del*) which entirely lacks the *q γ 27* locus. When different maize lines were crossed to the null K0326Y-*Del* line, the frequency of DNA rearrangement could be determined because the PCR products arose entirely from the parents contributing the *q γ 27* allele. The frequency with which *q γ 27* rearranges to single copies from one generation to another is on the order of 10^{-3} and varies dramatically among different lines, with the highest in A188 and lowest in Mo17. It occurs significantly higher in male than female gametogenesis in all lines. Due to a relatively higher frequency in W22, the triplication of 27-kD γ -zein gene was identified in a small number of different UniformMu stocks (W22 background). The greatly enhanced amount of 27-kD γ -zein protein by the triplication allele is sufficient to confer vitreous kernels when α -zeins are suppressed by RNAi. Our results highlight a novel approach to directly determine the frequency of DNA rearrangements, in this case resulting in copy number variation at the 27-kD γ -zein locus. Furthermore, this provides a highly effective way to test suitable parents in QPM breeding.

Keywords: Quality Protein Maize (QPM); *o2* modifier; Copy number variation; DNA rearrangement

Abstract 76

Identification and high-resolution mapping of quantitative trait loci related to grain water content in maize

Jianju Liu¹, Hui Yu¹, Yuanliang Liu¹, Suining Deng¹, Qingcai Liu¹, Baoshen Liu^{2*}, Mingliang Xu^{1*}

¹State Key Laboratory of Plant Physiology and Biochemistry/College of Agronomy and Biotechnology/National Maize Improvement Center/Center for Crop Functional Genomics and Molecular Breeding, China Agricultural University, 2 West Yuanmingyuan Road, Beijing 100193, P. R. China

²College of Agronomy/State Key Laboratory of Crop Biology, Shandong Agricultural University, Taian 271018, P. R. China

*Corresponding author: Mingliang Xu, mxu@cau.edu.cn; Baoshen Liu, liubs@sdau.edu.cn

(submitted by 刘建菊<jianjuli23@163.com>)

Abstract: Grain water content (GWC) of maize is a critical parameter to mechanical harvesting. High GWC at harvest results in a high proportion of broken kernels, thereby reducing grain yield and quality. In the current study, a recombinant inbred line (RIL) population was built from the cross of 844 and 807, which differ in GWC at physiological maturity (PM). A genetic linkage map was constructed which consisted of 782 SNPs with a total genetic distance of 1522.48 cM. The RIL population was repeatedly evaluated for GWC at PM in three field trials. The initial QTL mapping revealed 31 QTL related to GWC and 17 related to grain dehydration rate (GDR). Seven GWC QTL were consistently detected in at least two of the three field trials, each of which could explain 6.92–24.78% of the total GWC variation. Similarly, one GDR QTL was consistently detected, accounting for 9.44–14.46% of the total GDR variation. Three major GWC QTL were found to overlap with GDR QTL in bins 1.05/06, 2.06/07, and 3.05. One of the GWC QTL, *qGwc1.1*, which has been consistently detected in three field trials, were selected for fine-mapping. With the four rounds of fine-mapping efforts, *qGwc1.1* was narrowed from a 27.22 Mb to a 2.05 Mb region flanked by markers SSR-75.2 and STS-77.2. *qGwc1.1* was found to act in a semi-dominant manner to reduce GWC at PM by 1.49–3.31%. Hence, genetic introgression of *qGwc1.1* into maize may prove beneficial for decreasing GWC at harvest, thereby minimizing the breakage of kernels.

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Abstract 77

Association mapping of stay-green traits for the panel adapted to China northern spring maize planting region

Lili Liu^{1,2}, Huaqing Hao^{1,3}, Xue Feng^{1,2}, Zhigang Li^{1,2,3}, Fang Sun^{1,2}, Haichun Jing^{1,2,3*}

¹Key Laboratory of Plant Resources, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China

²University of Chinese Academy of Sciences, Beijing 100049, China

³Engineering Laboratory for Grass-based Livestock Husbandry, Chinese Academy of Science, HuLunBuir, 021000, China

*Corresponding author: Haichun Jing@ibcas.ac.cn

(submitted by 刘丽丽 <liulili@ibcas.ac.cn>)

Abstract: Drought is one of the major constraints to maize yield production. Stay green, a crucial trait in maize genetic breeding, is closely related with drought tolerance and grain yields. Analysis the genetic bases of stay green is of great value to assist breeders to improve maize hybrid breeding. In the present study, a panel of 384 inbred lines from China northern spring maize planting region was genotyped using Maize SNP50 array, and two pools with extreme stay-green phenotypes from an F_{2:3} population were established and sequenced with average effective depth higher than 34.05 ×. The 384 inbred lines were subjected to analysis of population structure, genetic diversity and GWAS of stay green traits. Our results revealed that the panel can be divided into five hybrid subgroups, by SPT with the highest genetic diversity and PG with the highest stay green degree. By association study, 44 genetic loci for stay green trait were identified, among which 14 loci were overlapped with the reported QTLs for stay green and 19 were located in QTLs for maize grain yields. 204 genes tagged by the 44 loci were obtained and they have been involved in various metabolic pathways related with leaf senescence and abiotic stress. In addition, three candidate intervals of 7.87-12.89 Mb, 53.05-84.63 Mb, and 120.3-132.89 Mb on chromosome 2 were found significantly associated with stay green by BSA analysis of the F_{2:3} population. Two candidate loci identified in GWAS analysis also resided in the region of 53.05-84.63 Mb on chromosome 2. For these overlapped loci, more investigations are being carried out to validate the exact target genes and their functional analysis. Taken together, these results not only facilitate us further understanding the genetic basis for stay green but also provide new available targets for marker-assisted selection for maize in northern spring maize planting region.

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Abstract 78

Genetic Components Controlling Drought Responsiveness in Maize Seedlings

Shengxue Liu¹, Cuiping Li², Hongwei Wang³, Shuhui Wang¹, Xiaohong Yang¹, Jianbing Yan⁴, Bailin Li⁵, Shuhui Song², Feng Qin^{1*}

¹ State Key Laboratory of Plant Physiology and Biochemistry, College of Biological Sciences, China Agricultural University, Beijing 100193, China; ²BIG Data Center, Beijing Institute of Genomics, Chinese Academy of Sciences, Beijing 100101, China; ³Yangtze University, Jingzhou Hubei Province 434023, China; ⁴National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China; ⁵DuPont Pioneer, Wilmington, Delaware 19880

*Corresponding author: qinfeng@ibcas.ac.cn

(submitted by 刘升学<liushengxue1983@163.com>)

Abstract: Maize (*Zea mays*) responds to drought stress through a variety of physiological, biochemical, and transcriptional mechanisms. The altered gene expression is the key determinant of cellular phenotype in response to drought stress, which requires concerted action of both cis- and trans-regulatory cascade in genome. Although the genetic variants that influence gene expression levels have been identified by expression quantitative trait locus (eQTL) mapping in maize in tissue-dependent effects (kernel and leaves). There has been no systematic study to uncover genetic loci influence the dynamic and complex patterns of gene expression occurring under drought. Here we present an efficient experimental design to infer the genetic loci controlling the dynamics across three continuous drought gradient using 224 maize inbred lines, and apply it to characterize the local and distant regulatory effects on gene expression. The genome-wide association study (GWAS) identifies 73,573 eQTL, including 23,771 in WW (well watered), 22,945 in WS1 (moderate drought), 26,857 in WS2 (severe drought). Notably, 6,522 eQTLs were consistently identified across the three conditions, designated as statics-eQTLs, providing strong evidence that how these gene expressions are genetically regulated. In response to the stress, totally 5,492 dynamic-cis-eQTLs and 25,205 dynamic-trans-eQTLs were identified, capturing an unprecedented range of expression variation employed in drought response. Focusing on the TF mediated-stress responses, dynamic-eQTLs of TFs and their targets were collected, a hierarchy consisting 19 TF gene families was defined, generating a 3-hierarchical structure of regulatory network, regarding calcium signaling, transporter, oxidative detoxification, ABA response etc. Importantly, through Mendelian randomization analysis, we integrated eQTL, gene expression, and drought tolerance phenotype data, and identified 100 genes whose expression levels directly contributing to maize drought tolerance. It contains both candidate genes with predictable functions on drought response such as *ZmABA8ox1b*, *ZmP5CS*, *ZmPP2C*, but also genes with unclear roles in this regard. They can either serve as direct targets for genetic engineering for the trait improvement or important candidates for further functional investigations. Overall, this study unraveled the effect of local and distant genetic variation on the gene expression dynamics across the drought gradient, which provides insight into the genetic basis for natural variation in maize drought tolerance.

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Abstract 79

***Actin Depolymerizing Factor 5* was Identified Contribution to the Regulation of Drought Tolerance in Maize**

Liu Bojuan[#], Wang Nan[#], Zhou Yueheng, Cheng Ming, Wang Xiaonan, Chen Yong, Li Xinhai^{*}, Hao Zhuanfang^{*}

Institute of Crop Sciences, Chinese Academy of Agricultural Sciences, Beijing, 100081

[#]These authors contributed equally to this work; ^{*}Corresponding authors: Li X, linxinhai@caas.cn; Hao Z, haozhuanfang@163.com

(submitted by 郝转芳< haozhuanfang@163.com>)

Abstract: Actin depolymerizing factors (ADFs) play important role in the actin motion and conformation dynamics, which are considered sustaining cells free from loss of vitality after suffering biotic or abiotic stresses in eukaryotes. Many ADFs have been proved resistance to drought stress in *Arabidopsis* and rice, except in maize rarely reported. Accidentally, a candidate gene located on the peak of constitutive quantitative trait locus (QTL) from a meta-analysis of drought tolerance in maize, was also identified associated with grain yield under drought stress with genome-wide association mapping, which was named *ZmADF5*. Phylogenetic analysis indicated that *ZmADF5* was classified group IV commonly with *ADF5* and *ADF9* of other species. Overexpression of *ZmADF5* enhanced the drought tolerance in *Arabidopsis* with a significant increase of anthocyanin. Casein kinase and nfa class showed interactions with *ZmADF5* in yeast system. Fifty-five genes were identified contribution to high survival rate of transgenic *Arabidopsis* in improving drought tolerance of transgenic lines plants under drought stress.

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Abstract 80

Engineering of ‘Purple Maize’ with a multigene expression system

Xiaoqing Liu , Wenzhu Yang, Bona Mu, Suzhen Li, Ye Li, Yong Jiao, Xiaojin Zhou, Chunyi Zhang, Yunliu Fan and Rumei Chen*

Department of Crop Genomics & Genetic Improvement, Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, 12 ZhongGuanCun South Street, Beijing 100081, China

* Corresponding author: chenrumei@caas.cn

(Submitted by 柳小庆 liuxiaoqing01@caas.cn)

Abstract: Bidirectional promoters are identified in diverse organisms with widely varied genome sizes, including bacteria, yeast, mammals, and plants. However, little research has been done on any individual endogenous bidirectional promoter from plants. Here, we developed a simple and efficient multigene expression system based on an embryo-specific bidirectional promoter and 2A linker peptides. Eleven genes were successfully introduced into transgenic maize to rebuild the anthocyanin biosynthesis pathway in HiII embryos, resulting in an anthocyanin-rich purple embryo maize germplasm. Moreover, we also describe a strategy to modify the tissue specificity of a maize embryo-specific bidirectional promoter. Six types of cis-elements, i.e. RY repeats (R), GCN4 (G), the prolamin box (P), Skn-1 (S), and the ACGT and AACA (A) motifs, were collected and fused to PZmBD1 to generate eight chimeric putative bidirectional promoters. Qualitative and quantitative analysis of reporter genes driven by the promoters showed that two promoters exhibited high seed-specific bidirectional activity in maize transient and stable transformed systems. The stronger one was chosen and fused to the intergenic region of two gene clusters consisting of four anthocyanin biosynthesis-related genes (ZmBz1, ZmBz2, ZmC1 and ZmR2) and seven reporter genes, resulting in the first embryo and endosperm anthocyanin-rich purple maize. Anthocyanin analysis showed that the total anthocyanin content reaches 2,910mg kg⁻¹ DW in transgenic maize and cyanidin is the major anthocyanin in transgenic maize, as in natural varieties. Our results indicate that the multigene expression system, promoter modification strategy and these functionally characterized tissue-specific bidirectional promoters generated could be used for genetic research and development of plant biotechnology products. The anthocyanin-rich purple maize could provide economic natural colorants for the food and beverage industry, and valuable germplasm for developing anthocyanin-rich fresh corn.

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Abstract 81

玉米株高 *qPH10* 的 QTL 定位及候选基因预测

卢晓环¹, 周志强¹, 李海灵¹, 李冬冬², 刘文欣², 陈绍江², 郝转芳¹, 张德贵¹, 雍红军¹, 李新海¹, 李明顺^{1*}

¹ 中国农业科学院作科所, 北京, 100081

² 中国农业大学, 北京, 100193

*通讯作者: lims2013@126.com

(submitted by 卢晓环 <xiaohuanlu111@163.com>)

摘要: 玉米是我国重要的粮食、经济、饲料兼用型作物。玉米株高 (Plant height, PH) 作为生产中重要的农艺性状, 与玉米的抗倒伏性以及种植密度高度相关。本研究利用掖 478 为轮回亲本, 齐 319 为供体亲本构建的染色体片段代换系 (CSSLs) CL137 为父本与掖 478 杂交构建近等基因系 F₂ 分离群体, 根据齐 319、掖 478 重测序数据开发在双亲中具有多态性的 Indel 分子标记, 在两个环境中对控制玉米株高的 10 号染色体 QTL 进行定位, 2017 年的株高表型将 QTL 定位到标记 mk8-bnlg1655 之间, 位于 83.86-85.34 Mb (B73 RefGen_v3) 的 1.5Mb 区间, 表型贡献率为 7.57%; 2018 年株高表型将株高 QTL 定位到标记 mk5-bnlg1655 之间, 位于 82.76-85.34Mb 的 2.5 Mb 区间, 表型贡献率为 5.75%。同时检测发现, 该 QTL 主要以加性效应为主, 显性效应较小。通过对所定位的 QTL 重合区间内的基因进行功能注释, 预测可能控制株高的候选基因。本文研究结果可为后续精细定位第 10 号染色体株高 QTL 以及探索候选基因功能机制提供研究基础。

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Abstract 82

Identification and characterization of three phytosterol synthesis related genes in Maize

ShanShan Zhang^a, GeGe Lu^a, Chenchen Zhang, Tao Li, Wei Zhang, Beijiu Cheng, Xin Zhang^{*}

National Engineering Laboratory of Crop Stress Resistance Breeding, Anhui Agricultural University, Hefei 230036, China

^aThe first two authors contributed equally to this paper.

^{*}Corresponding authors: xinzhang@ahau.edu.cn

(submitted by 张晨晨<362491912@qq.com>)

Abstract: Phytosterols, the important composition element in plant cell bilayer membrane participate in the process of plant photosynthesis, reproductive, immune, and be response to external stress. They are also the important metabolite that could regulate plant senescence. Up to now, there are few reports about the genes that regulate the biosynthesis of phytosterols and gene mapping, which resulted in the lack of understanding of the genetic basis of natural variation. In the present study, the phytosterols in maize kernel of 244 maize inbred lines were extracted by the method of saponification, the composition and content were identified by GC-MS. According to the GWAS analysis with the phenotype data of these maize, nine SNPs and 32 candidate genes which associated with phytosterol content were identified. To investigate the relationship between gene expression level and sterol content, quantitative RT-PCR was performed with RNA preparations isolated from special maize kernels which had different sterol contents. Three genes were identified to be closely related to sterol content and probably participate in the synthesis of sterols. These three genes were transferred into *S. cerevisiae*, and the content of ergosterol in transgenic yeast were significantly increased. Further analysis showed that these proteins located to the Golgi, cell membrane and nucleus in protoplast of maize.

Abstract 83

ZmPP2Ca gene regulating drought tolerance in maize by alternative splicing

Fengzhong Lu¹, Haoqiang Yu¹, Yang Cao¹, Fengling Fu^{1*}, Wanchen Li^{*1}

¹Maize Research Institute, Sichuan Agricultural University, Chengdu, Sichuan, 611130, China

*Corresponding author: aumdys@sicau.edu.cn

(submitted by 路风中 <1550460423@qq.com>)

Abstract: Serine/threonine protein phosphatase 2C (PP2C), a key component in ABA signaling pathway, dephosphorylates various proteins. Alternative splicing (AS), re-processing precursor-mRNA at the post-transcription level, regulates gene expression, produces multiple variants. In our previous studies, we identified a maize PP2C clade B gene, named PP2Ca, with AS producing two splicing variants, the first intron of 213nt removed in the shorter spliced variant was retained in the longer variant, and they might relate to regulate drought tolerance in maize. We transformed these two variants to wild type *Arabidopsis* for functional validation and subcellular localization. The heterologous expression of the two variants increase the sensitivity of wild type *Arabidopsis* to drought stress. The result of subcellular localization show that the longer spliced variant was localized in the Nuclear and chloroplast, whereas the shorter spliced variant was localized in the Nuclear and cytoplasm. Therefore, we consider that these two spliced variants regulate maize drought tolerance with different mechanism by alternative splicing.

Abstract 84

基因索引的玉米突变体库助力功能基因组学研究

路小铎^{1,2} 刘基生¹ 任雯³ 杨群¹ 柴振光¹ 陈茹梅² 王磊² 赵军² 郎志宏² 王海洋²
范云六² 赵久然^{3*} 张春义^{2*}

¹齐鲁师范学院玉米分子育种研究院, 济南, 250200

²中国农业科学院生物技术研究所, 北京, 100081

³北京市农林科学院玉米研究中心, 北京, 100097

*通讯作者: maizezhao@126.com; zhangchunyi@caas.cn

(submitted by 路小铎<lu.xiaoduo@163.com>)

摘要: 玉米是世界上主要的粮食作物。B73参考基因组的公布开启了从全基因组水平进行玉米基因功能定性的新时期。在玉米大约39000个基因中, 仅有部分研究比较透彻, 绝大多数基因由于缺乏高通量的反向遗传学资源而进展缓慢。为了获得一套饱和的、基因索引的突变体集合; 我们利用EMS诱变B73的花粉, 然后结合外显子捕捉和高通量测序对1942份突变体进行变异挖掘, 总共检测到346,714个潜在导致蛋白变化(包括终止获得/丢失, 错义剪切, 起始获得/丢失和非同义变异)的CG>TA突变位点, 平均每个突变体携带178个变异。这些变异覆盖34,186个玉米基因, 占全部注释基因的86.9%, 平均每个基因10.1个突变。导致提前终止和错义剪切(剪切位点供体/受体变异)的CG>TA突变共有26,290个, 覆盖15,065个基因, 其中6,124个基因携带2个及以上该类型变异。为了方便广大科研人员查找感兴趣的EMS突变体, 我们构建了玉米EMS突变体数据库(<http://www.elabcaas.cn/memd/>)。该突变体库将大大加速玉米功能基因组学的进展。

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Abstract 85

Single gametophyte sequencing reveals that crossover events differ between sexes in maize

Cheng Luo¹, Xiang Li^{1*}, Qinghua Zhang¹, Jianbing Yan^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

* Corresponding author: lixiang1989@webmail.hzau.edu.cn (Xiang Li), yjianbing@mail.hzau.edu.cn (Jianbing Yan)

(submitted by 罗成<luochengxiansheng@webmail.hzau.edu.cn>)

Abstract: Meiotic crossover (CO) plays a key role in producing gametophytes and generating genetic variation. The patterns of CO production differ inter- and intra-species, as well as between sexes. However, sex-specific patterns of CO production have not been accurately profiled independently of genetic backgrounds in maize. Here, we develop a method to isolate single female gametophyte for genomes sequencing in maize. We show that more COs are observed in male (19.3 per microspore) than in female (12.4 per embryo sac). Based on Beam-Film model, the more designated class I and II COs are identified in male than in female. In addition, CO maturation inefficiency (CMI) is detected in some genetic backgrounds, suggesting that maize may be an ideal model for dissecting CMI. This research provides insights toward understanding the molecular mechanism of CO production between sexes and may help to improve maize breeding efficiency through paternal selection.

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Abstract 86

Molecular dissection of maize seedling salt tolerance using a genome-wide association analysis method

Meijie Luo¹, Yunxia Zhang¹, Panpan Zhang¹, Kuan Chen¹, Wei Song¹, Xiaqing Wang¹, Baishan Lu¹, Yanxin Zhao^{1*}, Jiuran Zhao^{1*}

¹Beijing Key Laboratory of Maize DNA Fingerprinting and Molecular Breeding, Maize Research Center, Beijing Academy of Agriculture and Forestry Sciences (BAAFS), Beijing 100097, China

*Corresponding author: rentlang2003@163.com; maizezhao@126.com

(submitted by 骆美洁 <mjl108@163.com>)

Abstract: Soil salinity is a major devastating abiotic factor that affects maize growth and productivity worldwide. However, knowledge of the molecular mechanisms of responses to salt stress in maize remains limited. To elucidate the genetic basis of salt tolerance traits, a genome-wide association study was performed on 348 maize inbred lines under normal and salt-stressed conditions. The phenotypic data for 27 traits revealed coefficients of variation > 25%. In total, 149 significant SNPs were identified, explaining 6.6%–11.2% of the phenotypic variation for each SNP. Of the 104 identified quantitative trait loci (QTLs), 83 were related to salt tolerance and 21 to normal traits. Additionally, 13 QTL were simultaneously identified by two to five traits. Ten and six QTLs controlling salt tolerance traits and root growth were co-localized with reported QTL intervals from linkage populations, respectively. Based on functional annotations, 16 candidate genes for salt tolerance were predicted. Two genes were identified as known maize salt response genes, one of which, *plasma membrane protein 3 (PMP3, GRMZM2G477325)* was characterized to involve in ion homeostasis. One of the candidate genes, *GRMZM2G071119*, were located in a QTL harboring 11 peak SNPs and its *Arabidopsis* homolog is responsible for chloride transmembrane transport, making it a promising target for further functional investigations. These results aid in elucidating the genetic variation in salt tolerance and provide novel loci for the genetic improvement of maize with salt tolerance.

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Abstract 87

***ZmMs7* functions as a transcriptional activator to regulate anther and pollen development in maize**

Biao Ma^{1,2}, Youhui Tian¹, Quancan Hou^{1,2}, Zhenying Dong^{1,2}, Xueli An^{1,2}, Ziwen Li^{1,2}, Taotao Zhu^{1,2}, Simiao Zhang¹, Wei Zhao¹, Chang Liu¹, Suowei Wu^{1,2}, Ke Xie^{1,2}, Jinping Li², Xiangyuan Wan^{1,2*}

¹Biology and Agriculture Research Center, University of Science and Technology Beijing, Beijing 100024, China; ²Beijing Engineering Laboratory of Main Crop Bio-Tech Breeding, Beijing International Science and Technology Cooperation Base of Biotechnology Breeding, Beijing Solidwill Sci-Tech Co. Ltd., Beijing 100192, China.

* Corresponding author: wanxiangyuan@ustb.edu.cn (Submitted by 马彪<mabiao@ustb.edu.cn>)

Abstract: Genic male-sterility (GMS) mutants are optimal materials for research on anther development and valuable for hybrid breeding and seed production. Our previous study (*PBJ*, 2018, 16: 459-471) revealed that maize *GMS* gene *ZmMs7* encodes a plant homeodomain (PHD)-finger transcription factor involved in maize anther and pollen development. *ZmMs7* is orthologous to rice *PTCI* and *Arabidopsis MSI*. In this study, we further explored the cytological and molecular mechanisms of *ZmMs7* gene regulating male fertility in maize. Cytological analyses showed that *ms7-6007* mutant displayed delayed tapetal degeneration, abnormal pollen exine formation and aborted microspore development, and ultimately led to complete male sterility. Quantitative RT-PCR analysis revealed that *ZmMs7* is specifically expressed in maize anthers at stages 8b (tetrad) and 9 (free haploid microspore), indicating that *ZmMs7* functions during post-meiosis stages of anther development. *ZmMs7* protein is predominantly localized to the nucleus as revealed by maize leaf protoplast transient assay. RNA-seq analysis indicated that 488 genes, which are mainly associated with tapetal development and pollen exine formation, had altered expression in *ms7-6007* mutant anthers. Transient dual-luciferase reporter assay using maize protoplast showed that *ZmMs7* has transcriptional activation activity. Yeast two-hybrid, CoIP and BiFC assays identified one bZIP and four NF-Y-type transcription factors that interact with *ZmMs7* protein, implying that *ZmMs7* may function together with a multi-protein transcription complex. Further studies are required to elucidate the molecular mechanism of transcriptional regulation of *ZmMs7*.

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Abstract 88

黑龙江省三、四积温带玉米新品种抗倒伏性比较研究

马德志, 杨德光*, 于乔乔, 孙玉珺, 吴玥, 张倩

东北农业大学农学院, 哈尔滨 150030

*通讯作者: deguangyang@sina.com

(submitted by 马德志<1353505920@qq.com>)

摘要: 将适合于黑龙江省东部湿润区第三、四积温带种植的 36 个玉米品种进行了高密植抗倒伏性评价, 试验对玉米的农艺性状、茎秆力学指标和产量进行聚类分析, 同时验证评价玉米抗倒性强弱的各项重要指标。结果表明, 通过系统聚类统计方法将品种分为三大类, 其中德美亚 3 号等 21 份品种为高度抗倒性(I)、禾田 6 号等 10 份品种为中度抗倒性(II)、德美亚 1 号等 5 份品种为低度抗倒性(III)。高度抗倒性品种的力学指标、单位茎长干物质和茎节直径均高于其他级别品种, 且这 3 个指标与茎秆抗倒伏指数呈极显著正相关, 可作为评价品种抗倒伏能力的重要指标。随着种植密度的增加, 不同级别品种的穗位高、重心高度和产量均增加, 单位茎长干物质、茎节直径和力学指标均下降, 其中 II 级品种抗倒性在增密后变化幅度不显著, 因此可考虑在高密度下种植。

项目基金: 国家重点研发计划 (2018YFD0300103)

Abstract 89

Resistance of Seed Internal Tissues to *Fusarium verticillioides* in Maize: Identification Methods, Resistant Lines, and Breeding Strategies

Peipei Ma¹, Zijian Zhou¹, Jingyang Gao¹, Yunxia Song¹, Kewei He¹, Cong Mu¹, Zhao Wang¹, Chaopei Dong¹, Yuna Chen¹, Jiafa Chen², Jianyu Wu^{12*}

¹ College of Agronomy, Synergetic Innovation Center of Henan Grain Crops and National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China.

² College of Life Sciences, Synergetic Innovation Center of Henan Grain Crops and National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China.

* Corresponding author: Dr. Jianyu Wu, E-mail: wujianyu40@126.com

(submitted by 马培培<2624634227@qq.com>)

Abstract: Seeds carrying fungi, both internally and externally, are an important factor restricting maize yield and quality. In this study, we compared the resistance responses of the internal and external tissues of maize (*Zea mays* L.) seeds from a resistant line and a susceptible line to *Fusarium verticillioides* by observing tissue morphology and quantifying the pathogen. Histological and morphological analyses revealed that the pathogens inside the seeds broke through the seed coat near the inoculation point and spread to the entire epidermis. In addition, the content of *F. verticillioides* was significantly higher in the susceptible line than in the resistant line after 5 days of culturing. We also found that the incidence of *F. verticillioides* in seeds was negatively correlated with seed germination rate. On this basis, we established an identification system for the resistance of maize seed internal tissues to *F. verticillioides* and identified 121 maize inbred lines. The proportions of highly resistant, resistant, moderately resistant, susceptible, and highly susceptible lines were 14.05%, 39.67%, 28.93%, 14.05%, and 3.31%, respectively. In addition, we found that the resistance of internal and external seed tissues to *F. verticillioides* was not consistent for most maize inbred lines. Notably, our findings showed that the internal tissues of seeds belonging to P group lines (Tropical/subtropical origin, multiple sources of resistance) were less resistant to *F. verticillioides* (only 38.7%) than other maize lines, although this group is widely used by Chinese breeders to improve disease resistance. The results of the present study improve our understanding of the pathogenesis and resistance mechanisms of seed internal tissues to fungi and offer new perspectives for maize breeders to improve disease resistance.

