本科生发表论文情况

序号	学生姓名	位次	论文名称	期刊	年份及卷期页码
1	Jin Liu(刘锦)	3	Transcriptomic changes in Nicotiana benthamiana plants inoculated with the wild type or an attenuated mutant of Tobacco vein banding mosaic virus	Molecular Plant Pathology	2017, 18(8): 1175-1188
2	Peng Ji（季鹏）, Weiguang Li（李伟光）, Yuxin Zheng（郑玉欣）, Zhaohui Wang（王朝辉）, Qixin Huo（霍启欣）, Chengyao Hua（华程遥）	1 至 6	Isolation and Identification of Four Novel Biocontrol Bacillus Strains against Wheat Sharp Eyespot and their Growth-promoting Effect on Wheat Seedling	International Journal of Agriculture and Biology	2019, 21: 282-288
3	Weiguang Li（李伟光）, Peng Ji（季鹏）, Qinzheng Zhou（周勤政）, Chengyao Hua（华程遥）	1 至 4	Insights into the synergistic biodegradation of waste papers using a combination of thermostable endoglucanase and cellobiohydrolase from Chaetomium thermophilum	Molecular Biotechnology	2018, 60(1): 49-54
4	Xiutao Chen（陈秀涛）, Weiguang Li（李伟光）, Peng Ji（季鹏）, Yang Zhao（赵洋）, Chengyao Hua（华程遥）	1 至 5	Engineering the conserved and noncatalytic residues of a thermostable β-1,4-endoglucanase to improve specific activity and thermostability	Scientific Reports	2018, 8: 2954-2963
5	Qinzheng Zhou（周勤政）, Peng Ji（季鹏）, Jianye Zhang（张建业）, Xue Li（李雪）	1 至 4	Characterization of a novel thermostable GH45 endoglucanase from Chaetomium thermophilum and its biodegradation of pectin	Journal of Bioscience and Bioengineering	2017, 124(3): 271-276
6	Qinzheng Zhou（周勤政）, Jianchao Jia（贾建超）, Peng Ji（季鹏）	1 至 3	A novel application potential of GH6 cellobiohydrolase CtCel6 from thermophilic Chaetomium thermophilum for gene cloning,	International Journal of Agriculture and Biology	2017, 19(2): 355-362

			heterologous expression and biological characterization		
7	Chengyao Hua（华程遥），Weiguang Li（李伟光）	并列第一	Characterization of a novel thermostable GH7 endoglucanase from Chaetomium thermophilum capable of xylan hydrolysis	International Journal of Biological Macromolecules	2018, 117: 342-349
8	Yifan Liu（刘一帆），Mengyu Liu（刘梦宇），Siqi Wang（王思琦）	2至4	Improving the thermostability of a thermostable endoglucanase from Chaetomium thermophilum by engineering the conserved noncatalytic residue and N-glycosylation site	International Journal of Biological Macromolecules	2020, 164:3361-3368
9	Ruirui Yang（杨瑞睿），Yanxu Sun（孙衍旭），Mengyu Liu（刘梦宇），Lifan Zhou（周立凡）	2至5	Identification and characterization of a novel hyperthermostable bifunctional cellobiohydrolase-xylanase enzyme for synergistic effect with commercial cellulase on pretreated wheat straw degradation	Frontiers in Bioengineering and Biotechnology	2020, 8:296-308
10	Xiaoyuan Hou（侯晓媛）	4	A Conserved Glycoside Hydrolase Family 7 Cellobiohydrolase PsGH7a of Phytophthora sojae Is Required for Full Virulence on Soybean	Frontiers in Microbiology	2020, 11:1285-1296
11	Yanxu Sun（孙衍旭），Ruirui Yang（杨瑞睿），Mengyu Liu（刘梦宇），Siqi Wang（王思琦），Yifan Liu（刘一帆），Lifan Zhou（周立凡）	3至8	Improvement of the catalytic activity and thermostability of a hyperthermostable endoglucanase by optimizing N-glycosylation sites	Biotechnology for Biofuels	2020, 13:30-40
12	Weiguang Li（李伟光），Chengyao Hua（华程遥）	2和3	Enhancement of catalytic activity and thermostability of a thermostable cellobiohydrolase from Chaetomium thermophilum by site-directed mutagenesis	International Journal of Biological Macromolecules	2018, 116: 691-697