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Abstract 90

Integrate transcriptome and GWAS for functional characterization of long non-coding RNAs under deficient nitrogen in maize seedling

Peng Ma, Dan Liu, Ling Wu, Shibin Gao*

Maize Research Institute, Sichuan Agriculture University, Chengdu, China

*Corresponding author: shibingao@163.com

(submitted by 马鹏<mapeng561@163.com>)

Abstract: Nitrogen(N) is one of the most important components of nucleic acids and proteins which are necessary for plant growth and development. Long non-coding RNAs (lncRNAs) are a class of transcripts more than 200 nucleotides that are not translated into proteins. They have been reported to participate in regulation of plant biological processes and development. To greatly expand our understanding in regulatory mechanisms of lncRNAs as a kind of competing endogenous RNAs (ceRNA) in maize under nitrogen deficiency, lncRNAs and miRNAs libraries were constructed for deep sequencing of leaves and roots under low nitrogen and high nitrogen during seedling stage. 8836 mRNAs, 894 lncRNAs and 38 known miRNAs were identified. Co-expression network analysis showed that most of the low-nitrogen response lncRNAs were involved in abiotic stress responses, nitrogen metabolism and signal conduction process. In order to explore valuable candidate genes related to nitrogen stress, genome-wide association studies (GWAS) and published data which included a total of 40 consistently LN-responsive candidate genes were combined for analysis. Hundreds of lncRNAs contained trait-associated significant SNPs suggested to be considered as putative trait-related lncRNAs that probably contributed to development and related traits during the early stage of nitrogen deficiency in maize. Moreover, the expression profiling of selected N-responsive lncRNAs and target genes were validated by qRT-PCR. These results will provide new insights to illustrate the potential regulatory roles of lncRNAs in response to N stress.

Abstract 91

Characterization of a new maize high-lysine mutant

Xuexia Ma², Qiuzhen Tian¹, Hongyu Chen¹, Jihua Tang¹, Qinghua Yang¹ and Guifeng Wang^{1*}

¹ Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China;

² Shanghai Key Laboratory of Bio-Energy Crops, School of Life Sciences, Shanghai University, Shanghai 200444, China.

*Corresponding author: guifeng.wang@henau.edu.cn

(submitted by Xuexia Ma <1010663604@qq.com>)

Abstract: Maize kernel is nutritionally poor for monogastric animals as the lack of key essential amino acids, like lysine, tryptophan and methionine. A large class of kernel mutants with opaque or floury endosperm that could drastically improve protein quality. Here, we characterized a new maize opaque mutant 1754, of which the lysine and methionine contents increase 70% and 93%, respectively. The developing mutant kernels could be clearly distinguished as early as 10DAP from the self-crossed heterozygous plants, featured by the pale appearance. Paraffin sections showed that the development of embryo of the mutant was impaired and the starch granule and protein body in endosperm became smaller, relative to the wild type counterparts. The mutant 1754 gene was mapped into a physical interval of about 90kb covering 12 candidate genes, by analyzing an F2 population of 1212 individuals. DNA sequencing indicated that a G-to-A transition appears in exon of gene A, thus causing an amino acid substitution from glycine to glutamic acid. Transcriptomic data analysis revealed that the casual gene mainly affects starch and sucrose metabolic pathways, amino acid biosynthesis pathways and glycolysis pathways. This data provides a foundation for later functional analysis, which is of great significance for improving the protein quality of maize seeds.

Funding acknowledgement: National Natural Sciences Foundation of China (31771800, U1804235), Outstanding Youth Foundation of Henan Province (2018JQ0018), and Shanghai Pujiang Talent Program (17PJD014).

Abstract 92

Fine mapping of the main QTL of ear length in maize

Shujun Meng¹, Qiyue Wang¹, Jiong Wan¹, Wen Zhang¹, Shuanhong Ma¹, Dong Ding^{1*}, Jihua Tang^{1*}

¹State Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University.

*Co-corresponding authors: Jihua Tang (tangjihua1@163.com)

(submitted by 孟淑君18237116524@163.com>)

Abstract: Ear length is one of the most important traits of maize, which plays an important role in the formation of maize yield per single ear. In this study, a SSSL (single segment substitution lines) which contained one DNA fragment of Zong 3 in Xu 178 background was distinguished. Compared with the background material Xu 178, the ear length and kernel number per row of the SSSL were significantly increased ($P=4.44752E-14$). However, there was no significant difference in ear width, ear row number, grain length, grain width and grain thickness between Xu 178 and SSSL. The BC₁F₁ population was constructed by crossing with SSSL and Xu 178 and then backcrossing. The main QTL of maize ear length was fine mapped by fitting 418 InDel marks into 101 BC₁F₁ recombinant seedlings. Based on the B73 reference genome, the candidate gene was located in about 586 kb region of maize chromosome 10, where 12 candidate genes were predicted by map-based cloning.

Abstract 93

Gibberellins synthesis is involved in the reduction of cell flux and elemental growth rate in maize leaf under low nitrogen supply

Xiaohuan Mu^{1,2}, Qinwu Chen¹, Xiangyu Wu¹, Fanjun Chen¹, Lixing Yuan¹, Guohua Mi^{1*}

¹Department of Plant Nutrition, China Agricultural University, Beijing 100193, PR China

²Present address: The author is working in Mingyue Gou's group.

State Key Laboratory of Wheat and Maize Crop Science, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding author: miguohua@cau.edu.cn

(Submitted by Xiaohuan Mu<muxiaohuan1988@qq.com>)

Abstract: One strategy for plants to adapt to low nitrogen is to reduce shoot growth and allocate more nitrogen and carbon for root growth. The mechanism underlying the response of leaf growth to low-nitrogen remains unknown. In this study, we investigated cell division and elongation in maize leaf growth in response to low-nitrogen by using an integrated approach. Kinematic analysis revealed that low-nitrogen inhibited leaf elongation mainly by shortening length of division zone, reducing cell flux and elemental growth rate. Hormone analysis revealed that changes of gibberellins caused by low-nitrogen correlate with the observed changes in division zone size and elemental growth rate in response to low-nitrogen, suggesting role of gibberellin in low-nitrogen induced inhibition of leaf elongation. RNA-Seq identified that GA20ox4 (GRMZM2G060940), a key enzyme for synthesis of gibberellins, was down-regulated in both division and elongation zone of leaf under low-nitrogen supply. Furthermore, exogenous GA3 application on low-nitrogen plants restored leaf growth. However, application of gibberellin biosynthesis inhibitor reduced leaf growth. It is concluded that low-nitrogen reduces cell flux and elemental growth rate in maize leaf via reducing gibberellin synthesis. As a result, leaf elongation rate was slower and leaf area was smaller, freeing nitrogen for export to roots.

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Abstract 94

A novel protein is essential for mitochondrial function and seed development in maize

Jiacheng Ni², Yongyan Wang¹, Hongyu Chen¹, Jihua Tang¹ and Guifeng Wang^{1*}

¹ Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China;

² Shanghai Key Laboratory of Bio-Energy Crops, School of Life Sciences, Shanghai University, Shanghai 200444, China.

*Corresponding author: guifeng.wang@henau.edu.cn

(submitted by Jiacheng Ni <867373597@qq.com>)

Abstract: To identify new factors implicated in the accumulation of storage substance and seed development, we dissected a maize mutant 1750 that generates small and partially collapsed kernels, leading to either embryo or seedlings lethality. The mutant gene was mapped into a physical interval of about 200kb, which contains 4 candidate genes, through positional cloning with a F2 population of 2000 individuals. DNA sequencing revealed that gene A contains a SNP (G-A) located at the exon-intron boundary, giving rise to an alternative splicing and an expected truncated protein. Indeed, we detected two distinct transcripts with premature termination from the mutant kernels. In support of this, the protein encoded by gene A can not be detected in the mutant relative to the wild-type kernels using immunoblotting with antibody against gene A. Gene A encodes a novel protein localized to mitochondrial and is constitutively expressed with high levels during kernel development. Biochemical analysis revealed a protein A-related complex is essential for mitochondrial function. Together, we elaborate the indispensable role of this complex that is required for mitochondrial function and seed development.

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Abstract 95

Maize endosperm-specific transcription factor *PBF1* dynamically regulates targeted genes balancing protein and carbohydrate storage in a nitrogen dependent manner

Ning Lihua, Du Hongyang, Wang Yuanchong, Nasr Ullah Khan, Zhou Ling, He Bing, Ge Min, Xu Jinyan, Liang Shuaiqiang, Zhao Han✉

Jiangsu Provincial Key Laboratory of Agrobiolgy, Institute of Crop Germplasm and Biotechnology, Jiangsu Academy of Agricultural Sciences, Nanjing 210014, China

✉To whom correspondence should be addressed. Email: zhaohan@jaas.ac.cn

Phone: +86-25-84390751 Fax: +86-25-84390297

Abstract: Maize is an important crop that exhibits marked seed compositions in response to supplemental nitrogen (N). The prolamin-binding factor (PBF1) is a key transcription factor regulating the expression of genes encoding maize storage proteins, besides controlling starch synthesis. However, the effect of N supply on expression profile of genes targeted by PBF1 is still not well understood. In this study, N limitation orchestrated the expression of key genes involved in N and carbon (C) metabolism. We identified that many differential expressed genes were specifically bound by PBF1 under SN and DN conditions, respectively. GO analysis showed that PBF1 mainly regulated genes involved in endosperm storage products synthesis and accumulation under SN conditions, but affected transcription of abiotic stress genes under DN condition. In addition, we showed that under SN conditions, PBF1 binding tend to activate storage protein-related genes and repress carbohydrate synthesis and metabolism related genes. Taken together, this study provides insights into the regulatory network of the PBF1 and its role in the regulation of carbohydrate and protein synthesis and metabolism in an N-dependent manner.

Abstract 96

A new role for ethylene as a developmental signal controlling ear length and kernel number in maize

Qiang Ning^{1, §}, Yinan Jian^{1, §}, Yanfang Du¹, Ran Zhao¹, Xiaomeng Shen¹, Haitao Jia¹, Lei Liu^{1,2}, Fang Yang¹, David Jackson^{1,2,*}, Zuxin Zhang^{1,*}

¹ National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, PR China

² Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, NY 11724, USA.

*Corresponding author: zuxinzhang@mail.hzau.edu.cn; jacksond@cshl.edu

(submitted by 宁强 ningqiang_404@163.com)

Abstract: Maize (*Zea mays* L.), one of important cereal crop plants, produces two spatially separated inflorescences, the ear and the tassel. Axillary meristems produced on the ear inflorescence are initially indeterminate spikelet-pair meristems that form determinate spikelets which terminate with production of two pistillata florets through the arrest of the stamens. Therefore, meristematic activity of inflorescence meristems determines the number of florets on the ear, and fate of floral organs determines sex of floret. Here, we provide a direct evidence on ethylene level in the regulation of the meristem activity and then kernel number. We characterized that a gene encoding an ethylene biosynthesis enzyme the 1-aminocyclopropane-1-carboxylate oxidase2 (ACO2), is responsible for QTL *qEL7* for ear length and kernel number per ear by map-based cloning, gene expression, enzyme kinetic assay and transgenic validation. A 7_bp insertion/deletion closely nearby a FASCIATED EAR4 binding TGACG motif in *ZmACO2* promoter alters *ZmACO2* expression level and endogenous ethylene level. Silencing *ZmACO2* lines result in 14.6% to 24.3% of increase for ear length and 12.0% to 27.3% of increase for kernel number per row. The high ethylene level induces expression of BARREN INFLORESCENCE4 (BIF4) and AP2/EREBP transcription factors including INDETERMINATE SPIKELET1 and BRANCHED SIKLESS1 in the ear inflorescence. We propose a regulatory pathway of ethylene for activity maintenance of inflorescence meristem and provide a potential tool for improving grain yield by optioning endogenous ethylene level in maize.

Abstract 97

Cloning and characterization of a maize kernel defective mutant encodes a PPR protein

Yixuan Peng¹, Kaijian Fan¹, Riliang Gu¹, Li Li^{1*}, Jianhua Wang^{1*}

¹ Seed Science and Technology Research Center, Beijing Innovation Center for Seed Technology (MOA), Beijing Key Laboratory for Crop Genetic Improvement, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China.

*Corresponding Authors: lili2016@cau.edu.cn; wangjh63@cau.edu.cn

(submitted by 彭奕萱<pengyixuan@cau.edu.cn>)

Abstract: The development of maize kernel is tightly related to the yield, quality, and the seedling establishment as the seeds. Thus, studying the molecular mechanism of maize kernel development is essential which provides the knowledge of modern crop breeding based on molecular biology and genetics improvement. In our research, the mutant *small kernel X* (*sk-X*) was discovered from the offspring of self-inbred Ye478 which occurred spontaneous mutation, the stable inherited was confirmed as a recessive trait controlled by a single recessive nuclear gene with several generations. The *sk-X* mutant showed defective in both seed development (lower weight than normal kernels) and the plant establishment (shorter plant height). Moreover, delayed embryo and endosperm development was observed through DAPI-stained paraffin sections. The *sk-X* mutant was out-crossed to another inbred Zheng58, followed by self-pollination and generated the segregated F₂ population for gene mapping, the pooled RNA samples for normal kernels (WT) and the small kernels (*sk-X*) were collected respectively and sequenced on an Illumina HiSeq4000 platform, BSR-seq analysis showed a 18Mb interval. Using 864 individuals from F₂, the mutative gene *SK-X* was narrowed down to a 1.1Mb region with 12 codominant genetic markers. Within this small interval, a candidate gene encodes a PPR protein was down-regulated in *sk-X* mutant compared to the normal wild-types. Given that the expression difference, further sequencing was performed and a 414 bp Ds transposon insertion was detected on the first exon of this candidate, leading to a decreased expression in *sk-X*. Additionally, we performed a reverse genetic screen on a Mutator-derived mutants pool, another Mu-insertional allele was identified which showed exactly same phenotypic variation as *sk-X*, Further study of the molecular mechanism on this gene will continued for function exploration.

Abstract 98

玉米不同回交世代DH系遗传分析研究

齐欣，刘欣芳，马骏，孟庆国，姜敏*

辽宁省农业科学院玉米研究所，沈阳 110161

*通讯作者：姜敏<jm3192@126.com>

(齐欣<lnsnkyqx@163.com>)

摘要：以“辽3162”为受体，自交系“A619”为供体，通过杂交、回交得到回交群体，利用高频诱导系诱导加倍形成回交世代BC1F1和BC5F1的DH系各50份；以100份DH系为材料，通过比较不同回交世代的DH系和父母本的农艺性状，对群体做出评价；利用SSR分子标记分析群体的遗传分离特性。研究表明：DH系间各性状存在较大的遗传变异，株高、穗位高、穗重、百粒重、行粒数等农艺性状符合正态分布，DH系内整齐一致。通过筛选后得到的35对SSR分子标记引物对100个DH系进行遗传分离研究，不同世代回交群体DH系中没有出现明显偏分离比例，通过 χ^2 检测，未达到显著水平，所以在DH系群体中亲本的SSR标记在DH系中不存在明显的偏分离现象。

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Abstract 99

Systematic identification of endogenous RNA polymerase III promoters for efficient RNA guide based genome editing technologies in maize

Xiantao Qi², Le Dong¹, Changlin Liu¹, Long Mao¹, Fang Liu¹, Xin Zhang², Beiji Cheng², Chuanxiao Xie¹*

¹Institute of Crop Science, Chinese Academy of Agricultural Sciences, Beijing 100081, China

²Anhui Agricultural University, Anhui Province, Hefei 230036, Anhui, China

*Corresponding author: xiechuanxiao@caas.cn. (C. Xie).

(submitted by 祁显涛<xiantaoq@126.com>)

Abstract: Single-guide RNA(sg RNA) is one of the two core components of the CRISPR(clustered regularly interspaced short palindromic repeat)/Cas(CRISPR-associated) genome-editing technology. We established an in vitro Traffic Light Reporter (TLR) system, which is designated as the same colors as traffic lights such as green, red and yellow were produced in cells. The TLR can be readily used in maize mesophyll protoplast for a quick test of promoter activity. The TLR assay indicates the variation in transcription activities of the seven Pol III promoters, from 3.4%(U6-1) to over 21.0%(U6-6). The U6-2 promoter, which was constructed to drive sgRNA expression targeting the ZmWx1 gene, yielded mutation efficiencies ranging from 48.5% to 97.1%. Based on the reported and unpublished data, the in vitro TLR assay results were confirmed to be a readily system and may be extended to other plant species amenable to efficient genome editing via CRISPR/Cas. Our efforts provide an efficient method of identifying native Pol III-recognized promoters for RNA guide-based genome-editing systems in maize.

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Abstract 100

Dynamic patterns of protein-coding and noncoding elements across maize development in a maize F₁ hybrid and its inbred parents

Jia Qian¹, Linqian Han¹, Junfeng Wei¹, Zi Luo¹, Sijia Chen¹, Juan Li¹, Xi Wang¹, Lin Li^{1*}, Yirong Zhang^{2*}, Zhongfu Ni^{2*}

¹ National Key Laboratory of Crop Genetic Improvement, Crop Information Center, Huazhong Agricultural University, Wuhan 430070, China

² State Key Laboratory for Agrobiotechnology, Key Laboratory of Crop Heterosis Utilization (MOE), Beijing Key Laboratory of Crop Genetic Improvement, China Agricultural University, Beijing 100193, China

* Corresponding author

(Submitted by Jia Qian < qianjiapingxu@126.com >)

Abstract: Heterosis is widely utilized in agriculture, but its genetic and molecular basis is largely unclear. With the development of omic technologies, we now have the unprecedented chance to collect large omic datasets to decipher heterosis. In order to evaluate the contribution of different expressed elements to heterosis throughout maize development, we have collected a comprehensive transcriptomic dataset on 26 different tissues or developmental stages of two maize inbreds, B73 and Mo17, as well as their F₁ hybrid. We have detected 153,400 mRNAs (38,067 genomic loci), 10,152 lncRNAs (8,149 genomic loci), 61,936 circRNAs (>12,830 genomic loci), 34,3515 small RNAs (343515 small RNA genomic clusters), and 148 fusion transcripts across 26 different stages and tissues, which exhibited dramatic expression variation in B73, Mo17 and F₁s. The global comparison of all different functional elements across 26 tissues/stages for B73, Mo17, and their F₁s is ongoing.

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Abstract 101

Dissecting the genetic architecture that controls the domestication of maize leaf morphology

Yu-Hua Fu^{1,3#}, Guanghui Xu^{3#}, Wenchao Qin³, Huafeng Chen³, Xufeng Wang³, Qiuyue Chen³, Cheng Huang^{3,4}, Dan Li³, Dingyi Xu³, Jinge Tian³, Weihao Wu³, Sijia Lu⁵, Cong Li^{2,3} and Feng Tian^{3*}

¹ Guizhou Institute of Subtropical Crops, Guizhou Academy of Agricultural Sciences, Xingyi, 562400, China; ² College of Biological Science and Technology, Shenyang Agricultural University, Shenyang 110866, China; ³ State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Key Laboratory of Biology and Genetic Improvement of Maize (MOA), Beijing Key Laboratory of Crop Genetic Improvement, China Agricultural University, Beijing 100193, China; ⁴ College of Agronomy, Hunan Agricultural University, Changsha 410128, China; ⁵ School of Life Sciences, Guangzhou University, Guangzhou 510006, China

These authors contributed equally to this work

* Correspondence: Cong Li (lnlicong_123@163.com) and Feng Tian (ft55@cau.edu.cn)

(submitted by 秦文超 <qwenchao1995@163.com >)

Abstract: From its wild progenitor teosinte (*Zea mays* ssp. *parviglumis*), maize has experienced a dramatic morphological transformation. Although significant advances have been achieved in the identification of the genes that control the changes in overall plant architecture during domestication, the genetic basis that controls the changes in leaf morphology, an important component of plant architecture, remains poorly understood. Here, using a large population of 866 maize-teosinte BC₂S₃ recombinant inbred lines genotyped with 19838 SNP markers, we performed high-resolution quantitative trait locus (QTL) mapping for three leaf morphological traits, including leaf length, leaf width, and sheath length. We demonstrate that the three leaf traits were associated with distinct genetic architecture features and under relatively independent genetic control. This genetic independence was further validated by the analysis of near isogenic lines for target QTLs. QTL characterization revealed that the three leaf traits might have experienced directional selection for increased leaf size during maize domestication. We found that known leaf development genes identified by mutagenesis were significantly enriched in the support intervals of leaf trait QTLs, potentially indicating their important roles in regulating the natural variation in leaf traits. Our findings provide novel insights into the genetic basis that controls maize leaf evolution.

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Abstract 102

Evolutionary, structural and expression analysis of core genes involved in starch synthesis

Jianzhou Qu^{1,2}, Shutu Xu^{1,2}, Zhengquan Zhang^{1,2}, Guangzhou Chen^{1,2}, Yuyue Zhong^{1,2}, Linsan Liu^{1,2}, Renhe Zhang^{1,2}, Jiquan Xue^{1,2*}, Dongwei Guo^{1,2*}

¹The key Laboratory of Biology and Genetics Improvement of Maize in Arid Area of Northwest Region, Ministry of Agriculture and Rural Affairs, College of Agronomy, Northwest A&F University, Yangling, 712100, Shaanxi, China; ²Maize Engineering Technology Research Centre of Shaanxi Province, Yangling, 712100, Shaanxi, China

*Corresponding author: xjq2934@163.com; gdwei1973@126.com

(submitted by 渠建洲<qujz0220@163.com>)

Abstract: Starch is the main storage carbohydrate in plants and an important natural resource for food, feed and industrial raw materials. However, the details regarding the pathway for starch biosynthesis and the diversity of biosynthetic enzymes involved in this process remains largely unknown. Here we used a comprehensive phylogenetic analysis of 74 sequenced plant genomes to revisit the evolutionary history of the genes encoding ADP-glucose pyrophosphorylase (AGPase), starch synthase (SS), starch branching enzyme (SBE) and starch de-branching enzyme (DBE). We found that AGPase, SS, SBE and DBE have undergone complicated evolutionary processes in plants and that gene/genome duplications are likely responsible for the observed differences in isoform numbers. A structure analysis of these protein isoforms suggested that the deletion/mutation of amino acids in some active sites resulted in not only structural variation but also sub-functionalization or neo-functionalization. Meanwhile, AGPase-, SS-, SBE- and DBE-encoding genes exhibit spatio-temporally divergent expression patterns related to the composition of functional complexes in starch biosynthesis. Global gene co-expression network analysis also captured multiple second genes likely as node genes affect communication between core genes of starch biosynthesis, or indirectly involved in upstream metabolism processes to regulate the downstream starch metabolism. This study will be helpful for future studies aimed at increasing understanding of starch biosynthesis and the functional evolutionary divergence of AGPase, SS, SBE, and DBE in plants.

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Abstract 103

基于核磁共振的玉米不同籽粒类型单粒质量和含油率分析

李浩川¹, 曲彦志¹, 杨继伟¹, 崔丽洋¹, 毛熙岚¹, 刘宗华^{1*}

¹河南农业大学农学院, 郑州, 450002

*通讯作者: zhliu100@163.com

(刘宗华 <zhliu100@163.com>)

摘要: 针对现有玉米单倍体核磁共振分选系统基于一个含油率阈值,无法对胚败育籽粒和单倍体籽粒正确分选的问题,分别对玉米生物诱导产生的二倍体、单倍体和胚败育 3 种不同籽粒类型的单粒质量和含油率进行分析,提出了利用籽粒含油率双阈值提高单倍体正确识别率的分选方法。该研究以 2 个普通玉米杂交种和 3 个自交系为母本,以高油型诱导系为父本,进行生物诱导产生的 3 种不同类型籽粒为研究对象,利用核磁共振分选系统分别对不同类型籽粒的单粒质量和含油率进行测定,结果表明:单粒质量整体表现为单倍体二倍体胚败育,除二倍体籽粒与胚败育籽粒间存在极显著差异外,其他籽粒类型间差异不显著;不同类型籽粒的单粒质量平均变异系数为 16.62%,并且每个材料的 3 种籽粒类型间出现较大的重叠区域。而不同类型籽粒含油率整体表现为二倍体单倍体胚败育,变异性以二倍体最小,平均变异系数仅为 12.52%,其次是单倍体,而胚败育籽粒最高(34.14%),但其含油率最低且均 $\leq 2\%$;每个材料各自的 3 种类型籽粒间含油率呈现梯度分布,存在较明显的界限。由此可见,利用籽粒含油率能够区分玉米生物诱导的 3 种不同籽粒类型,而单粒质量则不能;通过设置二倍体籽粒的最小含油率为上限,胚败育籽粒的最大含油率为下限,利用含油率的双阈值可提高单倍体的正确识别率,为玉米生物诱导单倍体高效自动化分选提供依据。

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Abstract 104

***DEK43* Regulates Maize Kernel Development by Affecting *nad4* Splicing**

Li Li Wang¹, Ru Chang Ren¹, Lin Zhang, Ya Jie Zhao, Xian Sheng Zhang*, Xiang Yu Zhao*

State Key Laboratory of Crop Biology, College of Life Sciences, Shandong Agricultural University, Taian, Shandong 271018, China

¹ These authors contribute equally to this work.

*Correspondence: Xiang Yu Zhao (zhxy@sda.u.edu.cn), Xian Sheng Zhang (zhangxs@sda.u.edu.cn)

(submitted by 赵翔宇 < zhxy@sda.u.edu.cn >)

Abstract: Mitochondria is an important organelle in plant cell and mitochondrial respiratory chain complexes are essential for mitochondrial function. *nad4* gene codes mitochondrial respiratory chain complex I subunit IV, however, the mechanism underlying *nad4* gene splicing is still unclear. In this study, a defective kernel mutant *defective kernel 43* (*dek43*) was identified in maize. The *dek43* mutant has small, lethal, light-colored kernel. The mutated gene was identified through map-based cloning strategy and named *DEK43*. It encodes a P-type Pentatricopeptide repeat (PPR) protein that was mainly involved in the splicing process of organelle mRNA in higher plants. Then the splicing efficiency of the 22 mitochondrial group II introns was compared between wild type and *dek43* mutant. The result revealed that the splicing efficiency of the first and the third introns of the *nad4* was dramatically reduced. The further analysis showed that the abundance of complex I and super complex I+III₂ in *dek43* were significantly reduced compared with WT. In-gel NADH dehydrogenase assays indicated that the activities of complex I and super complex I+III₂ were both significantly reduced in *dek43*. Furthermore, the mitochondrial ultrastructure was damaged. The RNA-seq analysis revealed that the expression of mitochondrial related genes in *dek43* were up-regulated and several starch biosynthesis related genes were down-regulated. Taken together, our data suggested that *DEK43* played an important role in maintaining the function of mitochondria and the development of maize kernel.

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Abstract 105

Maize *Empty pericarp602* encodes a P-type PPR protein that is essential for seed development

Zhenjing Ren^{1,2,#}, Kaijian Fan^{1,2,#}, Ting Fang¹, Jiaojiao Zhang¹, Li Yang¹, Jianhua Wang^{2,*}, Guoying Wang^{1,*}, and Yunjun Liu^{1,*}

¹ Institute of Crop Sciences, Chinese Academy of Agricultural Sciences, Beijing 100081, China

² College of Agronomy and Biotechnology, China Agricultural University, Beijing 100094, China

These authors contributed to the work equally.

* Correspondence: Yunjun Liu (liuyunjun@caas.cn), Guoying Wang (wangguoying@caas.cn), or Jianhua Wang (wangjh63@cau.edu.cn)
(submitted by 任珍静 renzhenjinggogo@163.com)

Abstract: PPR (pentatricopeptide repeat) proteins play crucial roles in intron splicing, which is important for RNA maturation. Identification of novel PPR protein with the function of intron splicing would help to understand the RNA splicing mechanism. In this study, we identified the maize *empty pericarp602* (*emp602*) mutants, the mature kernels of which showed empty pericarp phenotype. We cloned the *Emp602* gene from *emp602* mutants and revealed that *Emp602* encodes a mitochondrial-localized P-type PPR protein. We further revealed that *Emp602* is specific for the *cis*-splicing of mitochondrial *Nad4* intron 1 and intron 3, and mutation of *Emp602* led to the loss of mature *Nad4* transcripts. The loss of function of *Emp602* nearly damaged the assembly and accumulation of complex I and arrested mitochondria formation, which arrested the seed development. The failed assembly of complex I triggers significant up-regulation of *Aox* expression in *emp602* mutants. Transcriptome analysis showed that the expression of mitochondrial-related genes, e.g. the genes associated with mitochondrial inner membrane presequence translocase complex and electron carrier activity, were extensively upregulated in *emp602* mutant. These results demonstrate that EMP602 functions in the splicing of *Nad4* intron 1 and intron 3, and the loss of function of *Emp602* arrested maize seed development by disrupting the mitochondria complex I assembly.

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Abstract 106

Dissecting Heterosis During the Ear Inflorescence Development

Stage in Maize via a Metabolomics-based Analysis

Xia Shi^{1*}, Xuehai Zhang^{1*}, Dakun Shi¹, Xiangge Zhang¹, Weihua Li^{1*}, Jihua Tang^{1,2 *}

¹National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China; ²Hubei Collaborative Innovation Center for Grain Industry, Yangtze University, Jingzhou 434025, China

*Corresponding authors: tangjihua1@163.com (submitted by Shixia<mrshi0614@126.com>)

Abstract: Heterosis can increase the yield of many crops and has been extensively applied in agriculture. In maize, female inflorescence architecture directly determines grain yield. Thus, exploring the relationship between early maize ear inflorescence development and heterosis regarding yield-related traits may be helpful for characterizing the molecular mechanisms underlying heterotic performance. In this study, we fine mapped the overdominant heterotic locus (hlEW2b), associated with ear width, in an approximately 1.98-Mb region based on analyses of chromosome segment substitution lines and the corresponding testcross population. Maize ear inflorescences at the floral meristem stage were collected from two inbred lines, one chromosome segment substitution line that carried hlEW2b (sub-CSSL16), the receptor parent lx9801, and the Zheng58 × sub-CSSL16 and Zheng58 × lx9801 hybrid lines. A total of 256 metabolites were identified, including 31 and 24 metabolites that were differentially accumulated between the two hybrid lines and between the two inbred lines, respectively. Most of these metabolites are involved in complex regulatory mechanisms important for maize ear development. For example, nucleotides are basic metabolites affecting cell composition and carbohydrate synthesis. Additionally, nicotinate and nicotinamide metabolism is important for photosynthesis, plant stress responses, and cell expansion. Moreover, flavonoid and phenolic metabolites regulate auxin transport and cell apoptosis. Meanwhile, phytohormone biosynthesis and distribution influence the cell cycle and cell proliferation. Our results revealed that changes in metabolite contents may affect the heterotic performance related to ear width and yield in maize hybrid lines. This study provides new clues in heterosis at the metabolomics level and implies that differentially accumulated metabolites made distinct contributions to the heterosis at an early stage of ear inflorescences development.

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Abstract 107

Overexpression of maize heat stress transcription factor gene *ZmHsf14* enhances thermo tolerance in transgenic *Arabidopsis*

Qianqian Qin, Nannan Song, Jing Wang, Muhammad Yahya, Manli Zhan, Weina Si*,
Haiyang Jiang*

National Engineering Laboratory of Crop Stress Resistance Breeding, Anhui Agricultural University,
Hefei 230036, China

*Corresponding authors: hyjiang@ahau.edu.cn

(Submitted by 宋楠楠< 18709856246@163.com>)

Abstract: Heat shock transcription factor (Hsfs) are highly conserved among eukaryote and always play vital role in plant stress responses. Whereas, function and mechanism of Hsfs in maize is limited. In the present study, an HSF gene *ZmHsf14* was cloned from maize, which was up-regulated under heat shock and salinity treatment. Subcellular localization analysis by transient expression assay in maize protoplasts indicated that *ZmHsf14* was located on the nucleus and cell membrane. Yeast one-hybrid assay showed that *ZmHsf14* has transcription activity in yeast cells. In addition, overexpression of *ZmHsf14* gene in *Arabidopsis* significantly increased the survival rate of transgenic plants under heat shock treatment. Whereas, no significant phenotypic differences were observed between transgenic plants and WT plants under salt or drought stresses. Subsequently physiological indexes measurements exhibited that under heat stress, MDA content of *ZmHsf14* transgenic plants was significantly lower than that of WT plants, while proline content was dramatically higher than that of WT plants. Meanwhile, our results showed that expression level of some heat responsive genes in transgenic plants were significantly higher than WT plants. To conclude, our results suggest that overexpression of the *ZmHsf14* gene enhanced the heat tolerance in transgenic *Arabidopsis*.

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Abstract 108

Dual functions of *ZmNF-YA3* in photoperiod dependent flowering and abiotic stress responses in maize

Huihui Su^{1*}, Yingying Cao^{1*}, Lixia Ku^{1*†}, Wen Yao¹, Yanyong Cao², Zhenzhen Ren¹, Dandan Dou¹, Huitao Wang¹, Zhaobin Ren¹, Huafeng Liu¹, Lei Tian¹, Yaogang Zheng¹, Chen Chen¹, and Yanhui Chen^{1†}

¹ Synergetic Innovation Centre of Henan Grain Crops and National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China

² Henan Academy of Agricultural Science, Zhengzhou, Henan, 450002, China

* These authors contributed equally to this work.

† Correspondence: chy9890@163.com or kulixia0371@163.com

(submitted by 苏慧慧 <1079483412@qq.com>)

Abstract: Nuclear factor-Y (NF-Y) transcription factors are important regulators of several essential biological processes, including embryogenesis, drought resistance, meristem maintenance, and photoperiod-dependent flowering in *Arabidopsis*. However, the regulatory mechanisms of NF-Ys in maize (*Zea mays*) are not well understood yet. In this study, we identified an NF-Y transcription factor, *ZmNF-YA3*. Genome-wide analysis showed that *ZmNF-YA3* bound to >6000 sites in the maize genome, 2259 of which are associated with genic sequences. *ZmNF-YA3* was found to interact with CONSTANS-like (CO-like) and flowering promoting factor1 (FPF1) through yeast two-hybrid and bimolecular fluorescence complementation (BiFC) assays. Quantitative real-time reverse transcription-PCR (qRT-PCR) combined with yeast one-hybrid assay and EMSA suggested that *NF-YA3* could promote early flowering by binding to the FLOWERING LOCUS T-like12 (FT-like12) promoter in maize. Moreover, we also showed that *ZmNF-YA3* could improve drought and high-temperature tolerance through binding to the promoter regions of *bHLH92*, *FAMA*, and the jasmonic acid activator *MYC4*, respectively. These results contribute to a comprehensive understanding of the molecular mechanisms and regulatory networks of NF-Y transcription factors in regulating maize flowering time and stress response in maize.

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Abstract 109

Fine Mapping of *qLA3* Controlling Leaf Angle in Maize

Fei Sun; Weiya Li; Zhixing Yang; Jieyuan Yao; Yangzi Zhao; Jiansheng Li; Xiaohong Yang*

State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center of China, China Agricultural University, Beijing, 100193, P. R. China

*Corresponding author: yxiaohong@cau.edu.cn

(submitted by 孙飞<sunf00@cau.edu.cn>)

Abstract: Maize, as one of three major crops around the world, exhibits the highest grain yield and the largest planting area in the world as well as in China. However, there is still a large gap in maize yield per unit area compared with the developed countries such as the United States of America. Erect maize variety has a small leaf angle and a large leaf delamination under dense planting conditions, meanwhile the microclimate and permeability is ameliorated, which is favorable for capturing the luminous energy and carbon dioxide between each lamina, enhancing photosynthesis and respiration, and subsequently improving luminous energy utilization as well. Leaf angle is a key factor for plant architecture, increasing the angle between the leaf midrib and the ground maintains light capture under high plant density. Previously, a major QTL, *qLA3*, controlling leaf angle (LA) was identified on chromosome 3. To fine-map *qLA3*, we used a heterogeneous inbred family (HIF), XMO10, which was heterozygous only at *qLA3* to generate a large near-isogenic line (NIL) population by backcrossing to Mo17 two times. Seven InDel markers were developed in the QTL region, and 11 recombinants were screened using 4056 individuals derived from XMO10. Using the progeny test strategy, *qLA3* was finally narrowed down to 18kb interval, which contained one annotated gene. Furthermore, the functional analysis of *qLA3* will be performed by using microscopic observation, expression analysis, genetic transformation, and so on. These results will provide useful information for theoretical guidance for genetic improvement of plant architecture in maize.

Abstract 110

***Empty pericar23* encoding a mitochondrial pentatricopeptide repeat protein, is involved in the cis-splicing of *nad1* intron 2 and seed development in maize**

Feng Sun, Yun Shen, Yiwei Liu and Bao-Cai Tan*

Key Laboratory of Plant Development and Environmental Adaptation Biology, Ministry of Education, School of Life Sciences, Shandong University, Qingdao 266237, China

*Corresponding author: bctan@sdu.edu.cn

(submitted by Feng Sun <epusun@sdu.edu.cn>)

Abstract: Group II introns in plant mitochondria have lost the essential self-splicing capability that is distinct from their bacterial ancestors. To achieve the intron splicing, mitochondria have to recruit a variety of host-derived cofactors for cis- and trans-splicing. The pentatricopeptide repeat (PPR) is the largest family of RNA binding proteins and governs nearly all aspects of RNA metabolism at the post-transcriptional level. Because of the embryo-lethality caused by the loss-of-function mutants, only a few PPRs are characterized. Here, we defined the function of EMPTY PERICARP23 (EMP23), in the mitochondrial *nad1* intron 2 splicing and complex I biogenesis and seed development in maize. *Emp23* encodes a P-subfamily PPR protein that localizes to mitochondria. The loss-of-function of *Emp23* severely arrested the embryogenesis and endosperm development, giving rise to an empty pericarp phenotype in maize. The cis-splicing of the second intron of *nad1*, was specifically affected, accumulating an unspliced *nad1* transcript. The splicing defect of *nad1* intron 2 leads to a disassembly of complex I, which greatly reduced its activity. The alternative pathway is compromised to be activated and the accumulation of other complexes in the respiratory chain is increased, manifesting severe morphological defects of mitochondria in *emp23* mutants. These results reveal that *Emp23* is specifically required for the cis-splicing of *nad1* intron 2, mitochondrial complex I biogenesis, and hence is crucial to the embryogenesis and endosperm development in maize.

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Abstract 111

细胞周期转换蛋白 APC/C^{ZmCCS52B} 调节玉米生物量的研究

孙瑞琪¹, 路小铎², 曹梦强¹, 吕家发¹, 张春义³, 张宪省⁴, 李翠玲^{1*}, 丁兆军^{1*}

¹ 山东大学生命科学学院, 青岛, 266237

² 齐鲁师范学院生命科学学院, 济南, 250013

³ 中国农业科学院生物技术研究所, 北京, 100081

⁴ 山东农业大学生命科学学院, 泰安, 271018

*通讯作者: cuilingli@sdu.edu.cn

(submitted by 孙瑞琪<15737954304@126.com>)

摘要: 植物生物量的分配和改善不仅是植物功能生态学的核心科学问题之一, 也是遗传育种上需要解决的科学问题之一。因此, 对于植物是生物量的研究具有非常重要的意义。我们从玉米自交系 B73 的 EMS 诱变突变体库筛选到一株具有矮化, 茎秆变细变短, 叶片和雌穗变小等生物量减少表型的突变体。通过 EcMutMp 的方法, 我们定位到了该突变位点, 证明该突变体是由基因 ZmCCS52B 发生突变引起的。详细的表型分析发现, *zmccs52b* 突变体矮化的表型是由于茎节变短引起的; 对叶片下表皮细胞的观察发现 *zmccs52b* 突变体叶片变窄的表型是由于该突变体单个细胞体积增大引起的; 通过对授粉不同天数后的籽粒进行切片分析发现, *zmccs52b* 突变体胚乳由于细胞核内复制增加导致单个细胞明显增大, 但由于整体细胞数目的减少, 使得胚乳及整个植株表现出整体器官变小及生物量明显减少的表型。

ZmCCS52B 是人和动物 Cdh1 的同源蛋白, 是细胞周期中 APC/C 复合体的激活因子, 参与到 APC/C 特异性的识别底物的过程之中。通过酵母双杂交筛选鉴定到了 ZmWEE1 能够和 ZmCCS52B 蛋白互作, 我们的 CO-IP 和 BIFC 实验也验证了该结果。ZmWEE1 作为一种蛋白激酶, 可以抑制 CDK1 激酶的活性从而抑制细胞进入有丝分裂。ZmWEE1 含有 APC/C 复合体特异性识别底物的 KEN Box 说明 ZmCCS52B 很可能通过特异性识别并降解底物 ZmWEE1 来参与细胞周期的调控, 并最终影响玉米生物量的积累和植株高度。

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Abstract 112

A large-scale circular RNA profiling reveals universal molecular mechanisms responsive to drought stress in maize and Arabidopsis

Pei Zhang¹, Yuan Fan¹, Xiaopeng Sun¹, Lu Chen¹, William Terzaghi², Etienne Bucher³, Lin Li¹, Mingqiu Dai^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China.

²Department of Biology, Wilkes University, Wilkes-Barre, Pennsylvania 18766

³IRHS, Université d'Angers, INRA, AGROCAMPUS-Ouest, SFR4207 QUASAV, Université Bretagne Loire, 49045 Angers, France

Corresponding author: mingqiudai@mail.hzau.edu.cn

(submitted by 孙霄鹏<mingqiudai@mail.hzau.edu.cn>)

Abstract: In this study, a large scale circRNA profiling identified 2174 and 1354 high-confidence circRNAs in maize and Arabidopsis, respectively, and most were differentially expressed in response to drought. A substantial number of drought-associated circRNA hosting genes were involved in conserved or species-specific pathways in drought responses. In most cases, maize circRNAs were negatively correlated with sRNA accumulation. In 368 maize inbred lines, the circRNA-hosting genes were enriched for SNPs associated with circRNA expression and drought tolerance, implying either important roles of circRNAs in maize drought responses or their potential use as biomarkers for breeding drought-tolerant maize. Additionally, the expression levels of circRNAs derived from drought-responsible genes encoding calcium-dependent protein kinase and cytokinin oxidase/dehydrogenase were significantly associated with drought tolerance of maize seedlings. Specifically, Arabidopsis plants overexpressing circGORK (Guard cell outward-rectifying K⁺-channel) were hypersensitive to ABA, but insensitive to drought, suggesting a positive role of circGORK in drought tolerance. We report the transcriptomic profiling and transgenic studies of circRNAs in plant drought responses, and provide evidences highlighting the universal molecular mechanisms involved in plant drought tolerance.

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Abstract 113

Map-based cloning, phylogenetic and microsynteny analyses of *ZmMs20* gene regulating male fertility in maize

Yanbo Wang¹, Dongcheng Liu^{1,2}, Youhui Tian¹, Suowei Wu^{1,2}, Xueli An^{1,2}, Zhenying Dong^{1,2}, Simiao Zhang¹, Jianxi Bao¹, Ziwen Li^{1,2}, Jinping Li² and Xiangyuan Wan^{1,2*}

¹Biology and Agriculture Research Center, University of Science and Technology Beijing, Beijing 100024, China; ²Beijing Engineering Laboratory of Main Crop Bio-Tech Breeding, Beijing International Science and Technology Cooperation Base of Bio-Tech Breeding, Beijing Solidwill Sci-Tech Co. Ltd., Beijing 100192, China.

*Correspondence: wanxiangyuan@ustb.edu.cn (submitted by 田有辉 < tianyouhui@ustb.edu.cn >)

Abstract: Genic male sterility (GMS) mutant is a useful germplasm resource for both theory research and production practice. The identification and characterization of GMS genes and mutants in maize (*Zea mays*) have deepened our understanding of the molecular mechanisms controlling anther/pollen development and enabled development and efficient use of many biotechnology-based male-sterility (BMS) systems for heterosis utilization. Here, we reported a complete GMS mutant (*ms20*), which displays abnormal anther cuticle and pollen development. Its fertility restorer gene *ZmMs20* was isolated by map-based cloning and found to be a new allele of *IPE1* encoding a glucose methanol choline (GMC) oxidoreductase involved in lipid metabolism in anther. Phylogenetic and microsynteny analyses showed that *ZmMs20* was conserved among gramineous species, which provides clues for creating GMS materials in other crops. Nevertheless, we predicted that gene duplication may take place after the divergence of monocots and dicots, because many orthologs of *ZmMs20* were found in dicots genomes and the defects of null mutants of these orthologous genes in *Arabidopsis* is mild compared to that of *ms20*. The expression pattern of all the 17 maize cloned GMS genes was analyzed based on RNA-seq data of maize anther during eight developmental stages (stages 5 to 11). The expression pattern of *ZmMs20* was found to be similar to those of *Ms7*, *Ms26*, *Ms6021*, *APV1*, and *IG1* genes, which will give some cues for deciphering their functional relationships in regulating male fertility. Finally, two functional markers of *ZmMs20/ms20* were developed and tested for creating maize *ms20* male-sterile lines in 353 genetic backgrounds, and then an artificial maintainer line of *ms20* GMS mutation would be created by using *ZmMs20* gene, *ms20* mutant and BMS system. Taking together, this work will promote our understanding to functional mechanism of male fertility and facilitate molecular breeding of *ms20* male-sterility lines for hybrid seed production in maize.

Funding acknowledgement: National Key R&D Program of China (2018YFD0100806, 2017YFD0102001, 2017YFD0101201), National Transgenic Major Program of China (2018ZX0801006B, 2018ZX0800922B) and National Natural Science Foundation of China (31771875, 31871702).

Abstract 114

顺反调控多态性调控玉米籽粒萌发杂种优势形成的网络解析

万炯¹, 丁冬¹, 王琪月¹, 孟淑君¹, 张稳¹, 马拴红¹, 汤继华^{1*}

¹河南农业大学, 郑州, 450046

*通讯作者: tangjihua1@163.com

(submitted by 万炯<wan__jiong@163.com>)

摘要: 杂种优势是在作物育种中得到广泛应用的生物学现象。但是目前关于杂种优势遗传机理的解析仍停留在假说阶段。显性假说 (Jones 1917)、超显性假说 (Shull 1908)、拟超显性假说 (Mangelsdorf 1952)。这三种假说均是基于位点的杂合性, 即单基因位点等位基因间的互作而提出。而在基因组层面, 杂合基因组中基因相对于中亲值的表达模式 (加性/非加性, 显性/超显性) 是造成杂种优势的原因和表现形式。本研究选取郑 58、昌 7-2、郑单 958、郑单 958 (反交) 籽粒萌发 24 小时的胚, 通过转录组、蛋白组测序, 结合生物信息学数据分析, 在全基因组的水平上对基因的表达模式进行分析以及等位基因特异表达进行分析。综合基因表达模式和等位基因特异表达分析, 并进一步为解释杂种优势的分子机理提供全基因组水平的视角, 并应用于玉米杂种优势利用育种实践。

Abstract 115

***ZmCom1* is required for both mitotic and meiotic recombination in Maize**

Yazhong Wang¹, Luguang Jiang¹, Ting Zhang¹, Juli Jing¹ and Yan He^{1*}

¹MOE Key Laboratory of Crop Heterosis and Utilization, National Maize Improvement Center of China, China Agricultural University, Beijing, China

*Corresponding author: yh352@cau.edu.cn

(submitted by 汪亚中 <wangyazhong@cau.edu.cn>)

Abstract: CtIP/Ctp1/Sae2/Com1, a highly conserved protein from yeast to higher eukaryotes, is required for DNA double-strand break repair through homologous recombination (HR). In this study, we identified and characterized the COM1 homolog in maize. The *ZmCom1* gene is abundantly expressed in reproductive tissues at meiosis stages. In *ZmCom1*-deficient plants, meiotic chromosomes are constantly entangled as a formation of multivalents and accompanied with chromosome fragmentation at anaphase I. In addition, the formation of telomere bouquet, homologous pairing and synapsis were disturbed. The immunostaining assay showed that the localization of ASY1 and DSY2 was normal, while ZYP1 signals were severely disrupted in *Zmcom1* meiocytes, indicating that *ZmCom1* is critically required for the proper SC assembly. Moreover, RAD51 signals were almost completely absent in *Zmcom1* meiocytes, implying that COM1 is required for RAD51 loading. Surprisingly, in contrast to the *Atcom1* and *Oscom1* mutants, *Zmcom1* mutant plants exhibited a number of vegetative phenotypes under normal growth condition, which may be partly attributed to mitotic aberrations including chromosomal fragmentation and anaphase bridges. Taken together, our results suggest that although the roles of *COM1* in HR process seem to be primarily conserved, the *COM1* dysfunction can result in the marked dissimilarity in mitotic and meiotic outcomes in maize compared to Arabidopsis and rice. We suggest that this character may be related to the discrete genome context.

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Abstract 116

Development of a Haploid-Inducer Mediated Genome Editing (IMGE) System for Accelerating Maize Breeding

Baobao Wang^{1,3,*}, Lei Zhu^{1,3}, Binbin Zhao¹, Yongping Zhao¹, Yurong, Xie¹, Zhigang Zheng², Yaoyao Li², Juan Sun² and Haiyang Wang^{2,*}

¹Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing 100081, China

²School of Life Sciences, State Key Laboratory for Conservation and Utilization of Subtropical Agro-Bioresources, South China Agricultural University, Guangzhou 510642, China

³These authors contributed equally

*To whom correspondence should be addressed: wangbaobao@caas.cn or whyang@scau.edu.cn

(submitted by 王宝宝 < wangbaobao@caas.cn >)

Abstract: Crop breeding aims to generate pure inbred lines with multiple desired traits. Doubled haploid (DH) and genome editing using CRISPR/Cas9 are two powerful game-changing technologies in crop breeding. However, both of them still fall short in rapid generation of pure elite inbred lines with integrated favorable traits. We report here the development of a Haploid-Inducer Mediated Genome Editing approach (IMGE for short), which utilizes a maize haploid inducer (HI) line carrying a CRISPR/Cas9 cassette targeting for a desired agronomic trait to pollinate an elite maize inbred line, and to generate genome edited haploids in the elite maize background. Homozygous pure DH lines with the desired trait improvement could be generated within two generations, thus bypassing the lengthy procedure of repeated crossing and backcrossing used in conventional breeding for integrating a desirable trait into elite commercial backgrounds. We envisage that this technology could be widely used to accelerate crop breeding.

Funding acknowledgement: National Key R&D Program of China (2016YFD0100303 and 2016YFD0101001) and Beijing Natural Science Foundation (6172032).

Abstract 117

百奥云-基于云计算的育种数据分析管理平台

周欢, 谢志春, 朱芳, 王冰冰*

长沙百奥云数据科技有限公司, 长沙, 410205

*通讯作者: bwang@biobin.com.cn

(submitted by 王冰冰<bwang@biobin.com.cn>)

随着大规模测序和自动化采集设备在遗传育种上的应用, 育种相关的数据呈现爆发式增长态势。为有效管理和分析育种大数据, 我们开发了一套基于B/S架构的全新软件—百奥云智能育种平台。百奥云平台可以整合各种育种相关数据(基因型, 表现型及环境数据), 应用统计方法对育种数据进行深入分析, 并结合移动互联网、可视化和机器学习等技术, 帮助育种家采集育种和管理数据、筛选育种材料, 进行科学的育种决策。

百奥云育种平台支持多作物/多语种/多机构/多用户/多数据源(CLOUD)管理模式, 并设计了多种数据接口, 支持从多种已有的育种数据库和Excel表格中获取数据。为便于用户采集田间数据, 百奥云开发了一款简便易用的微信小程序, 无需下载和安装任何软件, 通过手机微信即可快速采集性状数据。针对基因型数据快速增长的趋势, 百奥云采用了主流的大数据技术来管理SNP基因型数据, 可快速响应用户的请求。数据分析上, 百奥云平台除了常规的方差分析外, 还可对原始数据进行空间统计校正, 用混合线性模型估算不同材料的育种值, 以及对选定的区域进行分析等。百奥云平台可整合多年多点的历史数据, 通过可视化的方式展示育种材料在不同区域、不同时间的表现, 可帮助育种家进行准确的育种决策和科学的品种定位。

百奥云智能育种平台目前已正式上线, 对科研人员免费开放。访问地址:

<http://breeding.biobin.com.cn/>。

Abstract 118

Evolutionary divergence of the plant MCIA complex that is required for mitochondrial complex I assembly and seed development

Jiacheng Ni², Yongyan Wang¹, Hongyu Chen¹, Jihua Tang¹ and Guifeng Wang^{1*}

¹ Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China;

² Shanghai Key Laboratory of Bio-Energy Crops, School of Life Sciences, Shanghai University, Shanghai 200444, China.

*Corresponding author: guifeng.wang@henau.edu.cn

(submitted by Guifeng Wang < guifeng.wang@henau.edu.cn>)

Abstract: Mitochondria, the powerhouse of the cell, are present in almost all eukaryotic cells where their primary role is the generation of ATP through oxidative phosphorylation. Mitochondria are double membrane bound organelles that evolve from endosymbiotic purple non-sulphur gram-negative bacteria (α -proteobacteria) about 2 billion years ago. The respiratory chain contains five respiratory complexes that allow for the establishment and utilization of a proton gradient across the mitochondrial inner membrane. Mitochondrial Complex I (NADH:ubiquinone oxidoreductase) has 44 subunits of which 7 are encoded by mitochondrial DNA (mtDNA), which utilizes a flavin mononucleotide (FMN) and its hydrophobic domain to oxidize NADH. Complex I assembly is an intricate process that are coordinately controlled by assembly factors in a step-wise fashion. In human, the mitochondrial complex I assembly (MCIA) complex, harboring five factors, is involved in the early assembly of the P_P-b subcomplex. The four core proteins in the MCIA are NDUFAF1 (NADH:ubiquinone oxidoreductase complex assembly factor 1), ECSIT (evolutionarily conserved signalling intermediate in toll pathway), ACAD9 (acyl-coA dehydrogenase family member 9), and TMEM126B (transmembrane protein 126B), as well as possible involvement of TIMMDC1 (translocase of inner mitochondrial membrane domain containing 1). However, most of these assembly factors have lost corresponding orthologs in plant. Here, we discuss the evolutionary divergence of the plant MCIA complex and its functions in mitochondrial complex I assembly and seed development.

Funding acknowledgement: National Natural Sciences Foundation of China (31771800, U1804235), Outstanding Youth Foundation of Henan Province (2018JQ0018), and Shanghai Pujiang Talent Program (17PJD014).

Abstract 119

Characterization and fine mapping of a novel defective kernel mutant in maize

Kaijian Fan^{1,2,*}, Guoying Wang^{2,*}, Jianhua Wang^{1,*}

¹College of Agronomy and Biotechnology, China Agricultural University, Beijing 100094, China

²Institute of Crop Sciences, Chinese Academy of Agricultural Sciences, Beijing 100081, China

*Correspondence: Guoying Wang(wangguoying@caas.cn); Jianhua Wang(wangjh63@cau.edu.cn)

(submitted by Guoying Wang<wangguoying@caas.cn>)

Abstract: Endosperm is one of the most important part of maize kernel and has great impact on its size and quality. Although many biochemical metabolic pathways have been reported to be involved in endosperm development, its regulatory mechanism is still not entirely clear. Here, we report a new defective kernel mutant from the offspring of *Mutator* (*Mu*) active lines. It showed small and opaque kernel, dramatically reduced endosperm content and abnormal starch granule. The bulked segregant RNA-seq (BSR-seq) mapped the causal gene to the long arm of chromosome 5. Using 12268 mutant individuals from a F₂ population, we further mapped the gene to a physical region of 780-kb between the markers InDel140 and InDelM3, which includes 14 protein coding genes according to the corresponding interval of B73 reference genome. Compared with the wild type, the 14 genes were failed amplification and the expressed reads undetected in mutant. Hence, we deduced a large fragment with all the 14 genes was absent in the candidate region of mutant. Analysis of these genes and their homologous genes in the syntenic rice region found that none of these genes had been reported to be related to kernel development. Our study will facilitate the isolation of the underlying gene and advance our understanding of the regulation relationship of maize endosperm development.

Abstract 120

A natural allele *vitreous endosperm1* regulates endosperm texture in maize seed

Haihai Wang[#], Yongcai Huang[#], Qiao Xiao[#], Xing Huang and Yongrui Wu*

[#] These authors contributed equally to this work

*Corresponding Author: yrwu@sibs.ac.cn

National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology & Ecology, Shanghai Institutes

Abstract: Vitreous endosperm confers strength to withstand mechanic damage during harvesting, transportation and storage of seed and therefore is a critical agronomic trait for maize breeding. Many *opaque* or *floury* mutants resulting from a single gene mutation have defects in biosynthesis of proteins and starch, and even abnormalities in seed development. In the natural population, different inbred lines often display variable endosperm texture phenotypes ranging from complete opacity to full vitreousness. To understand the mechanisms for natural variation in endosperm texture, we introgressed QTLs from four vitreous inbred lines into an opaque inbred line (A619, as the recurrent parent) to construct the near-isogenic lines (NILs). In BC5F1, the four NIL populations segregated vitreous and opaque kernels approximately at 1:1 ratio. Based on BSA and map-based cloning, we mapped a QTL (designated *vitreous endosperm1*, *ven1*) in a 1.3 Mb region using the NIL population of W64A and A619. Consistently, this QTL was located at the same locus using the other three NIL populations. The expression of *ven1* was significantly lower in A619 and NIL^{A619} compared to W64A and NIL^{W64A}. RNA *in situ* analysis revealed that *ven1* predominantly expressed in starchy endosperm cells from the middle to crown area, but not in the aleurone, sub-aleurone cells and starchy endosperm cells from the middle to basal area. Transcript and protein levels of zein genes were similar in A619 and NIL^{A619}. However, the starch granules in 18 and 24-DAP endosperms of NIL^{A619} were obviously reduced, but the expression of starch synthetic genes and proteins were not apparently affected compared to NIL^{W64A}. By TEM observation, we found that the starch granules were enveloped by a dispersed and loosen membrane. Cloning and functional studying *ven1* will help us understanding vitreous endosperm formation in maize seed. We are currently actively working on its mechanisms.