13	Yang Liu（刘阳）	2	Selection of organosilicone surfactants for tank-mixed pesticides considering the balance between synergistic effects on pests and environmental risks.	Chemosphere	2019, 217, 591-598
14	Yang Liu（刘阳）	3	Easily tunable membrane thickness of microcapsules by using a coordination assembly on the liquid-liquid interface.	Frontiers in Chemistry	2018,6:387
15	Yang Liu（刘阳）	4	Alcohol ethoxylates significantly synergize pesticides than alkylphenol ethoxylates considering bioactivity against three pests and joint toxicity to Daphnia magna.	Science of the Total Environment	2018, 644, 1452-1459
16	Hao Su（苏浩), Xiaoqi Fan（范晓琪), Xiaotian Zhang（张笑天）	7 至 9	Identification of Anthocyanins Compositions and Functional Analysis of An Anthocyanin Activator in Solanum nigrum Fruits.	Molecules	2017,22,876
17	Wei Tang(唐伟)	4	First report of Tobacco vein banding mosaic virus infecting sesame in China	Plant Disease,	2017, 101(5):850
18	Dejie Cheng(程德杰)	1	First Report of Papaya ringspot virus associated with a ringspot disease of Zucchini in Northern China.	Plant Disease,	2017,101(5):847
19	Dejie Cheng(程德杰)	3	Tobacco vein banding mosaic virus 6K2 protein hijacks NbPsb01 for virus replication.	Scientific Reports	2017 年 2 月
20	徐子雯、方倩囡、曹玉博	并列第一	套作糯玉米对西兰花连作田土壤微生物量及酶活性的影响	生态学杂志	2016,35(1):149-157
21	黄峤璟	2	耕种模式对三江平原黑土细菌多样性的影响	山东农业科学	2016,48(7);76-81
22	靳帅	1	基于 DNDC 模型的秸杆还田量与氮肥的耦合效应对夏玉米农田 N2O 排放的影响研究	山东农业科学	2016,48(2):68-73

23	徐文文	4	Research on Production Conditions of High-grade Wine from Northern Blueberry	Agricultural Science &Technology	2016,17(2):417-419,448
24	徐文文	1	Effects of Seaweed Bio-organic Fertilizer on Growth and Yield of Winter Wheat	Agricultural Science &Technology	2016,17(11):2555-2559
25	郑宾、赵伟、徐峥、高大鹏、姜媛媛	1月5日	不同耕作方式与氮肥类型对夏玉米光合性能的影响	作物学报	2017,43(6):925-934
26	付苒、胡元峰	1	2016年山东省玉米种植成本收益分析	农业展望	2017年第7期37页
27	戴云驰、张鑫	并列第一	根瘤菌、超声和2,4-D处理对小麦幼苗生长的影响	山东农业大学学报	2018,49(2):240-243
28	阚茗溪	1	适期晚播对不同密度冬小麦产量和茎秆抗倒性能的调控效应	中国农学通报	2019,35(32):1-6
29	柏慧	1	氮肥水平对强筋小麦产量和氮素利用的影响	中国农学通报	2020,36(4):7-14
30	李仲烨	1	Multi-environments and multi-models association mapping identified candidate genes of lint percentage and seed index in Gossypium hirsutum L.	Molecular Breeding	(2019) 39:149
31	王菲	1	白花丹参和紫花丹参叶绿素荧光日变化比较研究	Special Wild Economic Animal and Plant Rescarch	2019.01.003
32	王菲	1	间作大葱对桔梗根系分泌物的影响	山东农业科学	2019,51(11):68~73

33	张琳、殷宇琪、边央	并列第一	一个新的丹参 CYP450 的克隆和生物信息学的分析	基因组学与应用生物学	2020,39（12）:5718- 5723
34	甄天悦	1	增施磷肥提高弱光环境中夏大豆叶片光合能力及产量	作物学报	2020,46(2): 249-258
35	李心如	5	OsHsfB4d Binds the Promoter and Regulates the Expression of OsHsp18.0-CI to Resistant Against Xanthomonas oryzae	Rice	(2020)13:28
36	赖旻祎、宋吉媛、田岳	并列第一	丹参水肥一体化对杂草生物量的影响	山东农业科学	2020,52(8):96 ~99
37	穆如宇	2	粗山羊草响应盐胁迫转录组分析	分子植物育种	2020, 18（21）: 7015-7022
38	吴紫萱	1	花生种皮颜色的研究进展	山东农业科学	无
39	马乙傲	1	授粉后降雨对玉米杂交种结实以及籽粒性状的影响	山东农业科学	2021,53(9):27~32
40	范国瑞	1	Germination characteristics of maize seeds with high and low vigour levels in response to on-farm seed primining	Seed Science and Technology	2021.49.2.04
41	杜斌、李宗尧	并列第一	有机肥施用及合理密植提高黄淮海地区夏大豆光系统性能与籽粒产量	植物营养与肥料学报	2021,27(8):1361-1375
42	王孜禾	2	中药渣制作平菇栽培种试验	食用菌	2019, 41（4）: 43~44
43	杨瑞睿	2	海南省永兴岛革螨新纪录（蜱螨亚纲:中气门目）	中国媒介生物学及控制杂志	2018 年 12 月第 29 卷第 6 期