Abstract 121

Arbuscular mycorrhizal symbiosis and abiotic stress in maize

Hao Wang¹, Yinglong Chen^{1,2*}

¹State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Institute of Soil and Water Conservation, Northwest A&F University, & Chinese Academy of Sciences, Yangling 712100, China

²The UWA Institute of Agriculture, & School of Agriculture and Environment, The University of Western Australia, Perth WA 6009, Australia

*Corresponding author: yinglongchen@hotmail.com

(submitted by 王浩<wanghao0225@163.com>)

Abstract: Maize is one of the staple crop cultivated around the globe and its productivity expect to improve in a context of stressed habitat. Maize roots, as with most land plants, have been colonized by arbuscular mycorrhizal fungi (AMF) to establish mycorrhizal association. Also, in the past decade, evidences that show the beneficial effects of AMF on maize performance and resistance to abiotic stress in filed or controlled conditions are emerging. We thus collect recent literature, try to a) unravel the underpinning mechanisms of AMF-mediated plant tolerance to crucial stresses including drought, salinity, nutrient-deficiency and toxic metals/metalloids; b) appraise the right maize host-AMF combination fit in with the specific context; c) highlight the prospective aspects for maize breeding in future work, supporting the idea that AM symbiosis can be drew upon to develop agronomic practices for sustainable maize production along with a challenging environment.

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Abstract 122

玉米 *PIN1a* 基因异源表达及介导百脉根-AM 真菌共生功能分析

李晓玉*, 朱苏文, 王鹤群, 周媛

安徽农业大学生命科学学院, 合肥, 230036

*通讯作者: lixiaoyu@ahau.edu.cn

(submitted by 王鹤群<wanghequn@ahau.edu.cn>)

摘要: 在高等植物中, 生长素及其极性运输参与控制了许多植物生理生化过程, 如胚胎发育、侧根形成、维管束发育、向光反应、以及根部向地性等。PIN 蛋白家族是目前研究较多的生长素输出载体, 对生长素在植物组织的极性运输中起重要的调控作用。项目组前期发现, 丛枝菌根真菌可与玉米形成很好的共生关系。AM 真菌可显著促进玉米侧根的生长, 这可能是因为 AM 真菌侵染改变植物根部生长素的重新分布从而诱导侧根的增多。因此, 本课题以 AM 真菌和玉米共生体为研究对象, 通过 PIN 基因家族的生物信息学和诱导表达模式分析, 克隆了调节玉米根系生长素重新分布的关键 PIN 基因 *ZmPIN1a*, 并通过转化百脉根体外毛状根和玉米敲除转化验证该基因在调节生长素分布及侧根生长中的作用, 并结合 AM 真菌侵染后的转录组分析结果, 解析调控的关键调控网络。结果表明, 过表达转基因植株表型根系数目是高于野生型百脉根的, 所以初步推断, *ZmPIN1a* 能够促进 AMF 与百脉根的共生, 将该基因敲除后, *ZmPIN1a* 的水平表达呈下降的趋势, 一定程度上影响了 AMF 的生长发育, 从而使得百脉根根系发育减缓, 并导致根系发育不发达, 植株瘦弱。过表达转基因百脉根中的孢子定殖率在 25 天后有大幅度增长的趋势, 其增长速度高于野生型百脉根, 且对 *ZmPIN1a* 的敲除证明, 该基因的敲除会使得 AMF 的孢子的生长有所限制, 即对 AMF 的无性生殖有所影响, 也干扰 AMF 与宿主的共生。该研究有助于丰富 AM 真菌与玉米共生的理论, 为发掘玉米新品种提供依据。

Abstract 123

Fine mapping of *qLM*, controlling lesion mimic and multiple disease resistance in maize

Hongze Wang¹, Zhibing Lai^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: zhibing@mail.hzau.edu.cn

(submitted by 王宏泽<wh_hongze@qq.com>)

Abstract: Lesion mimic, displaying spontaneous necrotic or chlorotic spots on leaves or stalks without pathogen infection, has been proved to be a kind of programmed cell death and contributing disease resistance in many plant species. However, the molecular mechanisms and genetic variation of lesion mimic in maize are largely unknown. Here, we reported that a teosinte (*sp. Mexicana*)-derived major QTL on chromosome 7: *qLM* contributes lesion mimic phenotype and enhances maize resistance against southern corn rust (SCR) and northern leaf blight (NLB). Through fine mapping, *qLM* has been narrowed down to a 1.1kb region, on the downstream of *ZmMM1*, which induces cell death. Molecular assays indicate that *qLM* functions as a cis-element and suppresses the transcription of *ZmMM1*. Genetic assays showed that two kinds of *qLM* haplotypes *qLM* (-R/-S) exist in the modern maize inbred lines, but *qLM*-teosinte does not; which indicates *qLM*-teosinte was not selected in domestication. The three haplotypes have different functions: *qLM*-S does not induce both of lesion mimic phenotype and disease resistance; *qLM*-R does not contribute lesion mimic phenotype but slightly enhances disease resistance; and *qLM*-teosinte induces strong lesion mimic phenotype and enhances stronger disease resistance. In summary, *ZmMM1* was identified to contribute lesion-mimic phenotype and disease resistance and its transcription is inhibited by the cis element (*qLM*-teosinte) on its 3' terminus.

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Abstract 124

Identification of heterotic loci associated with grain yield and its components using two CSSL test populations in maize

Hongqiu Wang¹, Xiangge Zhang¹, Huili Yang¹, Xiaoyang Liu¹, Huimin Li¹, Liang Yuan², Weihua Li¹, Zhiyuan Fu¹, Jihua Tang¹

¹ Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding author: tangjihua1@163.com

(submitted by 王洪秋<hqwang@cau.edu.cn>)

Abstract: Heterosis has widely been used to increase grain yield and quality. In this study, the genetic basis of heterosis on grain yield and its main components in maize were examined over 2 years in two locations in two test populations constructed from a set of 184 chromosome segment substitution lines (CSSLs) and two inbred lines (Zheng58 and Xun9058). Of the 169 heterotic loci (HL) associated with grain yield and its five components identified in CSSL × Zheng58 and CSSL × Xun9058 test populations, only 25 HL were detected in both populations. The comparison of quantitative trait loci (QTLs) detected in the CSSL population with HL detected in the two test populations revealed that only 15.46% and 17.35% of the HL in the given populations respectively, shared the same chromosomal regions as that of the corresponding QTLs and showed dominant effects as well as pleiotropism with additive and dominant effects. In addition, most of the HL (74.23% and 74.49%) had overdominant effects. These results suggest that overdominance is the main contributor to the effects of heterosis on grain yield and its components in maize, and different HL are associated with heterosis for different traits in different hybrids.

Funding acknowledgement: National Natural Science Foundation of China (91335205), the Major Science and Technology Projects in Henan Province (161100110500), and the State Key Basic Research and Development Plan of China (2014CB138203).

Abstract 125

Research progress and prospect on Crop Phenomics database

Jinglu Wang^{1,2}, Ying Zhang^{1,2}, Xianju Lu^{1,2}, Liming Ma^{1,2}, Xinyu Guo^{1,2*}

¹Beijing Research Center for Information Technology in Agriculture, Beijing Academy of Agriculture and Forestry Sciences, Beijing 100097, China

²Beijing Key Lab of Digital Plant, Beijing 100097, China

*Corresponding author: guoxy@nrcita.org.cn

(submitted by 王璟璐 < wangjl@nrcita.org.cn >)

Abstract: Crop Phenomics has emerged as a fast growing and data intensive discipline. As we know, the phenotypic data are usually multiple dimensional and in different data types. For example, image data, includes RGB, hyperspectral, near-infrared, thermal and fluorescent imaging data. Consequently, the development of biological models and data management systems in this field requires a rational use of these complex, dynamic and large scale phenotypic data. In genomics research field, there were many large, well-recognized and mature public databases. While in Phenomics, there were not enough general-purpose standard databases with strong comprehensiveness and wide universality. According to our search results, there were about 20 studies concerned on database construction, such as Planteome, Plant Genomics and Phenomics Research Data Repository, and OPTIMAS-DW. Therefore, constructed a comprehensive and standard Crop Phenomics database, or built a Phenomics database for a specific crop, would be the research interest of the researchers in this filed. Crop Phenomics database can help researchers better manage their phenotypic data, and it can also benefit the data sharing among researchers.

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Abstract 126

Positional cloning of maize *endosperm breakdown1* mutant

Qun Wang¹, Mingmin Wang², Weiwei Qi², Rentao Song^{1*}

¹ State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

² Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

*Corresponding author: rentaosong@cau.edu.cn

(submitted by 王群<qunwang@cau.edu.cn>)

Abstract: The key difference in seed development between monocots and dicots is the presence of the persistent and transient endosperms, respectively. However, the developmental mechanisms of these two kinds of endosperms remain largely unclear. Here, we identified a novel maize (*Zea mays*) seed mutant, *endosperm breakdown1* (*enb1*), which has a dramatically decreased endosperm. Yet the *enb1* kernels germinated normally, and could develop into normal-appearing and fertile plants, indicating that *enb1* has a fully functional embryo. Cytological observations and Evans blue assays revealed that the *enb1* mutation triggers the precocious endosperm breakdown during kernel development. The *ENB1* was mapped to an interval of 287.90-kb, which contains 8 predicted genes according to the maize B73 RefGen_V4 genome. The gene 3 contained a single nucleotide change in the exon of *enb1* allele, resulting in an amino acid substitution, whereas other genes had no sequence differences between the wild type (WT) and *enb1* alleles. The homozygous *enb1* kernels carrying the transgenic gene 3 were recovered to the WT phenotype, while the homozygous *enb1* kernels without the transgene were remained with the mutant phenotype. The result demonstrated that the transformed gene 3 complemented the *enb1* mutation, and the gene 3 is the *ENB1*.

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Abstract 127

Genome-wide identification and expression analysis of Dof (DNA binding with one finger) protein family in monocot and dicot species

Shuai Wang¹, Pinghua Li^{1*}, Xianglan Wang^{2*}

¹ State Key Laboratory of Crop Biology, College of Agronomy, Shandong Agricultural University, NO. 61 Daizong Street, Tai'an 271018, China

*Corresponding author: pinghuali@sdaa.edu.cn; chizizhanshen@163.com

(submitted by 王帅<18763802276@163.com>)

Abstract: Dof (DNA binding with one finger) proteins are widely associated with kinds of biological processes in plant development. Although Dof proteins were characterized in many species, little was known about the phylogenetic and collinear relationships, expression patterns among monocot and dicot species. In this study, a genome-wide analysis of Dof proteins in 7 monocot and 4 dicot species was performed systematically. 392 Dof proteins were identified and classified into 11 district groups based on phylogenetic analysis. Interestingly, the proteins identified from monocots clustered separately from those of dicots. The motif and gene structure analysis suggested that each subfamily has alike conserved motifs and similar exon/intron compositions. In addition, different motifs among groups might indicate different functions. Multiple collinearity analysis and expression patterns in 3 monocot and 2 dicot species illustrated that most of the ortholog pairs shared similar expression patterns. Functional mode diagram and expression patterns showed that Dof proteins' functions were generally associated with their expression patterns. This study offered novel insights in the phylogenetic, collinear, expressional and functional analysis of Dof proteins and contributed to the further investigation of Dof proteins in plants.

Funding acknowledgement: This work was supported by the NSFC (31271393) and National Key Research and Development Program of China.

Abstract 128

Dynamic plant height QTL revealed in maize through remote sensing phenotyping using a highthroughput unmanned aerial vehicle (UAV)

Xiaqing Wang¹, Ruyang Zhang¹, Wei Song¹, Liang Han^{2,3}, Xiaolei Liu⁴, Xuan Sun¹, Meijie Luo¹, Kuan Chen¹, Yunxia Zhang¹, Hao Yang², Guijun Yang², Yanxin Zhao^{1*}, Jiuran Zhao^{1*}

¹Beijing Key Laboratory of Maize DNA Fingerprinting and Molecular Breeding, Maize Research Center, Beijing Academy of Agriculture & Forestry Sciences, Beijing 100097, China; ²Key Laboratory of Quantitative Remote Sensing in Agriculture of Ministry of Agriculture, Beijing Research Center for Information Technology in Agriculture, Beijing 100097, China; ³College of Architecture and Geomatics Engineering, Shanxi Datong University, Datong 037009, China; ⁴Key Laboratory of Agricultural Animal Genetics, Breeding and Reproduction, Ministry of Education, College of Animal Science and Technology, Huazhong Agricultural University, Wuhan 430070, China.

*Corresponding author: Yanxin Zhao, rentlang2003@163.com; Jiuran Zhao, maizezhao@126.com (submitted by 王夏青<xiaqingwang427@163.com>)

Abstract: Plant height (PH) is a key factor in maize (*Zea mays* L.) yield, biomass, and plant architecture. We investigated the PH of diverse maize inbred lines (117 temperate lines, 135 tropical lines) at four growth stages using unmanned aerial vehicle high-throughput phenotypic platforms (UAV-HTPPs). We extracted PH data using an automated pipeline based on crop surface models and orthomosaic model. The correlation between UAV and manually measured PH data reached 0.95. Under temperate field conditions, temperate maize lines grew faster than tropical maize lines at early growth stages, but tropical lines grew faster at later growth stages and ultimately became taller than temperate lines. A genome-wide association study identified 68 unique quantitative trait loci (QTLs) for seven PH-related traits, and 35% of the QTLs coincided with those previously reported to control PH. Generally, different QTLs controlled PH at different growth stages, but eight QTLs simultaneously controlled PH and growth rate at multiple growth stages. Based on gene annotations and expression profiles, we identified candidate genes controlling PH. The PH data collected by the UAV-HTPPs were credible and the genetic mapping power was high. Therefore, UAV-HTPPs have great potential for use in studies on PH.

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Abstract 129

Fine mapping of *opaque endosperm 5 (oe5)*, a gene controlling amino acid content in maize

Yikai Wang¹, Xiao Zhang¹, Ling Wu¹, Dan Liu¹, Shibin Gao^{1*}

¹Maize Research Institute, Sichuan Agricultural University, 611130, Sichuan, Chengdu, China, and

²State Key Laboratory of Crop Genetics of Disease Resistance and Disease Control, Sichuan, Chengdu, China

*Corresponding author: shibingao@163.com

(submitted by 王怡凯<532274482@qq.com>)

Abstract: The opaque endosperm mutants are important for maize quality. The opaque endosperm 5 mutant (*oe5*) arose from a spontaneous mutation in WT and has a starchy endosperm of kernel at mature stage. There is no vitreous endosperm in *oe5* kernels compared to WT kernels, and the plant height of *oe5* was lower than that of WT. Scanning electron microscopy analysis revealed that the filling material in *oe5* starch grains is loose. Transmission electron microscopy analysis revealed that in the mutant endosperm, there is a reduction 39% in protein body numbers. Biochemical analysis indicated, that the amylose, amylopectin, total lipid, total protein and zein protein contents of *oe5* endosperm showed significantly decrease compared to WT (~ 19.3%, ~ 10.4% less for amylose and amylopectin, ~ 43.2% less for total protein and ~ 71.3% less for total lipid). Moreover, there is no obvious differences in content of nonzein protein between *oe5* and WT kernels. The *oe5* endosperm showed higher total amino acid content for most amino acids, and asparagine content of *oe5* was 1 higher than that of WT and lysine increase 2.5 times, but cysteine and β amino isobutyric acid was not found in WT. Genetic analysis indicated that *oe5* is a single recessive mutant. For fine mapping *oe5*, we constructed a F2 population including 2000 mutant lines, the *oe5* gene was genetically mapped to a 379kb genomic interval on chromosome 5. There are 10 predicted genes located in this interval.

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Abstract 130

Updating and interaction of polycomb repressive complex2 components in maize (*Zea mays*)

Jiacheng Ni¹, Xuexia Ma¹, Yu Feng¹, Qiuzhen Tian², Yongyan Wang², Ningkun Xu², Jihua Tang² and Guifeng Wang^{2*}

¹ Shanghai Key Laboratory of Bio-Energy Crops, School of Life Sciences, Shanghai University, Shanghai 200444, China;

² Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China.

*Corresponding author: guifeng.wang@henau.edu.cn

(submitted by Jiacheng Ni <867373597@qq.com>)

Abstract: The evolutionarily conserved polycomb group (PcG) proteins form multi-subunits polycomb repressive complexes (PRCs) that repress gene expression via chromatin condensation. In Arabidopsis, three distinct PRC2s have been identified, each determining a specific developmental program. However, the core components and biological functions of PRC2 in cereals remain obscure. Here we updated the information on maize PRC2 at a genome-wide scale. Maize PRC2 subunits are highly duplicated, with four *MSI1*, three *E(z)*, two *ESC* and two *Su(z)12* homologs. *ZmFIE1* is preferentially expressed in seeds whereas the other members are broadly expressed in many tissues. *MEZ1* and *ZmFIE1* are maternally expressed imprinted genes, in contrast to a paternal allele-preferential expression pattern of *ZmFIE2*. In maize, *E(z)* members likely provide a scaffold for assembling PRC2 complexes whereas *Su(z)12* and *p55/MSI1*-like proteins together reinforce the complex; *ESC* members probably determine its specificity: FIE1-PRC2 regulates seed development while FIE2-PRC2 controls vegetative development. Moreover, the duplicated Brassicaceae-specific *MEA* and *FIS2* also directly interact with maize PRC2 members. Together, this study establishes a roadmap for protein interactions of maize PRC2 components, providing new insights into their functions in the vegetative and reproductive development of cereals.

Funding acknowledgement: National Natural Sciences Foundation of China (31771800, U1804235), Outstanding Youth Foundation of Henan Province (2018JQ0018), and Shanghai Pujiang Talent Program (17PJD014).

Abstract 131

Three-dimensional shoot architecture evaluation of different maize varieties at early growth stage

Yongjian Wang^{1,2}, Weiliang Wen^{1,2}, Xinyu Guo^{1,2*}

¹ China National Engineering Research Center for Information Technology in agriculture (NERCITA)

² Beijing Research Center for Information Technology in Agriculture, Beijing 100097, China

*Corresponding author: guoxy@nercita.org.cn

(submitted by 王勇健<wyongjian1990@163.com>)

Abstract: The maize shoot architect at early growth stage was increasing interest in both phenotype identification and precise management while it was difficult to achieve accurate measurement and evaluation. Therefore, three-dimensional (3D) shoot architecture measurement and evaluation was needed. At present, few parameters can be used to evaluate plant 3D architecture. We proposed an extracting method of 3D parameters based on 3D shoot architecture models and a daily light interception evaluation model using these 3D parameters. The 3D shoot architecture models of four maize hybrids at early growth stage were reconstructed by 3D digitized data in phytomer scale. Ten parameters of 3D shoot architecture (leaf height, leaf top height, leaf tip height, leaf span and so on) were extracted from 3D models. Six of the ten parameters with significant difference among varieties were selected for Multivariate Linear Regression of daily light interception. The results showed that there were more significant differences of maize shoot architecture (leaf span, leaf tip height, leaf top height) on early growth stage in 3D. The calculated daily light interception was compared with regressions ($R^2 = 0.948$). The parameters that had the highly influence on light interception were leaf area (positive correlation) and leaf tip height (negative correlation). The daily light interception of four varieties at early growth stage was JK968 > ZD658 > DMY2 > AD268. The evaluation method can help breeders optimize maize shoot architecture and precise management under whole growth stages.

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Abstract 132

Analysis of cytology and expression of resistance genes in maize infected with *Sporisorium reilianum*

Fengkun Qi¹, Lin Zhang¹, Xiaojie Dong¹, Hong Di¹, Jiayue Zhang¹, Minhao Yao¹, Ling Dong¹, Xing Zeng¹, Xianjun Liu¹, Zhenhua Wang^{1*}, Yu Zhou^{1*}

¹ Northeast Agricultural University, Harbin, Heilongjiang Province, 150030, China

* Corresponding authors: zhouyu0924@126.com and zhenhuawang_2006@163.com (submitted by 王振华 < zhenhuawang_2006@163.com >)

Abstract: Head smut, caused by the fungus *Sporisorium reilianum*, is a devastating global disease of maize (*Zea mays* L.) that leads to severe degradation of grain quality and yield loss each year. The goal of this research was to study the *S. reilianum* infection process in different maize varieties inoculated using different methods. In the present study, maize seedlings were artificially inoculated with compatible mating-type strains of *S. reilianum* by Needle Inoculation of Mesocotyls (NIM) or by Soaking Inoculation of Radicles (SIR). After NIM or SIR, Huangzao4 mesocotyls exhibited severe damage with brownish discoloration and necrosis, whereas Mo17 mesocotyls exhibited few lesions. Fluorescence microscopy and electron microscopy showed that *S. reilianum* infected maize within 0.5 d after SIR and mainly colonized the phloem. With longer incubation, the density of *S. reilianum* hyphae increased in the vascular bundles, concentrated mainly in the phloem. Mo17 and Huangzao4 infected with *S. reilianum* were compared cytologically. In Mo17, the growth of *S. reilianum* hyphae was severely inhibited relative to that in the susceptible inbred line Huangzao4. In Mo17, infected cells exhibited apoptosis-like features, and hyphae became sequestered within dead cells. In contrast, in Huangzao4 pathogen invasion resulted in autophagy that failed to prevent hyphal spreading. The expression of disease-resistance (*R*) genes in head smut-resistant maize inbred line Mo17 and susceptible inbred line Huangzao4, were analyzed using quantitative Real-Time PCR (qRT-PCR) before and after inoculation. The growth of *S. reilianum* hyphae diminished at 6 d after inoculation when expression of the *R* genes *ZmWAK* and *ZmNL* peaked. Thus, 6 d after SIR inoculation might be an important time point for inhibiting the progress of *S. reilianum* infection in maize. The results of this study will provide a basis for further analysis of the mechanisms of maize resistance to *S. reilianum*.

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Abstract 133

***APP1* Encoding a P-Loop NTPase Is Involved in Maize Kernel Development**

Yi Ming Wei¹, Bo Hui Wang¹, Ya Jie Zhao, Xian Sheng Zhang^{1*}, Xiang Yu Zhao^{1*}

¹ State Key Laboratory of Crop Biology, College of Life Sciences, Shandong Agricultural University, Taian, 271000, China

¹ These authors contribute equally to this work.

*Correspondence: Xiang Yu Zhao (zhxy@sdaa.edu.cn), Xian Sheng Zhang (zhangxs@sdaa.edu.cn)

(submitted by 赵翔宇 < zhxy@sdaa.edu.cn >)

Abstract: Mitochondria is a semi-autonomous organelle that provides the energy for the activities of cells through oxidative phosphorylation. Here, we report the identification of maize *app1* mutant with defective kernels. *APP1* encodes a p-loop NTPase protein that belong to DAR GTPase family. This gene was constitutively expressed in all detected tissues and the highest transcripts level was detected in early development kernels. To determine the subcellular localization of APP1, transient expression experiments in tobacco leaves and maize mesophyll cells using a fused construct p35S: APP1-GFP demonstrated that GFP signal was colocalized with the mitochondrial probe, Mito Tracker Red, indicating a main mitochondria localization of APP1. Further analysis shows that more hydrogen peroxide and superoxide were accumulated in *app1* mutant endosperm indicated by DAB and NBT staining, respectively. To further explore the mechanisms involved in the altered ROS level in *app1* mutant, the abundance and activities of mitochondrial complexes in wild type and *app1* was analyzed. The results indicated that the abundance and activities of complex I were declined in the *app1* development kernels. These results suggest that the APP1 is essential for the development of maize kernels through affecting mitochondria function.

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Abstract 134

玉米 *ZmLBD1* 响应缺磷胁迫和调节根系生长的功能研究

吴锋锴^{1,2}, 刘玲^{1,2}, 熊静^{1,2}, Baba Salifu^{1,2}, 李静^{1,2}, 徐洁^{1,2}, 卢艳丽^{1,2*}

¹四川农业大学, 温江, 611130

²农业部西南玉米生物学与遗传育种重点实验室, 温江, 611130

*通讯作者: yanli.lu82@hotmail.com

(submitted by 吴锋锴 <wfk0909@163.com>)

摘要: 植物转录因子 LBD 家族在植物生长发育中具有重要的调控作用, 但其响应逆境胁迫的相关研究鲜有报道。本研究在 301 份玉米自交系组成的自然群体中检测 *ZmLBD1* 的序列多态性, 发现其完整基因结构的 DNA 序列中包含 32 个 SNP 和 40 个 InDel, 外显子中的多态性最低 (核苷酸多态性 $\pi=2.16\times 10^{-3}$)。中性检验发现 *ZmLBD1* 在驯化过程中受到强烈的纯化选择作用。利用一般线性模型 (GLM)、GLM+Q 和 MLM, 在 *ZmLBD1* 中分别鉴定到 87、32 和 24 个显著的 SNP-玉米苗期缺磷相关性状间的关联 ($P\leq 0.01$)。在 MLM 模型中, 检测到 6 个位点在缺磷胁迫条件下同时与根尖数 (RT) 和根冠比 (RSR) 显著关联, 最高可解释根冠比 4.52% 的表型变异。玉米原生质体中的瞬时表达和拟南芥稳定表达发现 *ZmLBD1* 定位于细胞核。实时荧光定量 PCR 检测表明, *ZmLBD1* 在玉米耐低磷自交系 178 根系中受缺磷胁迫显著诱导表达, 其表达量上调了约 10 倍。拟南芥中过量表达和突变体功能互补分析发现, *ZmLBD1* 具有促进侧根发育和初生根生长的功能, 在缺磷胁迫条件下的差异更加显著。拟南芥过表达株系中的磷积累显著高于野生型, *ZmLBD1* 蛋白的积累量随着缺磷胁迫时间的延长而增加。在玉米 EMS 突变体中, 无义突变体 *ZmLBD1*^{TAG} 幼苗期的总根长、根表面积和根尖数等根系相关性状均显著低于对照基因型。酵母双杂交文库筛选实验以及互作蛋白的 split-LUC 和 Co-IP 验证表明, *ZmLBD1* 与延展素蛋白 *ZmEXP7* 存在相互作用, 可能与促进根系生长发育有关。综上研究结果表明, *ZmLBD1* 在根系中响应缺磷胁迫被诱导表达, 有助于磷的吸收和根系的发育和生长, 为开展耐低磷分子育种奠定了理论基础。

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Abstract 135

Comparative proteomics combined with analyses of transgenic plants reveal *ZmREM1.3* mediates maize resistance to southern corn rust

Shunxi Wang^{1,2}, Zan Chen^{1,2}, Lei Tian^{1,2}, Yezhang Ding³, Jun Zhang⁴, Jinlong Zhou^{1,2}, Ping Liu^{1,2}, Yanhui Chen^{1,2}, Liuji Wu^{1,2*}

1 Henan Agricultural University and Synergetic Innovation Center of Henan Grain Crops, Zhengzhou 450002, China

2 Key Laboratory of Physiological Ecology and Genetic Improvement of Food Crops in Henan Province, Zhengzhou 450002, China

3 Section of Cell and Developmental Biology, University of California at San Diego, La Jolla, CA 92093-0380, USA

4 Cereal Crop Research Institute, Henan Academy of Agricultural Sciences, Zhengzhou 450002, China

*Corresponding author: wlj200120@163.com

(Submitted by 吴刘记<wlj200120@163.com>)

Abstract: Southern corn rust (SCR), which is a destructive disease caused by *Puccinia polysora* Undrew. (*P. polysora*), commonly occurs in warm-temperate and tropical regions. To identify candidate proteins related to SCR resistance and characterize the molecular mechanisms underlying the maize-*P. polysora* interaction, a comparative proteomic analysis of susceptible and resistant maize lines was performed. Statistical analyses revealed 1,489 differentially abundant proteins in the resistant line, as well as 1,035 differentially abundant proteins in the susceptible line. After the *P. polysora* infection, the abundance of one remorin protein (*ZmREM1.3*), increased in the resistant genotype, but decreased in the susceptible genotype. Plant-specific remorins are important for responses to microbial infections as well as plant signaling processes. In this study, transgenic maize plants overexpressing *ZmREM1.3* exhibited enhanced resistance to the biotrophic *P. polysora*. In contrast, homozygous *ZmREM1.3* UniformMu mutant plants were significantly more susceptible to *P. polysora* than wild-type plants. Additionally, the *ZmREM1.3*-overexpressing plants accumulated more salicylic acid (SA) and jasmonic acid (JA). Moreover, the expression levels of defense-related genes were higher in *ZmREM1.3*-overexpressing maize plants than in non-transgenic control plants in response to the *P. polysora* infection. Overall, our results provide evidence that *ZmREM1.3* positively regulates maize defenses against *P. polysora* likely via SA/JA-mediated defense signaling pathways.

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Abstract 136

Fine mapping of a quantitative trait locus for flowering time in maize

Jinliang Xia, Feng Tian*

National Maize Improvement Center, China Agricultural University, Beijing, China 100193

*Corresponding author: ft55@cau.edu.cn

(Submitted by 夏进良 <jlx@cau.edu.cn>)

Abstract: Flowering time is an important adaptive trait that plays critical roles in growth and development. Although a lot of genes controlling flowering time have been cloned, the underlying molecular mechanisms remain largely unknown. In this study, using a large BC₂S₃ recombinant inbred line (RIL) population derived from a cross between maize and its wild progenitor, teosinte, we performed high-resolution quantitative trait locus (QTL) mapping for flowering time, and fine mapped a flowering time QTL, *qDTP1* using HIF-derived near isogenic lines (NILs). An F₂ population containing 2,269 plants was created by self-fertilizing the HIF family. NIL(maize) and NIL(teosinte) identified using molecular markers were used to validate the effect of *qDTP1*. The results showed that NIL(maize) flowered 4 days earlier than NIL(teosinte) under long-day conditions. Using molecular markers that flank the 3-LOD support interval of *qDTP1*, a total of 71 recombinants were identified from the F₂ population. To more precisely determine the recombination breakpoints, 6 additional markers were developed and used to genotype all recombinants. We further narrowed down *qDTP1* to a 206 kb physical region using 19 recombinant families. Based on the gene annotation of B73 reference genome, there are 14 genes located in the 206kb target region. Our results set important basis of finally cloning and functional characterization of *qDTP1*.

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Abstract 137

硫代谢关键基因 *ZmSO* 调控玉米抗旱性的分子机制

夏宗良*, 武轲, 徐紫薇, 张华, 孙凯乐, 吴建宇

河南农业大学生命科学学院, 河南郑州, 450002

*通讯作者: xiazl08@163.com

(submitted by 夏宗良<xiazl08@163.com>)

摘要: 硫是玉米生长发育所必需的矿质营养元素, 在玉米应对干旱胁迫中起着重要作用。然而玉米硫素代谢关键基因参与干旱胁迫应答的调控机制尚缺乏深入的研究。课题组自 2007 年以来, 以硫同化途径中的亚硫酸氧化酶 (sulfite oxidase, SO) 功能研究为切入点, 综合运用遗传、生化和生理学实验手段, 取得了四方面的进展: (1) 明确了玉米 *ZmSO* 基因组分布特点、基因结构特征和表达特性 (夏宗良等, 中国农业科学, 2009; 夏宗良等, 西北植物学报, 2012); (2) 鉴定了玉米 *ZmSO* 的氧化酶活性及酶动力学特征, 发现其具有较高的催化活性 (Xia et al., PLoS ONE, 2012); (3) 利用农杆菌介导的幼胚/愈伤组织转化技术, 创制了 *ZmSO* 过表达和沉默表达的转基因玉米株系, 研究了其生物学功能。发现 SO 通过亚硫酸盐氧化和 H_2O_2 的清除两个途径, 提高植株对 SO_3^{2-} 的抗性反应机制 (Xia et al., PLoS ONE, 2012; Xia et al., PMBR, 2015); (4) 对 *ZmSO* 过表达和沉默表达的转基因玉米株系进行抗旱性鉴定, 发现过表达 *ZmSO* 显著提高了植株的抗旱能力, 而抑制表达 *ZmSO* 的玉米转基因株系对干旱较敏感。进一步分析发现过表达 *ZmSO* 显著提高了植株的硫素利用效率, 增加了 Cys 和 GSH 的水平, 从而提高了耐旱性 (Xia et al., Front Plant Sci, 2018)。这些结果充分表明 *ZmSO* 通过影响硫素代谢物水平调控玉米的抗旱性。

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Abstract 138

The effects of extreme temperatures on vitamins and their biosynthesis pathways in sweet corn (*Zea mays* L.) seedlings

Nan Xiang¹, Jianguang Hu², Tianxiang Wen², Xinbo Guo^{1, *}

¹School of Food Science and Engineering, South China University of Technology; Overseas Expertise Introduction Center for Discipline Innovation of Food Nutrition and Human Health (111 Center), Guangzhou, 510640, China

²Crop Research Institute, Guangdong Academy of Agricultural Sciences; Key Laboratory of Crops Genetics Improvement of Guangdong Province, Guangzhou, 510640, China

*Corresponding author: guoxinbo@scut.edu.cn

(submitted by 向楠<937293470@qq.com>)

Abstract: Extreme temperatures have impacts on the stability and activity of cells thus limit plant development and growth. Maize, supposed to be lack of adaption to unsatisfactory environment, is sensitive to temperature at seedling stage thus its growth is easily being limited under temperature stress. In order to investigate the effect on extreme temperature on the content of vitamin C, E, folates and carotenoids and expression of relative gene in biosynthesis pathway, high (40 °C) and low (10 °C) temperature stresses to sweet corn seedlings for 30 hours were applied. The results indicated that: 1. Low temperature stress limited the expression of relative gene in recycling pathway of ascorbic acid. Applying low temperature for long time (30 °C) may extremely decrease the content of ascorbic acid while high temperature raised it due to the low energy reserves. 2. Low temperature increased the content of 5-MTHF, which belongs to folates derivatives and indirectly participates in DNA methylation. The variation of 5-MTHF under low temperature stress was correlated to the relative gene expression. 3. Low temperature also raised vitamin E content and changes were correlated with relative gene expression. Applying high temperature for 30 hours increased the expression of relative gene thus raised accumulation in γ - and α -tocotrienol. 4. Low temperature stress accelerated the conversion of zeaxanthin, resulting a lower carotenoids content. With the increasing treated time, the content of carotenoids as well as the expression of relative gene were increased under high temperature stress. Therefore, the study identified the variations of vitamins and their biosynthesis pathway in sweet corn seedlings growing under extreme temperature stresses, which is an advance for exploring the biosynthesis regulative mechanism of sweet corn seedlings under stress and a guidance for cultivating sweet corn in unsatisfactory environment.

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Abstract 139

Genetic basis and prediction for 20 agronomic traits from inbred and hybrid populations in maize

Yingjie Xiao^{1,2}, Zhao-Bang Zeng^{2*} and Jianbing Yan^{1*}

¹National Key Laboratory of Crop Genetic and breeding, Huazhong Agricultural University, Wuhan 430070, China

²Bioinformatics Research Center, Department of Horticultural Science, North Carolina State University, Raleigh 27607, USA

Correspondence: Jianbing Yan (yjianbing@mail.hzau.edu.cn) and Zhao-Bang Zeng (Zeng@statgen.ncsu.edu).

Abstract: Understanding the genetic base of agriculturally important traits is vital an informed decision in breeding. In maize, the hybrid varieties phenotypically outperform inbred lines, known as heterosis, however, the underlying genetic relevance remains unclear. We established two hybrid populations by crossing the maize CUBIC population of 1404 inbred lines with two elite testers. With multiple environmental phenotyping and whole-genome sequencing, multiple methods were used to identify quantitative trait locus (QTL) and epistasis responsible for 20 agronomic traits. A total of hundreds of QTLs was identified, while more than one-half of which were uniquely detected in either inbred or hybrid populations. It might reflect the tremendous variability of phenotype between populations. The majority of detected QTLs showed low effects across the whole-genome, indicating the strong inheritance of polygenic nature. On the other hand, the variation partition of QTL and epistasis varied from trait by trait and between inbred and hybrid populations. The shrinking phenotypic variation from inbred to hybrid populations were illustrated to be due to the decline of heritable variance dominated by epistasis. The results demonstrate a large genetic resource responsible for agricultural traits, revealing the important role of global epistasis on particular traits' heterotic responses, which will help assist the gene mining and breeding designs in maize.

Abstract 140

Genome Editing and Double Fluorescence Proteins Enable Robust Maternal Haploid Induction and Identification in Maize

Le Dong¹, Lina Li¹, Changlin Liu¹, Chenxu Liu², Shuaifeng Geng¹, Xinhai Li¹, Changling Huang¹, Long Mao¹, Shaojiang Chen², Chuanxiao Xie^{1,*}

¹Institute of Crop Science, Chinese Academy of Agricultural Sciences, National Key Facility for Crop Gene Resources and Genetic Improvement, Beijing, 100081 China

²National Maize Improvement Center of China, China Agricultural University, No. 2 Yuan Ming Yuan West Road, Haidian District, Beijing 100193, China

(submitted by 谢传晓 <xiechuanxiao@caas.cn>)

Abstract: Genome editing technologies has paved the way for exciting and novel applications in plant biotechnology. Doubled haploid (DH) technology has significant and valuable advantage over traditional approaches in crop breeding. It is pivotal to establish maternal haploid induction (HI) coupled with a stable and efficient haploid identification (HID) system that can be utilized to create maternal haploid inducer at any genetic background in maize and can also be extended to other important cereal crops. Here we showed that the targeted creation of HI lines by targeting its *ZmMTL/ZmPLA1* gene using CRISPR/Cas9 and then they were then crossed to stacked with a HID tool that carried double-fluorescence-protein (DFP) markers to identify maternal parthenogenesis haploid seeds. Simultaneously, the DFP cassette was stably transformed into the maize variety ZC01 after it had been transiently verified the tissues-preferred expression patterns. The DFP-mediated haploid inducer lines (DHILs), which the DFP cassette transformant were successfully stacked with previous targeting *MTL* mutations, were developed to validate the concept of both maternal induction and robust haploid identification. We found that their HI rate ranged from 4.7% to 11.0% with an average of 7.47%. Furthermore, the DFP cassette was found to be transiently expressed in maturing grains of bread wheat (*Triticumaestivum* L.), rice (*Oryza sativa* L.) and barley (*Hordeum vulgare* L.) in the similar tissue-preferred patterns with maize. The results indicate that this developed HID should also work as essential components of DH for those important cereal crops. In conclusion, we have developed an approach to create haploid inducer with robust HID marker for DH breeding system in maize and, possibly, for other important cereals as well. The CRISPR/Cas9 mediated *ZmMTL* (*ZmPLA1*) target mutation enabled to create maternal haploid inducer. The embryo- and endosperm- specific DFP markers were useful as an effective selection marker for maternal haploid identification for both mature seeds and young embryos. In addition, the system should be robust in open pollination environment. It also offers the potential for other important cereals like wheat, rice and barley which has no efficient HID selection method. With its broad application, this system should significantly contribute to DH breeding and hence yield increase in major cereal crops.

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Abstract 141

Proteomics Analysis of Two Maize Photo-Thermo-Sensitive Tasselless Lines

Jiayun Xing¹, Weiwei Jin³, Zhiping Deng^{2*}, Hongbing Luo^{1*}

¹Hunan Maize Engineering and Technology Center, Hunan Agricultural University, Changsha, China

²Institute of Virology and Biotechnology, Zhejiang Academy of Agricultural Science, Hangzhou, China

³National Maize Improvement Center, China Agricultural University, Beijing, China.

*Corresponding author: hbluo68@163.com; zhipingdeng@126.com

(submitted by 邢嘉韵<serfui@126.com>)

Abstract: The development of male sterility lines in hybrid crops, has made great contribution to crop productivity worldwide. The germplasms of photo-thermo-sensitive genic male sterile (PTGMS) lines have been widely used in hybrids breeding in major crops, such as rice, wheat and maize. While a number of maize PTGMS lines have been characterized, there are few studies on the molecular mechanisms of male sterility of PTGMS in maize at the proteome level. Two novel maize mutant lines I478 and I17, obtained after irradiation, showed tassel deficiency under short-day photoperiod and high temperature conditions during Summer period in Southern China. The two PTGMS lines and their background lines were sowed every 15 days in the field in Hunan Province from March 25th to August 25th in 2018. The two PTGMS lines displayed shortened shoot length compared to their control lines and showed lack of tassel. Leaves from six-leaf stage from these lines were sampled, and TMT reagent-based quantitative proteomic analysis was performed to identify proteins differentially expressed between both PTGMS lines and their control lines, respectively. Over 8000 proteins were identified in the proteomics studies, and statistics and bioinformatics analyses are in progress to reveal the proteome changes associated with both mutations.

Funding acknowledgement: National Natural Science Foundation of China (31671709), National key R&D Program of China (2016YFD0101803)

Abstract 142

***ZmLBD44* interacts with *ZmEXP7* to regulate root development and drought response in maize**

Jing Xiong¹, Xuanjun Feng¹, Fengkai Wu¹, Yanli Lu^{1*}

¹ Maize Research Institute, Sichuan Agricultural University, Wenjiang 611130, Sichuan in china,

*Corresponding author: yanli.lu82@hotmail.com

(submitted by 熊静<xiongjingsmile@163.com>)

Abstract: The LATERAL ORGAN BOUNDARIES DOMAIN (LBD) gene family encodes plant-specific transcription factors that regulate diverse processes such as development and stress responses. However, few of these transcription factors have ever been functionally characterized in maize (*Zea mays*). In this study, the LBD transcription factor *ZmLBD44* from maize, which is comprised of LOB domain and PKc-like Superfamily (PKc) domain, was phylogenetically characterized. The results indicate there is no another similar homologous gene both harboring LOB and PKc domains in maize and the closely related species. *ZmLBD44* was differentially expressed in various organs of maize and was induced by the treatments of low phosphorus, drought, indoleacetic acid (IAA), jasmonic acid (JA), salicylic acid (SA), as well as abscisic acid (ABA) in seedlings. The protein expression level of *ZmLBD44* increased with the treatment of JA while decreased with SA. *ZmLBD44* interacted with expansin7 (EXP7) and Cysteine/Histidine-rich C1 domain family protein revealed by Yeast two-hybrid assay. Interestingly, the interaction between EXP7 and full length of LBD44 is stronger than that between EXP7 and LBD44-C (LOB) domain, while no interaction was detected between EXP7 and the LBD44-N (PKc) domain, suggesting that the PKc domain may promote the interaction between LBD44-C and EXP7. In addition, root, especially seminal root of the *lbd44* mutant, with disabled LOB domain, was prominently less and shorter than that of wild type in seedlings and the adult plant of mutant was remarkably dwarf comparing to wild type. Meanwhile, the *lbd44* mutant was sensitive to drought, indicating the positive regulation of LBD44 on drought resistance. Furthermore, β -Glucuronidase (GUS) was driven by the promoter of *ZmLBD44* and expressed in *Arabidopsis*, and the activity of promoter was high in root tips and lateral roots based on histochemical stain. The protein localization was determined by conducting subcellular localization analysis, and we found *ZmLBD44* was different from canonical LBD protein and located in the cytoplasmic, nucleus and plasma membrane, implying the possibility of new function of *ZmLBD44*.

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Abstract 143

Gene expression and activity analyses of two terpene synthases responsible for biosynthesis of aroma compounds in *Osmanthus fragrans*

Rui Xiong, Yan Xiang*

Laboratory of Modern Biotechnology, School of Forestry and Landscape Architecture, Anhui Agricultural University, Hefei, 230036, China.

Abstract: Terpenoids are one of the main components of aroma in plants. In this study, we investigated these compounds in osmanthus (*Osmanthus fragrans* Lour.), a plant widely used in essential oils because of its fragrance. Through the Quantitative Real-time PCR results of enzymes associated with the 2-C-methylerythritol-4-phosphate pathway (MEP), the terpene synthase (TPS) is considered to be key enzymes which is responsible for terpenoid synthesis in osmanthus. In a series of experiments, we identified the TPS genes in osmanthus and revealed the underlying molecular mechanism. Because no genomic reference library exists for osmanthus, we sequenced and analyzed its transcriptome and identified two putative TPS genes, *OfTPS1* and *OfTPS2*. According to qRT-PCR analysis, the highest expression levels of both genes were at the full-bloom stage, thereby further suggesting that *OfTPS1* and *OfTPS2* are associated with osmanthus terpenoid synthesis. To verify this hypothesis, we constructed prokaryotic expression vectors and detected enzyme activity. This experiment revealed that *OfTPS1* and *OfTPS2* react with geranyl pyrophosphate ammonium salt (GPP) to produce (E)- β -ocimene and linalool, respectively. *OfTPS1* and *OfTPS2* are thus both monoterpene synthases.

Keywords: *Osmanthus fragrans*, Terpene synthase, qRT-PCR, Transcriptome, Prokaryotic expression

Abstract 144

Preliminarily analyzing the molecular mechanism of maize heterosis formation by high-throughput yeast two hybrid

Zimo Xiong, Jia Qian, Lin Li*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: hzaulilin@mail.hzau.edu.cn

(submitted by Zimo Xiong <2994270597@qq.com>)

Abstract: Heterosis is one of the most important biological phenomena, that is widely utilized in maize production. Understanding the molecular mechanism of heterosis will help to better apply heterosis in maize breeding. In this study, we have extracted mRNA of V4-stages leaves from B73, Mo17 and its hybrid offspring F1, and conducted reverse-transcribe experiment and constructed six cDNA libraries, which include as many transcripts as possible. Among them, about 10756 and 10354 transcripts were detected in the cDNA-AD library and cDNA-BD library of Mo17. Based on the high-throughput yeast two hybrid technology combined with next generation sequencing technology, we assembled a protein-protein interaction landscape of B73, Mo17 and F1 at the protein level for the V4-stage leaves, and found differentially expressed proteins between parents and hybrids, further characterized the differentially expressed protein-protein interactions associated with heterosis.

Abstract 145

EMB15 functions in plastid 30S ribosome assembly and embryogenesis in maize

Chunhui Xu¹, Yun Shen², Mengdi Zhang¹, Cuiling Li¹, Robert Meeley³, Donald R. McCarty⁴, Bao-Cai Tan^{*1}

¹ Key Laboratory of Plant Development and Environmental Adaption, Ministry of Education, School of Life Sciences, Shandong University, Qingdao, China. ² State Key Lab of Agrobiotechnology, Institute of Plant Molecular Biology and Agrobiotechnology, School of Life Science, The Chinese University of Hong Kong, N.T. Hong Kong. ³ DuPont Pioneer AgBiotech Research, Johnston, Iowa 50131-1004, USA. ⁴ Horticultural Sciences Department, University of Florida, Gainesville, FL 32611, USA

* For correspondence (e-mail bctan@sdu.edu.cn)

Abstract: The embryo defective (*emb*) mutants account for a major group of seed mutants in maize which display specifically arrested embryogenesis with normal endosperm development. Here we report the cloning and functional characterization of *Emb15*. The *emb15* mutant in W22 background displays *emb* phenotypes. *Emb15* was cloned by transposon-tagging and confirmed by multiple alleles. EMB15 contains an N-terminus domain with high similarity to prokaryotic RimM protein and a C-terminus domain with high similarity to UDP-GlcNAc pyrophosphorylases (UAP). *Emb15* appears to derive from fusion of two genes as algae and lower species host the two domains in two separate proteins. The RimM protein in *Escherichia coli* is implicated in assembly of 30S ribosome and is essential for growth. UAP is considered to catalyze a reversible reaction of UTP and GlcNAc to PPi and UDPGlcNAc, the precursor of N- and O-linked glycosylation. EMB15 was localized in the chloroplast. Expression of *Emb15* restores the growth of *E. coli* Δ rimM mutant. Y2H experiment suggests EMB15 interacts with chloroplast ribosome small subunit protein. Arabidopsis *AtEmb15* mutants created with CRISPR/Cas9 show shorter roots, smaller rosette leaves and dwarf phenotypes. These results suggest that EMB15 has a similar function of RimM in facilitating plastid 30S ribosome assembly. The function of UAP domain is under study.

Funding Acknowledgement: This research was supported by National Natural Science Foundation of China (91435201, 31630053) and Natural Science Foundation of Shandong Province, China (ZR2015CM031).

Abstract 146

The CLAVATA receptor *FASCIATED EAR2* functions through both heterotrimeric G-protein α and β subunit in meristem development

Fang Xu, Qingyu Wu, Lei Liu, Byoung Il Je, David Jackson *

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, USA

*Corresponding author: jacksond@cshl.edu

(submitted by 徐芳 <fxu@cshl.edu>)

Abstract: Plant survival depends on pools of stem cells, called meristems, which are maintained by a carefully orchestrated feedback between *CLAVATA* (*CLV*) and *WUSCHEL* (*WUS*) signaling. *CLV* signaling involves the secretion of the *CLV3* peptide and its perception by a number of Leucine-Rich-Repeat (LRR) receptors, including the receptor-like kinase *CLV1* and the receptor-like protein *CLV2*. The well-known *CLV-WUS* model has been established for more than a decade, however signaling events downstream of the *CLV* receptors remains largely a mystery. The alpha subunit of the maize heterotrimeric G protein *COMPACT PLANT2* (*CT2*) was previously shown as a downstream signaling component for *FASCIATED EAR2* (*FEA2*), the maize ortholog for *CLV2*. Here, we identified an additional player in this pathway through studying on a new fasciated ear mutant *fea*148*, which exhibited enlarged shoot apical meristem and inflorescence meristem as well as lesions on leaves caused by cell death. Map-based cloning combined with whole genome sequencing revealed a single amino acid change in the maize heterotrimeric G-protein β subunit (*ZmGB1*). The single amino acid mutation in *Zmgb1*^{*fea*148*} abolished its ability to form complex with *CT2*, which leads to the loss of function in *FEA2* signaling. Genetic analysis reveals that *ZmGB1* function together with *CT2* in inflorescence meristem while they function additively in shoot apical meristem, suggesting a diverse function mechanism for G protein complex. In addition, autoimmunity is detected in *Zmgb1*^{*fea*148*} with upregulation of defense marker gene, *PATHOGENESIS-RELATED 1* (*PR1*) and *PR5*. Our study thus suggest that maize heterotrimeric G-proteins play a dual role in controlling meristem size during meristem development and immune responses. Taken together, *ZmGB1* is a good candidate for fine-manipulation to optimize the tradeoff between yield and disease resistance.

Abstract 147

Genome-wide association study reveals the causes and consequences of natural variation in DNA methylation in maize

Jing Xu^{a,1}, Guo Chen^{a,1}, Peter J. Hermanson^b, Qiang Xu^a, Changsuo Sun^a, Wenqing Chen^a, Qiuxin Kan^a, Minqi Li^a, Peter A. Crisp^b, Jianbing Yan^a, Lin Li^a, Nathan M. Springer^{b,*}, Qing Li^{a,*}

^a National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

^b Department of Plant and Microbial Biology, University of Minnesota, St. Paul, MN 55108, USA

¹ Contributed equally

* Co-corresponding author

(submitted by 徐静 <xujing199506@qq.com>)

Abstract: DNA methylation can provide heritable information that is either coupled or uncoupled with genetic variations. Variation in DNA methylation plays important roles in shaping diversity of both gene expression and phenotypes. Here we investigated the genetic basis and biological functions of DNA methylation at a population scale in maize. DNA methylation was profiled across a diverse panel of 263 maize inbred genotypes. All genotypes show similar levels of DNA methylation, highlighting the importance of DNA methylation in maize development. More than 20,000 differentially methylated regions (DMRs) that are distributed over the 10 maize chromosomes were identified. These DMRs can differentiate maize populations that are defined using SNPs, although there is variation in how strongly different context-specific DMRs can separate sub-populations. Genome-wide association analysis with ~1 million SNPs revealed that > 50% of the DMRs were not tagged by genetic variations, suggesting the presence of unique information in DMRs. Association analysis of the DMRs with the expression levels for > 20,000 genes in both kernel and leaf tissues suggest that DNA methylation variation is associated with the expression of many genes. The direction of effect varies depending on both sequence contexts and the position of DMRs relative to the transcriptional start site. Association analysis with 983 metabolic traits suggests that DNA methylation is associated with phenotypic variations of 156 traits. There are some traits that only show significant associations with DMRs and not with SNPs. These results suggest that DNA methylation contributes to phenotypic diversity in maize and can provide unique information to explain phenotypic variation.

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Abstract 148

Arabidopsis *BRASSINOSTEROID INACTIVATOR2* is a typical BAHD acyltransferase involved in brassinosteroid homeostasis

Zhiqiang Zhang^{1,2}, Liping Xu¹ *

¹ National Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou, 450002, China

² National Key Laboratory of Crop Genetic Improvement, National Center of Rapeseed Improvement, Huazhong Agricultural University, Wuhan, 430070, China

(submitted by 徐莉萍<xulipingqq1@163l.com>)

Abstract: Brassinosteroids (BRs) are plant-specific steroidal hormones; BR homeostasis is crucial for various aspects of plant growth and development. However, to date, the BR inactivation process has not been thoroughly elucidated. In this study, we identified and characterized a novel BAHD family acyltransferase gene, *BRASSINOSTEROID INACTIVATOR2* (BIA2), involved in BR inactivation. BIA2-overexpressing (OE-BIA2) plants displayed typical BR-deficient phenotypes, which were rescued by exogenous BR treatment. Real-time qRT-PCR and transcriptome analyses showed that expression levels of virtually all of the BR biosynthetic genes were increased, whereas the expression of many BR inactivation genes was reduced in OE-BIA2 plants. Root inhibition assays showed that the root growth of OE-BIA2 plants was inhibited. We obtained plants with an intermediate phenotype by crossing the OE-BIA2 plants with *BRASSINOSTEROID-INSENSITIVE1* (BRI1)-overexpressing plants. The null BIA2 mutants had longer hypocotyls in the dark. BIA2 was predominantly expressed in roots, and its expression was induced by 24-epibrassinolide or dark treatment, but it exhibited a differential expression pattern compared with its homologue, BIA1. Furthermore, genetic transformation with point-mutant and deleted-BIA2 constructs confirmed that the HXXXD motif is essential for the function of BIA2. Taken together, these findings indicate that BIA2 is a typical BAHD acyltransferase that is involved in BR homeostasis and may inactivate bioactive BRs by esterification, particularly in roots and hypocotyls under dark conditions.

Abstract 149

Evolutionary metabolomics identifies significant metabolic divergence between maize and its wild ancestor, teosinte

Guanghui Xu^{1,3}, Jingjing Cao^{1,2,3}, Xufeng Wang¹, Qiuyue Chen¹, Weiwei Jin¹, Zhen Li^{1,*} and Feng Tian^{1,*}

¹State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Key Laboratory of Biology and Genetic Improvement of Maize (MOA), Beijing Key Laboratory of Crop Genetic Improvement, Center for Crop Functional Genomics and Molecular Breeding, China Agricultural University, Beijing 100193, China

²Institute of Plant Protection, Jiangsu Academy of Agricultural Sciences, Nanjing 210014, China

³These authors contributed equally to this work

*Corresponding author: Feng Tian (ft55@cau.edu.cn) and Zhen Li (lizhenchem@cau.edu.cn)

(submitted by 许光辉<gh99@cau.edu.cn>)

Abstract: Maize was domesticated from its wild ancestor, teosinte. Maize's new morphology and adaptation to diverse environments require coordinated changes in metabolic pathways that are essential for growth and development. However, how the metabolome was reshaped since domestication remains poorly understood. Here, we report a comprehensive assessment of divergence in the seedling metabolome between maize and teosinte. We demonstrate that teosinte, tropical maize and temperate maize experienced significant divergences in distinct sets of metabolites due to selection. To identify genetic factors controlling metabolic divergence, we assayed the seedling metabolome of a large maize-by-teosinte cross population. We show that the recent divergence between tropical and temperate maize was associated with more metabolite alterations and controlled by simpler genetic architecture. Using a statistical method that integrates transcriptome data, we identified candidate genes whose expression contributes to the metabolite variation in the maize-teosinte population and found that they are more likely under selection at nucleotide and transcript levels. Through overexpression or mutant analysis, we verified the roles of *FHT*, *Pr1*, and *ZmTPS1* in the divergence of their related biosynthesis pathways. Our findings not only provide important insights into domestication-associated changes in metabolism but also highlight the power of combining omics data for trait dissection.

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Abstract 150

Analysis of maize mutant library based on CRISPR/Cas9

Jiali Yan¹, Maolin Zhang¹, Gang Lu¹, Haijun Liu¹, Liumei Jian¹, Qinghua Zhang¹, Yong Peng¹, Jieting Xu², Yuming Lu², Jianbing Yan^{1,*}

¹ National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

² FOR FUTURE BREEDING Biotechnology (JiangSu) co. LTD., Changzhou 213000, China

* Corresponding author: yjianbing@mail.hzau.edu.cn

(submitted by 严佳丽 <yanjl95@163.com>)

Abstract: Maize is an important staple crop and has served as a powerful model system for genetic studies. Since the availability of the maize inbred B73 reference genome, the major goal of maize research has been to analyze the functions of all maize genes in different environment and physiological periods. Mutagenesis is one of the most effective and widely-used strategies for studying gene function. In the past decades, several maize mutant libraries have been constructed through EMS-induced, mutator-mediated mutagenesis and so on. Usually, however, all the mutations from these libraries are unidirectional and so they have complex background mutations, especially to the former, which makes the gene coverage rate cannot reach 100% and is difficult to identify the targeted mutations for the observed phenotypes. Currently, the applications of targeted genome editing using CRISPR/Cas9 in plants have been tremendously reported and provide the opportunities for us to precisely create saturated mutant library involved each gene. We established a high-throughput platform for genome editing through the CRISPR/Cas9 system in maize and took a group contained totally 1,000 genes as an example which involved all the genes of 50 QTLs gained through GWAS (genome-wide association studies) and QTL mapping involving agronomic traits, flowering and kernel development. The guides were detected by next-generation sequencing and the genotype of each T0 and T1 plant was identified using Sanger sequencing. Here we provide a detailed pipeline for genome-wide precisely targeted mutagenesis in maize. We detect the genotype of targeted genes to reveal and summarize the efficiency and features of the CRISPR/Cas9-based mutations in maize. Our results will help to precisely and quickly create a saturated mutant library involved each gene of maize, promoting the development of functional genomics and the breeding process.

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Abstract 151

A P-type pentatricopeptide repeat protein is involved in the splicing of multiple mitochondrial introns and seed development in maize

Huanhuan Yang¹, Zhihui Xiu¹, Xiulan Li¹, Feng Sun¹, Yiwei Liu¹,
Ruicheng Jiang¹, Baocai Tan¹ *

¹Key Laboratory of Plant Development and Environmental Adaptation Biology, Ministry of Education, School of Life Sciences, Shandong University, Qingdao 266237, China

*Correspondence: Baocai Tan (bctan@sdu.edu.cn)

(submitted by 杨欢欢<yanghuanhuan7@163.com>)

Abstract: Intron splicing is a post-transcriptional process that is pervasive in higher plants. Transcripts in mitochondria and plastids mostly contain group II introns, which require precise splicing by multiple nuclear encoded factors. The mechanism of introns splicing and splicing-specific trans-factors that are implicated in this process are not well understood. Here, we report the identification of *PPR231*, which is involved in the splicing of multiple mitochondrial introns. *PPR231* encodes a mitochondria-targeted P-type pentatricopeptide repeat (PPR) protein that harbors 10 PPR motifs. The loss-of-function in *PPR231* arrests the embryogenesis and endosperm development, generating a *small kernel* phenotype in maize. The mutation of *PPR231* impairs the *cis*-splicing of *nad2* intron 3 and affects the stability of *nad5* intron 1, 2, and 3, resulting in disrupted assembly and reduced activity of complex I. In response, the expression of alternative oxidase *AOX2* and compensation regulation from other complexes are activated. Taken together, these results reveal that *PPR231* is required for the splicing of multiple mitochondrial introns, which is essential to complex I biogenesis and embryogenesis and endosperm development in maize.

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Abstract 152

Genetic Dissection of Haploid Male Fertility in Maize (*Zea mays* L.)

Jiwei Yang¹, Yanzhi Qu¹, Qiong Chen¹, Jihua Tang¹, Thomas Lübberstedt²,
Haochuan Li^{1*}, Zonghua Liu^{1*}

¹College of Agronomy, National Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, Henan Agricultural University, Zhengzhou, China

²Department of Agronomy, Iowa State University, Ames, Iowa, United States of America

Corresponding author: lihaochuan1220@163.com; zhliu100@163.com

(submitted by 杨继伟 yangjiwei2009@126.com)

Abstract: Haploid genome doubling is a key limiting step of haploid breeding in maize. Spontaneous restoration of haploid male fertility (HMF) provides a more promising method than the artificial doubling process. To reveal the genetic basis of HMF, haploids were obtained from the offspring of 285 F_{2:3} families, derived from the cross Zheng58 × K22. The F_{2:3} families were used as the female donor and Yu high inducer No. 1 (YHI-1) as the male inducer line. The rates of HMF from each family line were evaluated at two field sites over two planting seasons. HMF displayed incomplete dominance. Transgressive segregation of haploids from F_{2:3} families was observed relative to haploids derived from the two parents of the mapping population. A total of nine quantitative trait loci (QTL) were detected, which were distributed on chromosomes 1, 3, 4, 7 and 8. Three major QTL, qHMF3b, qHMF7a and qHMF7b were detected in both locations, respectively. These QTL could be useful to predict the ability of spontaneous haploid genome doubling, and to accelerate the haploid breeding process by introgression or aggregation of those QTL.

Funding information: The National Key Research and Development Program of China; The National Natural Science Foundation of China; The Science and technology project of Henan Province.

Abstract 153

玉米草酸降解途径解析和籽粒品质遗传改良

王涛¹, 王琼², 杨俊^{1*}

¹ 作物抗逆育种与减灾国家地方联合工程实验室, 安徽农业大学, 合肥 230036

² 中国科学院分子植物科学卓越创新中心/植物生理生态研究所, 上海 200032

*通讯作者: jyang@ahau.edu.cn

(submitted by 杨俊<jyang@ahau.edu.cn>)

摘要: 草酸在自然界中的分布非常广泛, 部分物种中可占干重的3%—10%。草酸是一种典型的抗营养因子, 影响营养物质的吸收和利用。植物中的同时存在草酸的合成和降解途径, 调控草酸含量从而参与植物金属胁迫和生长发育等过程。一条经典的草酸降解途径由四种酶催化组成, 分别是草酰辅酶A合成酶(Acyl-activating enzyme3, AAE3 / O7)、草酰辅酶A脱羧酶(Oxalyl-CoA decarboxylase1, OCD1)、甲酰辅酶A转移酶(Formyl-CoA transferase, FCT)和甲酸脱氢酶(Formate dehydrogenase, FDH)。我们发现: 玉米高赖氨酸粉质突变体o7编码AAE3蛋白, 参与草酸降解第一步反应, 可以将草酸转变为草酰辅酶A。草酸第二步反应关键酶OCD1能降解化学合成的底物甲酰辅酶A, 而OCD1突变以后的玉米胚乳不能降解甲酰辅酶A。有意思的是, *ocd1*突变体籽粒也呈现粉质胚乳表型; 蛋白和淀粉的合成均受到影响, 草酸含量和重要代谢途径的多个关键中间物含量均发生显著改变。我们期望通过过量表达O7和OCD1两个关键酶, 降低玉米籽粒草酸含量, 提高品质。此外, 我们采用同源比较和组织表达分析等方法初步确定了草酸降解途径第三和第四步反应关键酶(FCT和FDH)的候选编码基因。目前正通过多种策略进行酶活验证, 创制相应遗传材料, 从而揭示玉米FCT和FDH可能的生物学功能。通过对fct/fdh突变体籽粒的草酸含量、胚乳发育、淀粉合成、蛋白合成、代谢组和元素含量等品质相关性状进行测定, 最终完整解析草酸降解途径关键酶的功能并探索其在改善籽粒品质中的潜力。

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Abstract 154

整合关联分析和转录组分析挖掘玉米抗轮枝镰孢菌穗粒腐病基因

姚丽珊, 马传禹, 李艳梅, 徐明良*

1 中国农业大学农学院国家玉米改良中心, 北京, 100193

*通讯作者: mxu@cau.edu.cn

(submitted by 姚丽珊<15117957503@163.com>)

摘要: 以 508 份玉米自交系为材料, 人工接种轮枝镰孢菌, 调查穗粒腐病, 利用 MLM 法进行关联分析。经过连续两年三点 (17 年北京夏季, 17 海南冬季, 18 北京夏季) 试验, 分别找到 6、28 和 7 个与穗粒腐病抗性相关的 SNP, 可以解释 5.15%-11.00% 表型变异。基于 B73 参考基因组, 共获得与这些 SNPs 共定位或相邻的基因 72 个。从上述关联群体中筛选极抗自交系 CIMBL47 和极感自交系 SY1035, 取授粉后 15 天的籽粒, 一分为二, 分别浸泡在无菌水和 $5 \times 10^6/\text{ml}$ 的病原菌孢子悬浮液中五分钟, 紧接着转移至 PDA 培养基上培养。在培养后的 0、0.5、1.5、3 和 6 小时 (hpi) 时间点分别取样, 利用高通量 RNA-seq 技术检测转录组, 比较抗、感材料在接种后不同时间点的差异表达基因。结果表明, 抗病材料 CIMBL47 接种后出现大量差异表达基因, 其中 2/3 为上调表达; 而感病材料 SY1035 差异表达基因较少, 且上调和下调基因数目相当。抗、感材料基因组的本底表达差异较大, 当病原菌接种后, 相比于感病 SY1035, 抗病 CIMBL47 中有大量上调表达基因, 这种趋势在 0.5hpi 达到峰值。这些结果暗示抗病材料含有较多处于“待激活”状态的抗病基因, 一旦检测到外界病原菌, 这些基因被快速激活来抵御入侵。随着侵染时间的延长, 感病材料 SY1035 中的下调表达基因数量上升, 并在 1.5hpi 时达到峰值。聚类分析发现差异表达基因多富集在具有催化活性、结合活性、代谢活性、胁迫相应活性、植物生长发育调控活性等功能途径。结合转录组信息和关联分析, 我们发现关联分析中大约 15% 的抗病候选基因在接种后的抗病材料中高表达, 这些基因多与植物激素信号转导、热激蛋白、植物细胞 P450、苯丙氨酸代谢等相关。研究表明, 整合关联和转录组分析可以有效挖掘玉米抗轮枝镰孢菌的微效数量抗病基因, 从而加快抗性基因的克隆过程。

Abstract 155

The Genetic Architecture of Chlorophyll Content by Linkage and Association Analysis in Maize

Yao Tianlong¹, Shi Dakun¹, Liu Nannan², Deng min³, Duan Haiyang¹, Wang luLin¹, Wan Jiong¹, Gao Jionghao¹, Xie Huiling¹, Tang Jihua¹, Zhang Xuehai^{1*}

¹College of Agronomy, Henan Agricultural University/National Key Laboratory of Wheat and Maize Crop Science, Zhengzhou 450002, China;

²Haixia Institute of Science and Technology, Fujian Agriculture and Forestry University, Fuzhou 350002, China;

³College of Agronomy, Hunan Agricultural University/Maize Engineering Research Center of Hunan Province, Changsha 410128, China.

*Corresponding author: xuehai85@126.com

(submitted by 姚天茏 <643173281@qq.com>)

Abstract: Chlorophyll content is one of the most important physiological traits as it is closely related to leaf photosynthetic efficiency and crop yield potential. Most cloned genes controlling chlorophyll content are from *Arabidopsis* and rice. However, genetic basis of chlorophyll content in maize is still unclear. In this study, the genetic basis of the chlorophyll content in maize was elucidated using an association mapping population (n=538) and two recombinant inbred line populations (RIL₃₃₅ and RIL₉₅₈). A genome-wide association study (GWAS) with three models (Q, K, Q+K) were carried out using 558 629 single nucleotide polymorphisms (SNPs), and K model has the greatest success in reducing false positive than other two models. Based on the result of K model, a total of 18 loci involving in 29 significantly SNP-traits associations were detected ($P \leq 3.99 \times 10^{-6}$), and 76 candidate genes were found. Of which, 85.5% (65/76) of the candidate genes have express QTLs (eQTLs) and 11.8% (9/76) of them were significantly associated with the corresponding phenotype ($P < 0.05$), indicating that these nine genes may affect phenotypic variation by regulating their expression. Moreover, 9 and 8 loci were identified in RIL₃₃₅ and RIL₉₅₈, respectively, two of which were also found in GWAS and two genes (*GRMZM2G168858* and *GRMZM2G371345*) within the two co-located loci may be participated in chlorophyll synthesis and degradation pathways. In details, *GRMZM2G168858* encodes NADPH-cytochrome reductase 2, which is an important reductase of cytochrome P450 family and participates in the catabolism of chlorophyll in leaves. *GRMZM2G371345* encodes flavonol 3-O-glucosyltransferase that as a substrate for UDP-glucose involved in chlorophyll synthesis and degradation pathways. The loci or candidate genes identified would be helpful in ideotype-based maize breeding with high photosynthetic efficiency.

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Abstract 156

ECOGEMS: efficient compression and retrieve of SNP data of 2058 rice accessions with integer sparse matrices

Wen Yao^{1,*}, Fangfang Huang¹, Xuehai Zhang², Jihua Tang^{2,*}

¹National Key Laboratory of Wheat and Maize Crop Science, College of Life Sciences, Henan Agricultural University, Zhengzhou 450002, China

²National Key Laboratory of Wheat and Maize Crop Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding author: tangjihua1@163.com, yaowen@henau.edu.cn

(submitted by yaowen@henau.edu.cn)

Abstract: We proposed to store large-scale genotype data as integer sparse matrices, which consumed much fewer computing resources for storage and analysis than traditional approaches. In addition, the raw genotype data could be readily recovered from integer sparse matrices. Utilizing this approach, we stored the genotype data of 1612 Asian cultivated rice accessions and 446 Asian wild rice accessions across 8,584,244 SNP sites in the ECOGEMS database with 310 MB of disk usage. Graphical interface for visualization, analysis and download of SNP data were implemented in ECOGEMS, which made it a valuable resource for rice functional genomic studies.

Availability and implementation: The code and data of ECOGEMS are freely available at <https://github.com/venyao/ECOGEMS>. ECOGEMS is deployed at <http://ecogems.ncpgr.cn> and <http://150.109.59.144:3838/ECOGEMS/> for online use.

Funding acknowledgement: This research was supported by the key grant science and technique foundation of Henan province (161100110500-0102) and the research start-up fund to top-notch talents of Henan Agricultural University (30500581).

Abstract 157

富含维生素 E 和植酸酶的玉米新材料创制

姚兴兰, 罗彦忠, 王磊, 张兰*

中国农业科学院生物技术研究所 北京 100081

*通讯作者: zhanglan20000@163.com

摘要: 玉米是家禽和单胃动物饲料的主要原料, 但玉米籽粒中高活性的 α -生育酚含量较低, 而且玉米籽粒中的磷主要是以有机磷——植酸磷的形式存在, 动物不能有效吸收利用。因此, 饲料中需要添加合成的 DL- α -生育酚醋酸酯和无机磷来满足动物生长发育的需要, 从而使得饲料成本大大增加; 而且植酸磷还会随动物粪便排出体外造成污染环境。所以, 一种有效的应对方法就是提高玉米籽粒中 α -生育酚的含量, 同时引入可以分解植酸磷的植酸酶。我们以胚特异性表达的启动子 13387 驱动玉米 γ -生育酚甲基转移酶 *ZmTMT* 基因的表达, 以胚乳特异性启动子 123387 驱动植酸酶基因 *phyA2* 的表达, 与抗除草剂基因 *Bar* 基因表达盒采用农杆菌侵染的方法转化玉米, 获得了 ZTPHY 转化体。侧翼序列分析表明, 三个外源表达盒已整合至玉米基因组的非编码区; qRT-PCR 和 western blotting 分析显示, 插入基因在玉米籽粒中高表达; 生育酚含量及植酸酶酶活测定结果表明, 玉米籽粒中 90% 以上的 γ -生育酚转化为了 α -生育酚, 植酸酶的酶活达到了 10000 单位/公斤种子, 已经能够满足动物的需要。此富含维生素 E 和植酸酶的玉米新材料用于玉米杂交种的开发, 可以降低饲料成本, 提高磷的利用率, 减少环境污染。

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Abstract 158

Ascorbic acid integrates the antagonistic modulation of ethylene and abscisic acid in the accumulation of reactive oxygen species

Yanwen Yu^{1,3}, Juan Wang^{1,3}, Shenghui Li^{1,3}, Xiamusiya Kakan¹, Yun Zhou², Yuchen Miao², Fangfang Wang¹, Hua Qin¹, and Rongfeng Huang^{1*}

¹Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing 100081, China

²Institute of Plant Stress Biology, Henan University, Collaborative Innovation Center 20 of Crop Stress Biology, Kaifeng 475001, Henan, China

³These authors contributed equally

* Corresponding author: rfhuang@caas.cn

(submitted by 于延文 yuyanwen1224@126.com)

Abstract: During plant growth and development, ethylene and abscisic acid (ABA) play important roles and exert synergistic or antagonistic effects on various biological processes, but the detailed mechanism underlying the interaction of the two phytohormones, especially in the regulation of the accumulation of reactive oxygen species (ROS), is largely unclear. Here, we report that ethylene inhibits but ABA promotes the accumulation of ROS in *Arabidopsis* (*Arabidopsis thaliana*) seedlings. Furthermore, changes in the biosynthesis of ascorbic acid (AsA) act as a key factor in integrating the interaction of ethylene and ABA in the regulation of ROS levels. We found that ethylene and ABA antagonistically regulate AsA biosynthesis via ETHYLENE-INSENSITIVE 3 (EIN3) and ABA INSENSITIVE 4 (ABI4), which are key factors in the ethylene and ABA signaling pathways, respectively. In addition, ABI4 is transcriptionally repressed by EIN3 in ethylene-regulated AsA biosynthesis. Via transcriptome analysis and molecular and genetic experiments, we identified VITAMIN C DEFECTIVE 2 (VTC2) as the direct target of ABI4 in the regulation of AsA biosynthesis and ROS accumulation. Thus, the EIN3-ABI4-VTC2 transcriptional cascade involves a mechanism by which ethylene and ABA antagonistically regulate AsA biosynthesis and ROS accumulation in response to complex environmental stimuli.

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Abstract 159

Preparation of NAC78 antibody and its expression under salt stress

Shan He¹, Zhikang Wang¹, Yun Qing¹, Guowu Yu^{1*}, Yubi Huang^{1,2*}

¹ Agronomy College, Sichuan Agriculture University, Chengdu, 611130, China

² Maize Research Institute of Sichuan Agriculture University; Chengdu, 611130, China

*Correspondence email: 2002ygw@163.com, yubihuang@sohu.com

(submitted by Guowu Yu<2002ygw@163.com>)

Abstract: Antibody is widely used in protein research of animal and plants. However, it is short of various antibodies to do protein investigation in maize. NAC78 is a novel transcription factor of NAC family. Here we constructed the prokaryotic expression vector pGEX-6t-GST-NAC78 to express GST-NAC78 fusion protein. Though subcutaneous injection of GST-NAC78 fusion protein antigen to New Zealand white rabbit to produce antibody, we successfully prepared NAC78 antibody from the rabbit serum. NAC78 antibody was purified by protein A+G beads from rabbit serum. Western blot was performed to analyze the expression of NAC78 protein in maize endosperm. The result shows that it is not significant difference in different development stages of maize endosperm. In addition, the expression of NAC78 in maize different tissues in mRNA level and NAC78 responding to salt stress were investigated. The results showed that the expression of NAC78 was different in different tissues, and the highest was found in endosperm. Salt treatment was performed at three-leaf stage. The results showed that NAC78 in leaves was induced by salt, and the highest level achieved after 6 hours of salt treatment.

Key words: maize, NAC78, antibody, protein expression

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Abstract 160

Fine mapping and functional analysis of *qDRI-2* for the dehydration rate of maize kernels

Yanhui Yu¹, Wenqiang Li¹, Jianbing Yan^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

(submitted by 余延辉<1414681677@qq.com>)

Abstract: The dehydration rate of maize kernels is one of the important agronomic traits, which would affect maize mechanized harvesting, storage, disease resistance and transportation. It is of great importance to improve the rate of dehydration of maize kernels after physiological maturity. Dehydration rate, which is controlled in a highly complex manner, making it difficult to detect and map dehydration rate related traits. Thus far, although some QTLs responsible for dehydration rate of maize kernels have been identified, but none of genes have been reported. Here, by fine mapping, we want to find and analyze the functions of genes that control the dehydration rate of maize kernels, and elucidate its molecular mechanism. In this study, we used a RIL population derived from the cross between DAN340 and K22 for QTL mapping. In the field, we arranged pollination dates of the population on the same day and used a hand-held moisture meter to measure the moisture content of maize kernels every 6 days after pollination. An index-area under the dry down curve (AUDDC) was used as the standardization to indirectly reflect the dehydration rate of kernels among different lines. Two major QTLs were detected using this population. Then we used a map-based cloning method to clone one of the QTLs (*qDRI-2*). Finally, the major QTL (*qDRI-2*) underlying was successfully detected within a 69.042kb region on chromosome 1, in which there are 3 genes based on B73 reference genome. We identified the relative expressions of candidate genes and found that one of the three genes exhibited significant differences but no significant differences for the other two. Furthermore, by identified the expression and phenotype of MU mutant of the candidate gene, we found that there were significant differences in the expression and phenotype, which confirmed that the candidate gene was a functional gene for dehydration rate. These findings implicated the opportunity for cloning the functional gene controlling dehydration rate and explaining its mechanisms, and provided the promising future for improving the dehydration rate of maize kernels.

Abstract 161

The Maize *Opaque 2 Regulated Kinase 1 (ORK1)* Phosphorylates *O2* and Reduces its Transcription Activation Activity

Yuan Yue¹, Xi Yang², Rentao Song^{1*}

¹State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, China Agricultural University, Beijing 100193, China

²Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

*Correspondence Author: rentaosong@cau.edu.cn

(submitted by 袁跃<yuan Yue6113@126.com>)

Abstract: *O2* is one of the most essential transcription factors that regulate the main maize storage proteins. Previous studies of *O2* protein have identified its phosphorylation form in vivo. However, the kinases involved in *O2*'s phosphorylation and function's regulation remain unknown. In this study, truncated *O2* was set as a bait for screening the maize endosperms' cDNA library in yeast and a putative kinase ORK1 was identified. Then their interaction was strongly confirmed by GST-pulldown and LCI. One step further, an in vitro phosphorylation assay was performed and its result showed that ORK1 phosphorylated *O2* in vitro. In order to test the effect of ORK1 on *O2*'s function, a dual luciferase transcription activation assay was performed in tobacco leaves, which showed that ORK1 reduced *O2* transcription activation activity. These results indicated that ORK1 was a direct upstream kinase of *O2* and regulated its transcriptional activation activity. In further studies, the site(s) of *O2* that can be phosphorylated by ORK1 will be obtained, and the effects of mimic phosphorylation on *O2*'s function will be revealed in vivo.

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Identification of maize brace-root quantitative trait loci in a recombinant inbred line population

Ao Zhang^{1,2,3*}, Zhenhai Cui^{1,2*}, Cong Li^{1,2}, Jinhong Luo⁴, Yixin Guan⁵, Lingli Liu^{1,2}, Zhuang Zhang⁶, Lijun Zhang^{1,2}, Yan He⁴, Yanye Ruan^{1,2*}, Haiqiu Yu^{3*}

¹College of Bioscience and Biotechnology, Shenyang Agricultural University, Shenyang 110866, Liaoning, China

²Shenyang City Key Laboratory of Maize Genomic Selection Breeding, Shenyang 110866, Liaoning, China

³College of Agronomy, Shenyang Agricultural University, Shenyang 110866, Liaoning, China

⁴Genetic Improvement, China Agricultural University, National Maize Improvement Center of China, Beijing Key Laboratory of Crop, Beijing 100094, China

⁵Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun 130102, Jilin, China

⁶Tieling County Agricultural Technology Extension and Service Center, Tieling 112000, Liaoning, China

*Corresponding author: yanyeruan@syau.edu.cn

(submitted by 张敖 <zhangao7@vip.qq.com>)

Abstract: Brace roots are vital constituents of the root system in maize. Their contribution to plant development is affected by brace-root traits (BRTs) including tier number (TN), root number (RN) and radius of the brace root (RBR). However, the genetic control of BRTs still remains elusive. Here, we have identified quantitative trait loci (QTLs) from 207 recombinant inbred lines of BY815/K22 grown in three environments to dissect the genetic architecture of BRTs in maize. All three of BRTs were highly heritable and were affected by genotype, environment and the interaction between them. RBR was positively correlated with both RN and TN. Eight QTLs were identified, 3 for TN, 3 for RN and 2 for RBR, and located on chromosome 1, 2, 9 and 10. They together explained 26.4% (TN), 21.5% (RN) and 13.4% (RBR) of phenotypic variation. Sixty of annotated genes were identified from the narrower QTLs by the bin-map method, including genes for signal transduction, gene expression regulation, and metabolism and related processes. The results also show that the interaction may occur between QTLs for BRTs. Our results can help to further study the genetic basis of BRTs and improve the approaches to control maize brace-root system through SNP marker-assisted selection.

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Abstract 163

The maize immune receptor *ZmWAK* recognizes secreted proteins from *Sporisorium reilianum*

Zhang Boqi¹, Zhang Nan¹, Zhang Qianqian¹, Zuo Weiliang¹, Mingliang Xu^{1*}

¹National Maize Improvement Center of China, China Agricultural University, Beijing 100193, China

(submitted by 张博琦 <zhangboqi@cau.edu.cn>)

Abstract: The maize pathogenic Smut fungi *Sporisorium reilianum* is a biotrophic pathogen causing disease threatening to maize production worldwide. Pathogenic interactions are governed by secreted fungal effector proteins. ZmWAK, a transmembrane protein with an extracellular receptor domain and an intercellular kinase domain, has been previously validated to confer the quantitative resistance to head smut disease. A pair of near-isogenic lines, a susceptible line Huangzao4 and its ZmWAK-converted resistant line Huangzao4R, were used to explore the molecular mechanism of ZmWAK-mediated resistance by recognizing protein secreted by *S. reilianum*.

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Abstract 164

Genomic prediction of the testcross performance across multiple populations

Dongdong Li^{1#}, Zhenxiang Xu^{2#}, Riliang Gu², Pingxi Wang¹, Jialiang Xu¹, Demar Lyle², Hongwei Zhang^{1*}, Guoying Wang^{1*}

¹ National Key Facility for Crop Gene Resources and Genetic Improvement, Institute of Crop Sciences, Chinese Academy of Agricultural Sciences, Beijing 100081, P. R. China.

² Center for Seed Science and Technology, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, P. R. China

These authors contribute equally to this work

* Corresponding authors: E-mail: wangguoying@caas.cn; zhanghongwei@caas.cn

(Submitted by Hongwei Zhang <zhanghongwei@caas.cn>)

Abstract: It is necessary to accelerate the development of new varieties using molecular breeding technologies such as genomic prediction. The prediction accuracy of cross population prediction should be enhanced to increase the efficiency of genomic prediction. In this study, four maize testcross populations were constructed and evaluated for yield per plant in three environments. We demonstrated using real data that the accuracy of cross population prediction was related to genetic relatedness, and generally decreased as the genetic relatedness between populations become distant. Including relatives of the test population in the training population increase the accuracy of genomic prediction drastically. The predicted best lines showed substantial overlap with the best lines selected based on observed yield per plant. Therefore, we suggested that, for genomic prediction across distinct populations, including relatives in the training population should be adopted in commercial plant breeding.

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Abstract 165

***PTPP*, a nucleotidase encoding gene, is activated by the heat shock transcription factor HSFA6a and positively regulates ascorbic acid biosynthesis and drought tolerance**

Hui Zhang^{1a}, Yanli Xiang^{1a}, Liping Fang¹, Fei Zhang¹, Xiaopeng Sun¹, Delin Zhang¹, Xingwang Li¹, William Terzaghi² and Mingqiu Dai^{1*}

¹National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

²Department of Biology, Wilkes University, Wilkes-Barre, Pennsylvania 18766

^aThese authors contributed equally to this article.

* Corresponding author: mingqiudai@mail.hzau.edu.cn

(submitted by 张卉<945046216@qq.com>)

Abstract: As a major abiotic stress, drought inhibits plant growth and yield. L-ascorbic acid (AsA), the other name Vitamin C, is the most abundant antioxidant against stress damages in plant. Our knowledge of the mechanisms in regulating AsA production in stress responses is very limited. In this study, we characterized a previously unidentified gene, *PTPP* (*PTP-like Phosphatase*). Arabidopsis *PTPP* (*AtPTPP*) was expressed in multiple tissues and upregulated by ABA and drought treatments. *Atptpp* mutants were hyposensitive to ABA but hypersensitive to osmotic and drought stresses. Overexpressing maize *PTPP* (*ZmPTPP*) promoted plant drought tolerance, indicating conserved and positive roles of *PTPP* in regulation of plant drought responses. *AtPTPP* and *ZmPTPP* released Pi by hydrolyzing GDP/GMP/dGMP/IMP/dIMP. *AtPTPP* positively regulated AsA production via endogenous Pi contents control. Overexpression of VTC2, the rate-limiting synthetic enzyme in AsA biosynthesis, promoted AsA production and plant drought tolerance, which is largely dependent of *AtPTPP* activity. We further demonstrated that the heat shock transcription factor HSFA6a directly bound *AtPTPP* promoter and activated *AtPTPP* expression. Genetic interaction assays showed that *AtPTPP* was required for *HSFA6a* to regulate ABA and drought responses. Our data indicate an ABA-dependent HSFA6a-*AtPTPP* pathway that positively regulates AsA production and plant drought tolerance.

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Abstract 166

Engineering Quantitative Trait Variation in maize by genome editing

Maolin Zhang, Jieting Xu, Jiali Yan, Liu Haijun, Lu Gang, Chen Hanmo, Wu Qilin, Liumei Jian, Mingliang Zhang, Jianbing Yan*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: yjianbing@gmail.com

(submitted by 张茂林<maolinz@126.com>)

Abstract: Current hybrid breeding relies on laborious selection of existed natural mutations that usually is time-consuming and inefficient leading the breeding speed will not meet the needs of people for food and feed. So the existing variations will not meet the breeders' demands and creating artificial beneficial variations has become a matter of urgency. The rapid developed genome editing technology provides an excellent opportunity to create the alleles precisely for breeding purpose. Here, we show that CRISPR/Cas9 saturated editing of promoters and aim to gain diverse *cis*-regulatory alleles involved in every *cis*-element and some of which have beneficial quantitative variation for breeding. We designed to rapidly assess the numerous promoter variants based of phenotype-linked analysis, hoping to provide more variants with continuous changes in quantitative traits to meet the needs of breeding. We performed this saturated mutation method to remodel the promoters of *BR2*, *CCT* and *crtRBI*, three classical key genes controlling plant height, flowering time and kernel nutrition, respectively, hoping to produce *cis*-regulator alleles that have proper plant height, flowering time and carotenoid content, respectively, without negative effects on other agronomic traits. Our results will help to rapidly create novel and beneficial quantitative variations to accelerate breeding process and dissect the mechanism of the relationship between *cis*-regulatory elements and gene functions.

Funding acknowledgement: The National Natural Science Foundation

Abstract 167

Genome-wide transcriptome analysis of miRNAs and mRNAs in maize for high-density planting

Min Zhang¹, Wenzong Li^{1,2}, LiJuan Xing¹, Lei Wang^{1*} and Miaoyun Xu^{1*}

¹ China Biotechnology Research Institute/The National Key Facility for Crop Gene Resources and Genetic Improvement, Chinese Academy of Agricultural Sciences, Beijing, 100081, China;

² College of Agronomy of Shihezi University/The key laboratory of Oasis Eco-Agriculture of Xinjiang Bingtuan, Xinjiang, 832003, China;

*Corresponding author: wanglei01@caas.cn ; xumiaoyun@caas.cn

(submitted by 张敏13126532159@163.com >)

Abstract: Maize (*Zea Mays* L.) plant density has significantly increased yield over the years. However, excessive plant density will restrain growth of maize owing to competition for water, light, and air. As plant density increased, the bald tip became longer and the cell length and size of silk were smaller. The main objectives of this study were to identify key regulators in maize under high-density planting. The crucial nutrient transporting tissue cob and silk of Zheng58 were selected at 6000 plants per mu, 8000 plants per mu, 10000 plants per mu, respectively, to performed miRNA-seq and RNA-seq analysis to detect differentially expressed genes (DEGs) and differentially expressed miRNAs (DEMs) at different plant density. 116, 102 and 104 in cob and 96, 79 and 87 known miRNAs in silk were identified respectively. 49 and 60 DEMs were identified in cob and silk were among the three comparison groups respectively. One and 3 candidate miRNA-mRNA modules were found in cob and silk by integrative analysis of DEMs and DEGs, respectively. It is mentioning that members of the miR169 family were dynamically expressed in the cob, which will support for exploiting new function in nutrient transport. Our study provided new insights into the molecular mechanisms and laid the foundation for future molecular study of maize for high-density planting.

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Abstract 168

Functional Study of An *O2* Downstream Target Gene

Mingliang Zhang¹, Liming Xu², Weiwei Qi², Rentao Song^{1*}

¹State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

²Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

*Correspondence Author: rentaosong@cau.edu.cn

(submitted by 张明亮<mingliangzhang@cau.edu.cn>)

Abstract: *OPAQUE2* (*O2*) plays an important regulatory role in the pathway of maize endosperm metabolism. *O2* encodes an endosperm-specific bZIP transcription factor and mainly regulates the expression of zein genes. In this study, we identified an *O2*-regulated gene and demonstrated that it can be activated directly by *O2* through transient transcription Dual-LUC assay. In order to analyze the spatiotemporal expression pattern of the gene, it was confirmed by quantitative Real-time RT-PCR that the gene was specifically expressed in the kernel. Through yeast two-hybrid (Y2H) library screening, the protein interacts with a protein involved in starch synthesis. After knocking out the gene by CRISPR-Cas9 technology, it was found that the ratio of amylose/amylopectin was decreased. Scanning electron microscopy showed that adjacent starch granules was in contact with each other and connected to each other. Understanding the molecular basis of this previously uncharacterized starch structure will accelerate the development of maize endosperm research.

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两个野生玉米回交 RIL 群体的 QTL 定位

吴伸伸¹, 李文强¹, 桂松涛¹, 杨宁¹, 张清华¹, 严建兵^{1*}

¹ 华中农业大学作物遗传改良国家重点实验室, 武汉, 430070

*通讯作者: yjianbing@mail.hzau.edu.cn

(submitted by 吴伸伸 shenshenwu666@163.com, 张清华 qhzhang@outlook.com)

摘要: 现代栽培玉米约于一万年前, 由野生玉米-大刍草 (*Zea mays ssp. parviglumis*) 驯化而来。而在人类对玉米的驯化过程中, 也导致玉米遗传多样性大大降低。鉴定与玉米驯化相关性状的遗传位点, 对于丰富我们对玉米驯化的认识具有重要意义。本研究选取了现代玉米的直系祖先, 大刍草亚种 *Zea mays ssp. parviglumis* 为父本, 以已有参考基因组序列的 B73 和 Mo17 为母本和轮回亲本, 通过 1 代回交和 6 代自交, 构建了两个野生玉米的 BC₁F₆ 回交 RIL 群体, *pariviglumis* × B73(PB) 和 *pariviglumis* × Mo17(PM) 群体分别由 800 个家系构成。随着二代测序技术的发展和成本的降低, 利用重测序数据构建遗传图谱已极其普遍, 本研究使用 Illumina HiSeq X TEN, 对 1600 份玉米-大刍草 BC₁F₆-RILs 进行双端重测序 (各 150bp)。大刍草亲本测序深度为 25X; PM 群体平均每份材料获得了 6.86G 数据, 平均测序深度达到 3.12X, 平均基因组覆盖度达到 79.60%, 共获得 10,979,442 个 SNP; PB 群体平均每份材料获得了 6.07G 数据, 平均测序深度达到 2.76X, 平均基因组覆盖度达到 74.71%, 共获得 11,092,342 个 SNP; 两个群体选取双亲分离的 2,504,840 个和 2,522,860 个高质量 SNP, 分别构建了含有 3553 和 4000 个 bin 的覆盖全基因组的高密度遗传图谱。通过一年两点四个重复的田间试验 (内蒙古巴彦淖尔和云南景洪), 调查了包括 3 个开花期性状、8 个农艺性状、8 个穗部性状、1 个驯化性状 (落粒性) 以及 1 个病害性状 (穗腐病) 在内的共计 21 个性状, 结合基因型和表型数据, 开展重要性状 QTL 定位, 为后续野生遗传资源挖掘提供了夯实的基础。同时我们将结合遗传图谱与参考基因组在全基因组水平上寻找结构变异、偏分离位点和重组热点, 解释其中的生物学问题。

关键词: 大刍草、RIL 群体、驯化、遗传图谱、QTL、结构变异

Abstract 170

Identification and characterization of three phytosterol synthesis related genes in Maize

ShanShan Zhang^a, GeGe Lu^a, Chenchen Zhang, Tao Li, Wei Zhang, Beijiu Cheng, Xin Zhang*

National Engineering Laboratory of Crop Stress Resistance Breeding, Anhui Agricultural University, Hefei 230036, China

^aThe first two authors contributed equally to this paper.

*Corresponding authors: xinzhang@ahau.edu.cn

(submitted by 张晨晨<362491912@qq.com>)

Abstract: Phytosterols, the important composition element in plant cell bilayer membrane participate in the process of plant photosynthesis, reproductive, immune, and be response to external stress. They are also the important metabolite that could regulate plant senescence. Up to now, there are few reports about the genes that regulate the biosynthesis of phytosterols and gene mapping, which resulted in the lack of understanding of the genetic basis of natural variation. In the present study, the phytosterols in maize kernel of 244 maize inbred lines were extracted by the method of saponification, the composition and content were identified by GC-MS. According to the GWAS analysis with the phenotype data of these maize, nine SNPs and 32 candidate genes which associated with phytosterol content were identified. To investigate the relationship between gene expression level and sterol content, quantitative RT-PCR was performed with RNA preparations isolated from special maize kernels which had different sterol contents. Three genes were identified to be closely related to sterol content and probably participate in the synthesis of sterols. These three genes were transferred into *S. cerevisiae*, and the content of ergosterol in transgenic yeast were significantly increased. Further analysis showed that these proteins located to the Golgi, cell membrane and nucleus in protoplast of maize.

Abstract 171

基于玉米基因组 PAV 结构变异及 5 个组织 RNA-Seq 数据的多性状全基因组预测

张体付¹, Alex B. Brohammer², 李知², Rafael Della Colleta², Candice N. Hirsch^{2*}

¹ 江苏省农业科学院种质资源与生物技术研究所以, 江苏省重点农业生物学实验室, 南京, 210014

² Department of Agronomy and Plant Genetics, University of Minnesota, Saint Paul, MN 55108

* 通讯作者: cnhirsch@umn.edu

(submitted by 张体付 <zhangtifu_82@163.com>)

摘要: 全基因组选择技术通过对目标性状的预测即可实现优良候选个体的选择, 在玉米高效育种中具有重要意义。目前, 对目标性状全基因组预测能力(prediction ability, PA)的研究主要通过基因组 SNP (single nucleotide polymorphism) 变异或单个组织的转录组开展的。玉米基因组 PAV (presence/absence variation) 结构变异及不同组织转录组如何作用于 PA 尚未明确。本研究利用鉴定的~4,000 个 PAV 基因及 5 个组织(授粉后 15 天胚乳、V7 节间、第七片完全展开叶、播种后 9 天幼苗及幼根)的 RNA-Seq 数据(~30,000 个基因)对 19 个玉米性状(株高、穗位高、上部叶片数、下部叶片数、茎粗、散粉期、抽丝期、轴长、轴粗、穗粗、穗行数、行粒数、粒长、粒宽、粒厚、籽粒面积、粒重、籽粒区产及穗轴区产)在 1~3 个环境下的 PA 进行了分析。研究发现, PAV 基因的 PA 与 SNP 相比并无明显差异, 且 PA 大小不受 LD (linkage disequilibrium) 的影响。5 个组织转录组对同一性状的 PA 相似, 该结果表明, 利用与目标性状具有相同组织来源的转录组进行预测并非是获得高 PA 必须的。进一步分析发现 PAV 基因与非 PAV 基因间、基因组与转录组间的 PA 相同, 基因组与转录组的整合也不能提高 PA。虽然少量(~50)基因的 PA 即可达到全基因组水平, 但其预测的稳定性较差。对高、低表达基因的 PA 分析显示转录组 PA 与基因的表达水平无关、但与基因的表达变异有关, 极端高表达变异(coefficient of variation ≥ 5)基因不利于 PA 的保持。综合来看, 多个性状在不同环境下的 PA 表现出较大差异, 这种差异主要由于相关性状狭义遗传力的改变引起的($r^2=0.81$, $p=7.4e-17$)。相关分析证实某一性状的 PA 可由其他与其高度正相关或负相关的性状的 PA 进行估计($r^2=0.82$, $p=5.9e-07$; $r^2=0.71$, $p=9e-36$)。

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Abstract 172

***ZmBRCA2* is dispensable for RAD51/DMC1-facilitated homologous recombination in maize meiosis**

Ting Zhang, Ju-Li Jing, Ya-Zhong Wang and Yan He

National Maize Improvement Center, China Agricultural University, Beijing 100193

(Submitted by 张挺<zhangting@cau.edu.cn>)

Abstract: Meiosis is the core process of sexual reproduction, initiated from the programmed formation of DNA double strand breaks (DSBs) and subject to homologous recombination (HR) repair. The homologs of BRCA2 have been demonstrated to be essentially required for recruiting RAD51 and DMC1 on the chromosomes to trigger the single-end invasion (SEI) during meiotic HR in various organisms. In this study, we characterized the meiotic functions of *BRCA2* in maize. We found that the disruption of *ZmBRCA2* caused aberrant chromosome behaviors and defective homologous pairing and synapsis, consequently leading to both male and female sterility. Surprisingly, we observed that although the number of RAD51 and DMC1 foci were substantially reduced, but not completely abolished in *Zmbrca2* mutant, suggesting that *BRCA2* is dispensable for RAD51 and DMC1 loading in maize. In addition, it is also surprised to observe 20 univalents in *Zmbrca2* mutant, rather than chromosome fragmentation as in other organisms, indicating that *ZmBRCA2*-dependent RAD51/DMC1 installation is specially recruited into crossover pathway. This novel finding further suggests that *ZmBRCA2* play a regulatory role in determining the choice of the different repair pathways. The functions of *ZmBRCA2* identified here have not been reported for any other BRCA2 homologs in other species.

Abstract 173

Multivariate analyses of root phenotype and dynamic transcriptome underscore valuable root system architectures and drought responsive gene networks in maize

Xuhui Ma¹, Junling Pang¹, Jun Zhao¹, Xia Zhang^{1*}

¹ Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing 100081, China

*Corresponding author: zhangxia@caas.cn

(submitted by Xia Zhang zhangxia@caas.cn)

Abstract: Roots are the major organ for water and nutrient acquisition, and also the first organ that initially senses the soil water deficit. As such, the root system is becoming an important breeding target for crop improvement. Root system architecture (RSA), referring to the spatial configuration of root components, is determined by length, number, spread, branching density, and angle of root components. RSA is of broad importance in plant adaptation to drought since it determines plant access to soil water and drives the growth of above-ground organs. Past efforts for improving crop productivity under drought have in general focused on shoot-related above-ground traits, whereas breeding via modifying root traits has been utilized to a lesser extent due to the difficulty of precise and fast RSA evaluation and uncertainty on the key root traits to be targeted. Here, we used two maize cultivars contrasting for drought tolerance and employed image-based phenotyping approach as well as dynamic root-specific transcriptome analysis to identify beneficial RSA components and molecular genetic factors in drought response. Several RSA features including larger root size, longer root length, and more number of lateral roots and seminal roots were found valuable to the improved drought tolerance. Multiple transcription factors, regulatory proteins and root-specific and root-predominant key hub genes were identified to be associated with root development and drought response. Findings will enable us to evaluate the potential value of different root traits for pre-screening of water-deficit tolerance during the early seedling stage and serve a valuable resource that merits in-depth functional analyses towards a better understanding of RSA-associated drought tolerance in maize.

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Abstract 174

Genetic variations in *ZmTIP1* are critical for root hair Development and Drought Tolerance of Maize Seedlings

Xiaomin Zhang^{1,2,3}, Shengxue Liu¹, Yue Mi¹, Hude Mao⁴, Feng Qin^{1*}

¹State Key Laboratory of Plant Physiology and Biochemistry, College of Biological Sciences, China Agricultural University, Beijing 100193, China

²Key Laboratory of Plant Molecular Physiology, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China

³University of Chinese Academy of Sciences, Beijing 100049, China

⁴State Key Laboratory of Crop Stress Biology for Arid Areas, College of Plant Protection, Northwest A&F University, Yangling 712100, Shaanxi, China

*Corresponding author: qinfeng@cau.edu.cn

(submitted by 张小敏<zxm19900718@163.com>)

Abstract: Drought is a primary abiotic stress on maize production globally. In a previous genome-wide association study, we identified a significant association between the natural variation of *ZmTIP1*, encoding a S-acyl transferase, with drought tolerance in maize seedlings. In this research, we characterized the entire genetic diversity on this locus among different maize varieties collected from tropical, subtropical and temperate regions worldwide. Enlightened by all the newly identified variations, two major haplotypes (Hap1 and Hap2) were found for *ZmTIP1*. The drought-sensitive inbred line MO17 carries Hap1, and the drought-resistant inbred line CIMBL55 harbors Hap2. The coding sequences of both haplotypes restored the short root hair phenotype of an Arabidopsis mutant with mutations in a S-acyl transferase gene. It suggested that ZmTIP1 encodes a functional S-acyl transferase and the genetic variants in the coding region did not affect ZmTIP1 protein function. Further analysis among 109 maize inbred lines showed that the expression level of Hap2 was significantly higher than that of Hap1, and the root hair length of Hap2 was remarkably longer than that of Hap1, implying that the gene expression level is determinant to the gene functional variation. Further analysis of the *ZmTIP1* Mu-insertional mutants demonstrated that the gene function was essential for root hair elongation and plant survival under water deficits, while the root hair length and drought resistance of *Ubi:ZmTIP1* transgenic maize were enhanced. ZmTIP1-GFP was observed to localize at Plasma membrane, PVC, and Golgi. Importantly, we identified a calcium-dependent protein kinase, ZmCPK28, as a putative substrate of ZmTIP1. The ZmTIP1-mediated-palmitoylation facilitated ZmCPK28 membrane association. Increasing *ZmCPK28* expression in transgenic maize consistently enhanced the root hair length and drought resistance. Collectively, we discovered that ZmTIP1-mediated localization of ZmCPK28 at the plasma membrane contributing to root hair elongation and plant drought tolerance. The *ZmTIP1* favorable haplotype may provide valuable genetic resource for maize drought tolerance improvement.

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Abstract 175

Integrated transcriptome, small RNA, and degradome analysis

reveals the complex network regulating starch biosynthesis in maize

Xiaocong Zhang^{1#}, Sidi Xie^{1,2#}, Jienan Han¹, Yu Zhou^{1,3}, Chang Liu^{1,3}, Zhiqiang Zhou¹, Feifei Wang¹, Zixiang Cheng¹, Junjie Zhang⁴, Yufeng Hu², Zhuanfang Hao¹, Mingshun Li¹, Degui Zhang¹, Hongjun Yong¹, Yubi Huang², Jianfeng Weng^{1*}, Xinhai Li^{1*}

¹Institute of Crop Science, Chinese Academy of Agricultural Sciences, Beijing 100081, China

²College of Agronomy, Sichuan Agricultural University, Chengdu 611130, China

³College of Agronomy, Northeast Agricultural University, Harbin 150030, China

⁴College of Life Science, Sichuan Agricultural University, Ya'an 625014, China

*Corresponding author: wengjianfeng@caas.cn, lixinhai@caas.cn

(submitted by 张晓聪 <cong_11101@126.com>)

Abstract: Starch biosynthesis in endosperm is a key process influencing grain yield and quality in maize. Although a number of starch biosynthetic genes have been well characterized, the mechanisms by which the expression of these genes is regulated, especially in regard to microRNAs (miRNAs), remain largely unclear. Sequence data for small RNAs, degradome, and transcriptome of maize endosperm at 15 and 25 d after pollination (DAP) from inbred lines Mo17 and Ji419, which exhibit distinct starch content and starch granule structure, revealed the mediation of starch biosynthetic pathways by miRNAs. Transcriptome analysis of these two lines indicated that 33 of 40 starch biosynthetic genes were differentially expressed, 27 of which were up-regulated and three of which were down-regulated in Ji419 compared with Mo17. Through combined analyses of small RNA and degradome sequences, 22 differentially expressed miRNAs were identified, including 14 known and eight previously unknown miRNAs that could target 35 genes. Furthermore, a complex co-expression regulatory network was constructed, in which 19 miRNAs could modulate starch biosynthesis in endosperm by tuning the expression of 19 target genes. Moreover, the potential operation of four miRNA-mediated pathways involving transcription factors, miR169a-NF-YA1-GBSSI/SSIIIa and miR169o-GATA9-SSIIIa/SBEIIb, was validated via analyses of expression pattern, transient transformation assays, and transactivation assays. Our results suggest that miRNAs play a critical role in starch biosynthesis in endosperm, and that miRNA-mediated networks could modulate starch biosynthesis in this tissue. These results have provided important insights into the molecular mechanism of starch biosynthesis in developing maize endosperm.

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Abstract 176

Characteristics of copy number variations and functional impact in maize population

Xiao Zhang^{1,2,3}, Karl A. G. Kremling¹, M. Cinta Romay¹, Robert Bukowski⁴, Qi Sun⁴, Edward S. Buckler^{1,5}, Dan Liu^{2,3}, Ling Wu^{2,3}, Shibin Gao^{2,3}, Fei Lu^{1,6*}

¹Institute for Genomic Diversity, 175 Biotechnology Building, Cornell University, Ithaca, New York 14853, USA

²Maize Research Institute, Sichuan Agricultural University, Chengdu 611130, Sichuan, China

³Key laboratory of Biology and Genetic Improvement of Maize in Southwest Region, Ministry of Agriculture, Chengdu 611130, Sichuan, China

⁴Bioinformatics Facility, Institute of Biotechnology, Cornell University, Ithaca, New York 14853, USA

⁵USDA-ARS, R. W. Holley Center, Cornell University, Ithaca, New York 14853, USA

⁶The State Key Laboratory of Plant Cell and Chromosome Engineering, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, Beijing 100101, China

*Corresponding author: flu@genetics.ac.cn

(submitted by 张 啸<hunterzap@163.com>)

Abstract: We jointly applied two read depth methods in next-generation sequencing (NGS) data of 271 inbred lines from ‘maize282’ population for the analysis of copy number variation (CNV) distribution and characteristics. CNV effect on gene expression was also studied by eQTL detection on expression abundance in seven developmental tissues and stages. The results showed that the average CNV length was accounted for nearly 10.2% of total genome. To understand the regulation rules of CNV in regulating gene expression, published RNA-Seq data and CNV alleles were combined for calling eQTLs by genome-wide association study (GWAS). The results indicated that gene expression was negatively correlated to CNV overlapped proportion of genes and eQTLs can be divided into *cis* and *trans* eQTLs according to the relative position to genes. Some eQTL hotspots were captured which can simultaneously impact expression of multiple genes. In addition, more than 90% significantly associated genes can be also identified using whole-genome SNPs. Our study not only helps to characterize CNVs in maize, but also paves a way for the functional and genetic research of CNVs.

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Abstract 177

The application of Short Tandem Target Mimic-mediated microRNAs inactivation in *Arabidopsis* and maize agronomic traits improvement

Zhanhui Zhang^{1*}, Tianxiao Yang¹, Jihua Tang^{1*}, Guiliang Tang^{1, 2*}

¹Collaborative Innovation Center of Henan Grain Crops, Henan Agricultural University, Zhengzhou 450002, China;

² Department of Biological Sciences, Life Science and Technology Institute, Michigan Technological University, Houghton, MI 49931, USA.

*Corresponding author: Guiliang Tang, gtang1@mtu.edu

Jihua Tang, tangjihua1@163.com

(Submitted by 张战辉 <amair0534@163.com>)

Abstract: Compared to animal miRNAs, plant miRNAs tend to have fewer targets that mainly encode transcription factors and F-box proteins. This indicates that miRNA is at the central position of gene expression regulatory networks of plant growth and development. Manipulation of mRNA transcript abundance via miRNA control provides a unique strategy for improvement of the complex agronomic traits of crops. In previous studies of our group, we have created 82 STTMs for targeting both conserved and species-specific miRNAs in *Arabidopsis*, and three STTMs in maize. Based on these STTM mutants, we have tried to explore the potential application of STTM technology in agronomic traits improvement. In *Arabidopsis*, we have generated a *STTMmiR165/166* and *STTMmiR160* double mutant by single mutant crossing. The double mutant plants exhibited a series of compromised phenotypes in leaf development and drought tolerance in comparison to phenotypic alterations in the single STTM lines. In maize, alternatively, we have screened according elite phenotypic variations in the three STTM mutants, almost without developmental defects. These elite mutant variations can be used for agronomic traits improvement via MAS. For example, *STTMmiR166* mutants have superior drought tolerance, *STTMmiR156* mutants have upward leaves, and *STTMmiR172* mutants have excellent lodging resistance. These preliminary results offered useful theoretic and technical support for crop improvement by manipulating miRNA expression.

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玉米抗禾谷镰刀菌茎腐病 QTL 定位

张智超 赵雅琪 张丁芳 杨琴

(旱区作物逆境生物学国家重点实验室/西北农林科技大学农学院, 陕西杨凌 712100)

摘要: 【研究背景】茎腐病是一种严重危害玉米生产的土传性病害, 随着气候环境变化、耕作制度改变和种植结构调整等因素, 玉米茎腐病危害在我国逐年加剧。玉米对茎腐病的抗性是复杂的数量性状, 由多基因控制, 受环境影响大。目前已克隆了两个抗禾谷镰刀菌茎腐病的 QTL-*qRfg1* 和 *qRfg2*, 其中 *qRfg1* 效应较大, *qRfg2* 是一个微效位点。挖掘玉米种质中新的优良抗病位点或抗病基因, 通过分子标记辅助的聚合育种, 培育抗茎腐病品种, 是保证玉米优质高产稳产的重要因素。【材料与方法】本研究选用抗病玉米自交系 KA105 和感病自交系 KB204 组配的 F_{2:3} 群体, 利用随机区组设计, 采用土埋伤根法人工接种禾谷镰刀菌, 采用劈茎观察法进行表型鉴定, 利用靶向测序基因分型技术 (GBTS-Maize10K) 获得全基因组分子标记, 构建连锁图谱, 进行 QTL 定位。【结果与分析】首先, 对已克隆的抗病位点 *qRfg1* 和 *qRfg2* 进行鉴定, 发现 KA105 和 KB204 均不带有抗病的 *qRfg1* 等位基因, 而 *qRfg2* 位点在两亲本中是分离的, KA105 带有抗病的 *qRfg2* 等位基因, KB204 是感病等位基因。2018 年夏天在陕西杨凌对两亲本及 250 份 F_{2:3} 家系的抗性鉴定表明, 病情分级、感病茎节数、感病茎高以及感病面积>75%的茎节数各指标之间相关性极显著 (相关系数为 0.75-0.95)。KA105 的病情分级是 2.36, KB204 的分级是 3.63, F_{2:3} 家系的病情分级呈正态分布, 其中 21 份材料表现高抗 (分级 0-1), 219 份的分级介于 2-4, 10 份表现高感 (分级 4-5)。【结论】两个亲本中均不带有已克隆的主效抗病基因 *qRfg1*, 因此, 我们有望发现新的抗茎腐病 QTL 位点。通过多个环境的抗病性重复鉴定, 将对抗茎腐病的 QTL 进行定位和验证。

关键词: 玉米茎腐病; 禾谷镰刀菌; 人工接种; QTL 定位;

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Abstract 179

Distribution of four bioactive flavonoids in maize tissues of five varieties and correlation with expression of the biosynthetic genes

Zhongbao Zhang ^{1,2}, Zhibin Liang ^{2,3}, Longfei Yin ¹, Qing X. Li ^{2,*}, Zhongyi Wu ^{1,*}

¹ Beijing Agro-Biotechnology Research Center, Beijing Academy of Agriculture and Forestry Sciences, Beijing 100097, China

² Department of Molecular Biosciences and Bioengineering, University of Hawaii at Manoa, Honolulu, Hawaii 96822, United States

³ Current address: Salk Institute for Biological Studies, 10010 N Torrey Pines Road, La Jolla, CA 92037.

*Corresponding author: qingl@hawaii.edu; zwu22@126.com

(submitted by 张中保<happyzzb@126.com>)

Abstract: Flavonoids are characteristic in maize and have diverse biological functions. C-Glycosylflavones are neuroprotective against β -amyloid-induced tau hyperphosphorylation and neurotoxicity in SH-SY5Y cells, which is relevant to Alzheimer's disease prevention and treatment. The content of the C-glycosylflavones eriodictyol, luteolin, isoorientin and maysin varied in pollens, silks, tassels and seeds among five maize varieties. Eriodictyol content was high (51 - 322 ng/g dw) in pollens, while luteolin content was low (0.2 - 106 ng/g dw) in all four tissues. The isoorientin content was approximately 3- to 10-fold greater than eriodictyol in pollens and tassels, particularly in the hybrid M1 and sweet corn M5 varieties. Maysin content was high in most silks and tassels. The differential expression of five genes involved in the maysin biosynthesis correlated well with the profiles of the four flavonoids among tissues and varieties. The present study offers valuable data for maize breeding and the use of maize flavonoids as functional food components.

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Abstract 180

Identification and Characterization of Maize ACD6-like Gene Reveal that *ZmACD6* Is the Maize Orthologue Conferring Resistance to *Ustilago Maydis*

Zhongqin Zhang*, Jinjie Guo, Yongfeng Zhao, Shangchuan Jiang, Jingtang Chen
Hebei Sub-center of Chinese National Maize Improverment Center, North China Key Laboratory for Crop Germplasm Resources of Education Ministry, College of Agronomy, Agricultural University of Hebei, Baoding, 071000 China

Correspondence: Zhongqin Zhang (zqzhang2015@126.com)

(Submitted by 张忠琴 zqzhang2015@126.com)

Abstract: Enhancing the broad-spectrum resistance is a major goal of crop breeding. However, studies on broad-spectrum resistance of crops are still lack and the underlying molecular mechanisms remain elusive. ACCELERATED CELL DEATH 6 (ACD6) has been demonstrated as a key component of broad-spectrum resistance in model plant *Arabidopsis* that acts in a positive feedback loop with salicylic acid (SA) to regulate multiple pattern receptors. However, the role of *ACD6* in disease resistance in crop plant is not known. Here, we found that the transcript of *ANK23*, one of 15 *ACD6*-like genes in maize, is induced by SA application and fungus *Ustilago maydis* infection. Importantly, *ANK23* can complement disease resistance of *acd6-2* mutant in *Arabidopsis*. The evidence demonstrated that *ANK23* is the orthologue of *ACD6*, thus we named it as *ZmACD6*. Furthermore, using CRISPR/Cas9 we got knockout maize plants of *ZmACD6*, which are more susceptible to *Ustilago Maydis* compared to wild type plants. We also present evidence that we obtain one *ZmACD6* high expression line SC-9 from a diverse maize natural population and SC-9 increase disease resistance against *Ustilago Maydis* providing a practical approach to cultivate elite varieties with enhanced disease resistance.

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Abstract 181

Acquisition of root-specific gene knockout transgenic plants by CRISPR/Cas9 gene editing approach in maize

Yan Zhang^a, Weiwei Qi^a and Rentao Song^{a, b*}

^aShanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China.

^bNational Maize Improvement Center of China, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

*Address for correspondence to rentaosong@cau.edu.cn.

(submitted by 章琰<1473985378@qq.com>)

Abstract: Maize is the cereal with the highest production worldwide. The root plays an important role during plant development and growth. Maize forms a complex root system to efficiently extract mineral nutrients and water from soil and allocates them to the energy-delivering. Thus maize plant architecture can be improved by root breeding to create an ideal phenotype for further yield increases. Nevertheless, the root system remains a largely untapped resource for future maize improvement. So getting the root-specific gene mutants mutant is important for maize root research and CRISPR/Cas9 technology is now the efficient genome-editing technique in the world. CRISPR/Cas9 system which has been remolded by us for its use in maize is used to make the mutant of four root-specific genes (*Rsg1-4*). We constructed two double-gene four-targets Cas9 vectors obtained genetically modified maize plants. Genotyping of T1 seedlings have shown that *Rsg1* and *Rsg2* have been edited. *Rsg1* has a single base mutation and *Rsg2* has occurred with a large fragment deletion.

Funding acknowledgement: This work was supported by the National Natural Sciences Foundation of China (91635303) .

Abstract 182

Genetic architecture of maize stalk rind penetrometer resistance in linkage and association mapping populations

Binghao Zhao^{1,4}, Kun Li^{2,4}, Min Wang¹, Yingjie Xiao³, Jianbing Yan³, Jiansheng Li¹ and Xiaohong Yang^{1,*}

¹State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center of China, China Agricultural University, Beijing, China

²Institute of Crop Science, Chinese Academy of Agricultural Sciences, Beijing, China

³National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan, China

⁴These authors contribute equally to this work.

*Corresponding author: yxiaohong@cau.edu.cn

(submitted by Binghao Zhao<zhaobinghao_93@163.com>)

Abstract: Maize (*Zea mays* L.) is a staple crop for humans and livestock. Maize yield suffers severe reduction due to stalk lodging worldwide annually. Although the stalk strength plays an important role in maize stalk lodging resistance, the genetic architecture of stalk strength remains largely unclear. Rind penetrometer resistance (RPR) is an effective way to measure stalk strength in maize. In this study, we measured RPR across 1,887 lines in a maize Random Open-parents Association Mapping population (ROAM) in four environments. With separate linkage mapping (SLM), joint-linkage mapping (JLM) and ROAM-based genome-wide association mapping (GWAS), we identified 31, 80 and 112 (26 remained after backward selection) loci significantly associated with RPR in ROAM, respectively. The phenotypic variation explained by each QTL within SLM ranged from 5.1%-17.0%. Two other methods, JLM and GWAS, could explain total phenotypic variation up to 55.5% and 36% (24% after backward selection), respectively. In addition, GWAS using 508 diverse inbred lines identified 81 SNPs in 22 loci were detected significantly associated with RPR at $P < 0.0001$. Among these loci, 122 consistent loci for RPR were detected by at least two methods. These results indicate numerous minor-effect loci contribute to the complex nature of maize RPR. Our results will be useful for improving maize stalk strength.

Abstract 183

Characterization and the Expression Analysis of Nitrate Transporter (NRT) Gene Family in maize

Qiufang Zhao ^{1,2}, Liqiang Jia ^{1,2*}, Shu Chen^{1,2}

¹South Subtropical Crops Research Institute, Chinese Academy of Tropical Agricultural Sciences, Zhanjiang, Guangdong 524091, China

²Key Laboratory of Tropical Fruit Biology, Ministry of Agriculture and Rural Affairs of China, Zhanjiang, Guangdong 524091, China

*Corresponding author: liqiangj@zju.edu.cn

Abstract: Nitrate is the preferred nitrogen source of higher plants and an essential nutrient for plant growth and development. Nitrate transporters (NRTs) play vital roles in the nitrate uptake and transportation. Maize (*Zea mays* L.) is an important cereal crop and is the staple food for most people around the world. However, the NRT gene family in maize is still unexplored. In this study, a comprehensive analysis of the maize NRT family was performed. We identified 91 NRT genes (ZmNRTs) distributed and ZmNRT1, ZmNRT2 and NRT3 with 84, 5 and 2 members, respectively. The ZmNRTs genes were distributed across 10 chromosomes at different densities. For convenience, the 84 ZmNRT1 genes were named ZmNRT1.1 to ZmNRT1.84 based on their location on the chromosome from chromosomes 1 to 10, and 5 ZmNRT2 genes were named ZmNRT2.1 to ZmNRT2.5, 2 of ZmNRT3 were name ZmNRT3.1 and ZmNRT3.2. By investigating the expression profiles of these genes in various tissues, we found that the expression pattern of some ZmNRTs genes is tissue-specific. Example, most ZmNRT2s genes were expressed in root and leaf, indicating that these genes are related to nitrogen uptake and transport in roots and leaves. We also examined the expression of the ZmNRT2s under nitrate starvation and found NRT2.2, NRT2.4 and NRT2.5 were strongly induced expression under low-nitrate nitrogen suggesting that ZmNRTs may play a broad role in the maize in response to nitrate deficiency.

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Abstract 184

Maize VP1-mediated ABA pathway is essential for proteome rebalancing between endosperm and embryo

Xixi Zheng^{1,2}, Qi Li^{1,2}, Changsheng Li^{1,3}, Dong An³, Yongrui Wu^{1*}

¹National Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Institute of Plant Physiology and Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200032, China

²University of the Chinese Academy of Sciences, Beijing 100049, China.

³Department of Plant Sciences, School of Agriculture and Biology, Shanghai Jiao Tong University, Shanghai 200240, China

*Corresponding author: yrwu@sibs.ac.cn

(submitted by 郑喜喜<xxzheng@sibs.ac.cn>)

Abstract: The double fertilization in flowering plants gives rise to the embryo and endosperm. Although they have different fates and biological functions, many evidences have shown that they appear to be developmentally coordinated through mutual communications. Here, we show that the embryo can sense and respond to the proteome alteration in the endosperm. Maize endosperm is the principal organ for the storage protein accumulation, and the main storage proteins in the endosperm are prolamins called zeins. Although the maize seed possesses a standardized proteome, mutations (like *o2*, *fl2*) or the α -zein RNAi (*α RNAi*) can induce the proteome rebalancing, by which the loss of zeins is compensated by the increase of non-zein proteins, leading to a relatively constant total protein level in the seed. We characterized the two most predominant increased proteins, Globulin1 and Globulin2 (GLB1 and GLB2), from non-zein proteins in *o2* and *α RNAi* seeds. Transcript levels of *Glbs* are also dramatically enhanced, indicating that the proteome rebalancing is regulated in part at the transcriptional level. In the double mutant of *α RNAi; vp1*, no obviously increased proteins were observed in the embryo, indicating that VP1 is the essential regulator in the embryo responding to the proteome alteration in the endosperm. By performing trans-activation assays, we proved that VP1 is able to strongly transactivate *Glbs* and diverse pathways induced by kernel proteome rebalancing in the presence of ABA. Overall, our results indicate that ABA-driven VP1 plays essential regulatory roles in signal and nutritional communications between endosperm and embryo.

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Abstract 185

Exploring genetic basis of regulation in root to response low-phosphorus stress by *ZmPAT1* in maize

Haixu Zhong¹, Xin Ding¹, Xiao Zhang¹, Bowen Luo¹, Dan Liu¹, Ling Wu¹, Shibin Gao^{1*}

¹Maize Research Institute, Sichuan Agricultural University, 611130, Sichuan, Chengdu, China, and

²State Key Laboratory of Crop Genetics of Disease Resistance and Disease Control, Sichuan, Chengdu, China

*Corresponding author: shibingao@163.com

(submitted by 钟海旭<602473599@qq.com>)

Abstract: According to genome-wide association analysis of maize seedling phenotypes under low phosphorus treatment and control conditions, candidate gene *ZmPAT1* was found from the loci associated with root dry weight index. The gene belongs to PAT1 super family and contains UBA-Like folding in its domain, which is related to ubiquitin activator UBA, and a GAPDH active site was found. The full-length sequence of genes amplified from 154 inbred lines of the related population was analyzed, the polymorphic loci were correlated with seedling traits and low phosphorus tolerance index (T/CK). Several loci significantly correlated with the number of seed roots were detected under low phosphorus tolerance index. An Indel was found in the exon region of *ZmPAT1* and validated in the isolated population of recombinant inbred line (RIL). The expression of *ZmPAT1* was continuously up-regulated in roots but down-regulated in leaves by qRT-PCR; the subcellular localization results showed that the gene was located in nucleus; the protein interaction between *ZmPAT1* and *ZmSPX1* was found by yeast two-hybrid. At the same time, we successfully transformed and obtained *ZmPAT1* overexpressed *Arabidopsis thaliana* and maize positive plants. Phenotypic identification was carried out on T4 positive plants of *Arabidopsis thaliana*.

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Abstract 186

Identification and characterization of a major QTL-*qRgls1* against gray leaf spot in maize

Tao Zhong¹, Mang Zhu¹, Yan Zhang¹, Ling Xu¹, Yancong Li², Zhijian Xie², Tingting Liu², Xingming Fan³ and Mingliang Xu^{1*}

¹National Maize Improvement Center of China, China Agricultural University, 2 West Yuanmingyuan Road, Beijing 100193, P. R. China.

²Baoshan Institute of Agricultural Science, Taibao North Road, Longyang District, Baoshan 678000, P.R. China.

³Institute of Food Crops, Yunnan Academy of Agricultural Sciences, Longtuo Street, Kunming 650205, P.R. China.

*Corresponding author: mxu@cau.edu.cn

(submitted by 钟涛<zhongtaomvp@163.com>)

Abstract: Gray leaf spot (GLS), caused by fungal pathogens *C. zeae-maydis* and *C. zeina*, is a foliar disease that poses a grave threat to global maize production. Thus, discovery of GLS resistant genes is beneficial to development of new maize varieties with improved resistance. Previously, a major quantitative resistance locus, *qRgls1*, was detected on maize Chr. 8 and could reduce the disease severity index (DSI) by 10%. *qRgls1* was later on fine-mapped into an interval of 1.4 Mb, flanked by the markers GZ204 and IDP5. Here, through sequential fine mapping, *qRgls1* was finally mapped into an interval of 60Kb. BAC contigs were built to reconstruct the *qRgls1* genomic sequences for the resistant parent line (Y32) and six genes were predicted in the 60-Kb mapped region. Of them, three encode kinases and one encodes a receptor-like kinase (RLK). Transgenic verification and expression analysis demonstrated that *ZmWAK-RLK* is the causal gene of *qRgls1*. *ZmWAK-RLK* is anchored on the plasma membrane, potentially serving as a receptor to perceive and transduce extracellular signals. The allelic difference in the extracellular domain is the cause of the difference in GLS resistance, while the intracellular domains of the alleles have kinase activities. *ZmWAK-RLK* interacts with *ZmHR*-like to mediate the GLS resistance. Taken together, the *qRgls1* could find its usefulness in the GLS resistant breeding in maize.

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Abstract 187

MicroRNA 对其靶基因的调控模式分析

周巧, 徐洁, 卢艳丽*

四川农业大学玉米研究所, 成都, 611130

*通讯作者: 63680982@qq.com

(submitted by 周巧<921704684@qq.com>)

摘要: MicroRNA (miRNA) 是一类大小约 21-24 nt 的内源性 RNA 分子, 在真核生物的器官建成、生长发育、信号传导及逆境胁迫响应等方面具有重要的作用。研究表明, miRNA 通过和目标靶基因 mRNA 互补配对, 对其切割或抑制其翻译, 进而在转录后水平或翻译水平上调控靶标基因的表达。目前, 大量研究证明 miRNA 能够有效降低其靶基因的表达水平。课题组前期通过小 RNA 测序及转录组联合分析发现, 部分 miRNA 与其靶基因之间的表达存在显著的正相关。为了解释这一现象, 本研究选取了测序结果中与其靶基因呈显著正相关的三组 miRNA (miR528、miR168、miR166) 进行实验验证。通过实时荧光定量 qPCR 检测玉米自交系 AC7643、AC7729/TZSRW 根系在正常灌溉和水分胁迫下 miRNA 和其靶基因表达水平, 结果与测序结果吻合, 即 miRNA 表达与其靶基因呈显著正相关。通过构建以目标 miRNA 为核心的基因共表达网络发现, miRNA 的模拟靶标基因与 miRNA 靶基因表达也显著相关。为进一步探究 miRNA 模拟靶标在 miRNA 对靶基因调控中的作用, 本研究分别构建了 miR528、miR168、miR166 三个 miRNA 的过表达载体及其对应的具有 miRNA 靶位点序列的荧光报告载体 (报告基因为 *eGFP*) 以及具有 miRNA 模拟靶位点序列的荧光报告载体 (报告基因为 *mCherry*), 在烟草中瞬时表达, 通过荧光强度变化检测 miRNA 与其靶基因和模拟靶标基因间的调控关系。结果发现, 当 miRNA 过表达载体与具有 miRNA 靶位点序列的荧光报告载体按 1:1 的比例共注射时, *eGFP* 其荧光强度低于单独注射 miRNA 靶位点序列载体的荧光强度。同样地, 当 miRNA 过表达载体与具有 miRNA 模拟靶位点序列的荧光报告载体按 1:1 的比例共同注射时, *mCherry* 其荧光强度也低于单独注射 miRNA 模拟靶位点序列载体的荧光强度。但将三者按 1:1:1 的比例一同注射时, *eGFP* 和 *mCherry* 的荧光强度较其各自与 miRNA 过表达载体共注射时的荧光强度均有所增强, 但未恢复至其单独注射时的荧光强度。这些结果说明 miRNA 模拟靶标基因可能通过吸附 miRNA 来阻碍 miRNA 对靶基因的调控作用, 以此间接地提高靶基因的表达, 导致 miRNA 与其靶基因之间出现正相关的表达关系。

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Abstract 188

A microtubule-associated protein was found to be involved in maize grain development.

Qingqian Zhou, Hongqiu Wang, Jihua Tang, Guifeng Wang and Zhiyuan Fu*

Key Laboratory of Wheat and Maize Crops Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou 450002, China;

*Corresponding author: fuzhiyuan2004@163.com

(Submitted by Qingqian Zhou <zqingqian2016@163.com>)

Abstract: Maize (*Zea mays* L.) is one of the most important cereal crops and is a model organism for plant genetic and molecular biology studies. Kernel is a main organ of interest for yield increase and quality improvement of maize. Here, we characterized a maize dek mutant, which produces small and collapsed kernels, leading to embryos and/or seedlings lethality. The causal gene was in a region of about 124kb covering 8 candidate genes, by analyzing an F₂ population of 32000 individuals. DNA sequencing indicated that GA deletion in the last exon of A gene, encodes a microtubule-associated protein that participates in the division and differentiation of endosperm cells, resulted in premature termination of translation. The Y2H and BiFC were used to verify the interaction protein. We used native gel electrophoresis, western blot and immunofluorescence to observe the polymerization and morphology of microtubules. It was found that the mutants could not polymerized microtubules because of premature termination, resulted in decreased number of cells, and eventually led to seed defects. The functional investigation of A will bring new insight to multi-pathway theory of kernel size and expand gene resource for high yield breeding of maize.

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Abstract 189

Molecular characteristics of segment 5, a unique fragment encoding two partially overlapping ORFs in the genome of rice black-streaked dwarf virus

Hongyue Zu¹, Hong Zhang¹, Minhao Yao¹, Jiayue Zhang¹, Hong Di¹, Lin Zhang¹, Ling Dong¹, Zhenhua Wang^{1*}, Yu Zhou^{1*}

¹ Maize Research Institute, Northeast Agricultural University, Harbin 150030, China

*Corresponding author: zhenhuawang_2006@163.com and zhouyu0924@126.com

(submitted by 周羽<zhouyu0924@126.com>)

Abstract: Rice black-streaked dwarf virus (RBSDV), a ds-RNA virus in *Fijivirus* genus with family *Reoviridae* which transmitted by the small brown planthopper, is responsible for incidence of maize rough dwarf disease (MRDD) and rice black-streaked dwarf disease (RBSDD). To resolve the variation and evolution of S5, a unique fragment in the genome of RBSDV which encodes two partially overlapping ORFs (ORF5-1 and ORF5-2), we analyzed 127 sequences from maize and rice exhibiting symptoms of dwarfism from a total of eight locations, with four subgroups (A58: 58 RBSDV isolates from all eight geographic locations, B35: 35 RBSDV isolates from the Beijing location, J34: 34 RBSDV isolates from the Jining location, and T127: the combination of A58, B35, and J34). The nucleotide diversity of ORF5-1 ($\pi = 0.039$) and ORF5-2 ($\pi = 0.027$) both were higher than that of the overlapping region ($\pi = 0.011$) ($P < 0.05$). ORF5-2 was under the greatest selection pressure base on codon bias analysis and its activation mechanism was possibly influenced by the overlapping region. The recombinant fragments of three recombinant events (14NM23, 14BM20, and 14NM17) cross the overlapping region. Based on neighbor-joining tree analysis, the overlapping region could represent the evolutionary basis of the full-length S5, which was classified into three main groups. RBSDV populations were expanding and haplotype diversity resulted mainly from the overlapping region. The genetic differentiation of combinations (T127-B35, T127-J34, A58-B35, A58-J34, and B35-J34) reached significant or extremely significant levels. Gene flow was most frequent between subpopulations A58 and B35, with the smallest $|F_{st}|$ (0.02930). In short, we have proved that RBSDV populations were expanding and the overlapping region plays an important role in the genetic variation and evolution of RBSDV S5. Our results enable ongoing research into the evolutionary history of RBSDV-S5 with two partly overlapping ORFs.

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优质蛋白玉米的研究进展

周昱婕¹, 李明顺^{1*}

¹中国农业科学院作物科学研究所, 北京, 100081

*通讯作者: limingshun@caas.cn

(submitted by 周昱婕<13126550889@163.com>)

摘要: *Opaque2* (*O2*) 调控玉米籽粒发育的多个代谢途径, *o2* 突变致使转录功能丧失, 醇溶蛋白降低, 赖氨酸含量显著增加, 由此育种家可以利用 *o2* 及其修饰基因选育出优质蛋白玉米 (QPM)。我们利用回交转育和分子标记辅助选择技术将此突变基因导入到适合温带种植的玉米自交系中, 成功构建出多个普通玉米和糯玉米 *o2* 近等基因系 (*o2*-NILs), 有效扩充了我国 QPM 的种质资源。研究发现多数 *o2*-NILs 籽粒赖氨酸含量显著增加, 但在丹 598/*o2* 和辽 2345/*o2* 中保持不变。针对该现象进一步分析发现在基因导入和回交过程中, *o2* 基因启动子与受体材料发生重组, 因此部分近等基因系突变恢复, 赖氨酸含量保持不变。利用 *o2*-NILs 与普通玉米自交系配制杂交组合进行产量和配合力鉴定, 结果表明 *o2* 基因对产量有一定影响, 但在不同的遗传背景下影响不同; 分析杂交组合的玉米品质, 发现 *o2* 基因突变导致赖氨酸、蛋白质和油分含量大幅增加, 其余必需氨基酸含量也发生了显著变化。

对 *o2*-NILs 胚乳蛋白质组学分析发现, *o2* 基因的导入不仅减少了醇溶蛋白质含量, 同时显著影响了氨基酸合成、淀粉-蛋白质平衡、应激反应和信号转导相关的众多蛋白质积累。近期发现 *o2*-NILs 与受体亲本相比, 成熟籽粒淀粉含量显著改变。进一步解析其中的机制可完善 *O2* 蛋白调控网络, 有助于深入认识 *o2* 在玉米品质育种中的应用。

项目基金: 国家玉米产业技术体系, 国家育种专项

Abstract 191

Variations in an LRR-Receptor Like Kinase Gene, *SRR1*, Impart Different Quantitative Resistance to Seed and Ear Rot in Maize Heterotic Groups

Zijian Zhou¹, Kewei He¹, Peipei Ma¹, Jianxin Wu², Wei Wang², Jingyang Gao¹, Ming Ju¹, Cong Mu¹, Yabin Wu¹, Chaopei Dong¹, Yunxia Song¹, Zongliang Xia², Jiafa Chen², Huiyong Zhang^{2,*}, Jianyu Wu^{1,2,*}

¹National Key Laboratory of Wheat and Maize Crop Science, Collaborative Innovation Center of Henan Grain Crops, College of Agronomy, Henan Agricultural University, Zhengzhou, 450002, China

² College of Life Sciences, Synergetic Innovation Center of Henan Grain Crops and National Key Laboratory of Wheat and Maize Crop Science, Henan Agricultural University, Zhengzhou 450002, China

*Corresponding Author: wujianyu40@126.com

(submitted by 周子健<zhouzijian19900601@163.com>)

Abstract: *Fusarium verticillioides* is a widely-distributed soil-borne fungal species, which can be transmitted to seeds and causes seed rot, stalk rot, and ear rot by systemic infection in maize. The most significant single nucleotide polymorphism associated with seed rot in the *qFRI* region, which is a quantitative resistance locus in maize, has been identified in the *GRMZM2G009818* gene. Here, we demonstrated that *seed rot resistance 1* (*SRR1*, *GRMZM2G009818*) encodes a leucine-rich repeat-receptor like kinase that functions as a pattern recognition receptor and imparts quantitative resistance to seed rot caused by *F. verticillioides*. We demonstrated that *SRR1* is localized in the cytomembrane and can activate PAMP-triggered immunity, ETI, and salicylic acid signal after *F. verticillioides* inoculation. Three key functional variations in the coding region are responsible for five functional haplotypes of *SRR1*, which impart different resistance to seed and ear rot. An undesirable functional haplotype of *SRR1*, appears to have occurred after domestication of maize resulting in the loss of ear rot resistance; this haplotype accumulated in the TSPT heterotic group, which includes most of the male parents in Huang-Huai-Hai, the main maize producing area of China. Our results would help in better understanding of *SRR1*, an important quantitative resistance gene.

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Abstract 192

The PPR protein DEK41 affects *cis*-splicing of mitochondrial *nad4* intron 3 and seed development in maize

Chenguang Zhu ¹, Guangpu Jin ¹, Peng Fang ¹, Yan Zhang ¹, Xuzhen Feng ¹, Yuanping Tang ¹, Weiwei Qi ¹, Rentao Song ^{1,2 *}

¹ Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China.

² State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100193, China

*Corresponding author: rentaosong@cau.edu.cn

(submitted by 朱晨光<cgzhu@shu.edu.cn>)

Abstract: The splicing of organelle-encoded mRNA in plants requires nucleus-encoded proteins. The mechanism of organelle mRNA splicing and the factors involved in this process are not well understood. Pentatricopeptide repeat (PPR) proteins are known to participate in such RNA-protein interactions. Maize *defective kernel 41* (*dek41*) is a seedling-lethal mutant that causes developmental defects. The *Dek41* gene was cloned by *Mu* tag isolation and allelic confirmation, and was found to encode a P-type PPR protein that targets mitochondria. Mitochondrial RNA transcript profile analysis revealed that *dek41* mutations cause reduced splicing efficiency of mitochondrial *nad4* intron 3. Immature *dek41* kernels exhibited severely reduced complex I assembly and NADH dehydrogenase activity. The up-regulated expression of alternative oxidase genes and deformed inner cristae of mitochondria in *dek41*, as revealed by transmission electron microscopy, indicate that proper splicing of *nad4* is essential for mitochondrial functions and morphology. Consistent with this finding, differentially expressed genes in the *dek41* endosperm included those related to mitochondrial function and activity. Our results indicate that DEK41 is a PPR protein affecting *cis*-splicing of mitochondrial *nad4* intron 3 that is required for mitochondrial function and maize kernel development.

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Gene Mapping and Cloning of Two Maize Kernel Mutants

Wanchao Zhu, Peng Tian, Sijia Chen, Mingzhu Li, Yan Liang, Lin Li*

National Key Laboratory of Crop Genetic Improvement, Huazhong Agricultural University, Wuhan 430070, China

*Corresponding author: hzaulilin@mail.hzau.edu.cn

(submitted by Wanchao Zhu <qfdwxzwc@163.com>)

Abstract: Maize (*Zea mays*) is one of the most important crops in the world. Its kernel, composed by the embryo and endosperm, is the primary organ for nutrient storage and the direct component of production. However, the regulatory mechanism of kernels development remains largely unknown. In this study, we combined multiple methods to map two maize kernel mutants which included one classic mutant *sem1* and one novel isolated mutant named as *ppr-like*. The *sem1* showed both a smaller embryo and endosperm than the wildtype while the *ppr-like* exhibited a small endosperm and no embryo. Combining SHOREmap and BSR-seq, the *SEM1* gene was genetically mapped to a 1-M genomic interval. Then, the interval was narrowed down to 260 kb by map-based cloning using 4000 mutant kernels. The novel mutant *ppr-like* was isolated and identified through phenotypic characterization and Chi-square test, which showed a perfect 3:1 segregation and exhibited recessive inheritance. After RNA extraction and sequencing, we used two strategies including BSR-seq and MMAPPR to map the mutant, which consistently mapped the candidate gene into a about 35-M genomic interval on the chromosome 1. Our results will be helpful to clone the key genes which controlled kernel development and accelerate the process of understanding the development mechanism of kernels.

玉米中介体亚基ZmMED7的初步功能分析

王晶, 赵军, 宗娜*

中国农业科学院生物技术研究所, 北京, 100081

*通讯作者: zongna@caas.cn

(submitted by 宗娜<zongna@caas.cn>)

摘要: 转录中介复合体是能够调控转录的蛋白复合体, 它能与转录因子相互作用, 增强转录因子的功能。转录中介体在植物对植物的生长发育的不同阶段都起到了重要作用, 并且还参与胁迫应答反应。我们从玉米 Qi319 中扩增出了与拟南芥具有高度同源性的转录中介体亚基基因 *ZmMED7-1* 和 *ZmMED7-2*。通过分析 *ZmMED7-1* 和 *ZmMED7-2* 在玉米不同发育时期和不同激素及其逆境条件下的表达谱、亚细胞定位和转基因拟南芥对干旱和高盐胁迫的抗性, 对该中介亚基在植物生长发育过程中的功能进行初步的分析。结果表明: (1) *ZmMED7-1* 和 *ZmMED7-2* 在玉米种子萌发初期, 营养生长阶段不同时期的根和叶, 未成熟雄穗、吐丝雌穗和授粉后不同时期的籽粒和胚乳中均有不同程度的表达; (2) *ZmMED7-1* 和 *ZmMED7-2* 在玉米叶片中的表达受 BR、IAA 和 GA₃ 的诱导, 并受到 ABA、盐和干旱等处理的抑制; (3) MED7 蛋白定位在细胞核和细胞质中; (4) *ZmMED7-1* 和 *ZmMED7-2* 在转基因拟南芥中过量表达显著降低了拟南芥植株对干旱、高盐胁迫的耐受性。上述结果表明, 玉米转录中介亚基 MED7 参与了在植物对非生物逆境的应答过程。我们将进一步研究 *ZmMED7-1* 和 *ZmMED7-2* 在玉米逆境应答中的生物学功能及其分子机理

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***Dek42* encodes an RNA binding protein that affects alternative pre-mRNA splicing and maize kernel development**

Yi Zuo¹, Fan Feng², Weiwei Qi², Rentao Song^{1,*}

1 State Key Laboratory of Plant Physiology and Biochemistry, National Maize Improvement Center, Beijing Key Laboratory of Crop Genetic Improvement, Joint International Research Laboratory of Crop Molecular Breeding, College of Agronomy and Biotechnology, China Agricultural University, Beijing 100094, China

2 Shanghai Key Laboratory of Bio-Energy Crops, Plant Science Center, School of Life Sciences, Shanghai University, Shanghai 200444, China

*Corresponding author: rentaosong@cau.edu.cn

(submitted by 左毅<zuoyi88@cau.edu.cn>)

Abstract: RNA binding proteins (RBPs) play an important role in posttranscriptional gene regulation. However, the functions of RBPs in plants remain poorly understood. Maize kernel mutant *dek42* has small defective kernels and lethal seedlings. *Dek42* was cloned by *Mutator* tag isolation and further confirmed by an independent mutant allele and CRISPR-Cas9 materials. *Dek42* encodes an RRM_RBM48 type RNA binding protein that localizes to the nucleus. *Dek42* is constitutively expressed in various maize tissues. The *dek42* mutation caused a significant reduction in the accumulation of DEK42 protein in mutant kernels. RNA-seq analysis showed that the *dek42* mutation significantly disturbed the expression of thousands of genes during maize kernel development. Sequence analysis also showed that the *dek42* mutation significantly changed alternative splicing in expressed genes, which were especially enriched for the U12-type intron retained type. Yeast two-hybrid screening identified SF3a1 as a DEK42-interacting protein. DEK42 also interacts with the spliceosome component U1-70K. These results suggested that DEK42 participates in the regulation of pre-mRNA splicing through its interaction with other spliceosome components. This study showed the function of a newly identified RBP and provided insights into alternative splicing regulation during maize kernel development.

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