44	李晨雨，臧传江 ，朱少杰	1 至 3	新烟碱类杀虫剂氟吡呋喃酮的研究开发现状与展望	农药	Vol.57,No.11 Nov.2018
45	柳浩琪	8	赤霉素对烟草幼苗生长发育的影响	山东农业科学	2019，51(4):139 ～143
46	杨阳	1	双孢蘑菇液体菌种培养基筛选及培养条件的优化	山东农业大学学报（自然科学版）接收	2018,49(3):393-395,401
47	宋超群	2	红平菇液体发酵培养基优化	食品研究与开发	2017/11/27
48	程德杰	1	甘蔗花叶病毒两个山东分离物的全基因组序列分析	植物病理学报，	2017，47(3)：357-363
49	段入心	并一	取食行为对丽蝇蛹集金小蜂雌雄成虫体内可培养真菌的影响	应用昆虫学报	2017 年 5 月
50	王立超，郑洁，李佩瑜	3 至 5	6 种杀虫剂对侧柏幼苗根系活力的影响	山西林业科技	2016 年 3 月第 45 卷 第 1 期
51	王立超，郑洁，李佩瑜	4 至 6	三种农药对苹果幼苗根系活力的影响	河北果树	2016.01.003
52	王立超	4	三种杀虫剂对桃树叶片叶绿素含量的影响	河北果树	2016.02.005
53	Mengyu Liu(刘梦宇)	3	Improving the thermostability of a thermostable endoglucanase from Chaetomium thermophilum by engineer the conserved no catalytic residue and N-glycosylation site	International Journal of Biological Macromolecules	（2020）3361－3368
54	王昭	2	德州市农作物病虫害统防统治发展现状及对策	德州学院学报	2019 年 12 月第 35 卷第 6 期

55	刘华新、王佳钰	1	Lauric acid is a potent biological control agent that damages the cell membrane of Phytophthora sojae	Frontiers in Microbiology	2021,12: 666761
56	Guangrui Cui(崔光睿)	3	Biological Activity of trans -2-Hexenal Against the Storage Insect Pest Tribolium castaneum (Coleoptera: Tenebrionidae) and Mycotoxigenic Storage Fungi	Journal of Economic Entomology	2021，114（2）：979-987.
57	张建	2	植物免疫诱抗剂的发现、作用及其在农业中的应用	世界农药	2020，42（10）：24-31.
58	周倩	1	草地贪夜蛾对山东玉米的为害情况研究进展	园艺学报	2021，已录用。
59	张俊杰	1	虫螨腈及其代谢物在韭菜和姜上的残留检测》	农药科学与管理	2021，42（6）：35-40.
60	张俊杰	3	新型介孔材料-QuEChERS-超高效液相色谱串联质谱法检测茶叶中 10 种农药残留》	分析化学	2021，49（5）：830-838.
61	Bao-Zhen Jiang(姜宝珍)	3	Janthinoid A, an Unprecedented Tri-nor-meroterpenoid with Highly Modified Bridged 4a,1-(Epoxymethano)phenanthrene Scaffold, Produced by the Endophyte of Penicillium janthinellum TE-43	Organic Letters	2021，DOI: 10.1039/d1qo01066b
62	姜伟涛	2	添加适宜氮磷对连作平邑甜茶幼苗生长及土壤环境的影响	植物生理学报	2017，53（9）：1687-1694
63	马雪婷	2	基于 web of science 的土壤微生物研究文献国际发展态势分析	北方园艺	2017（10）：198-207
64	王楚堃	1	纤维素降解菌剂对桃园土壤养分及果实品质的影响	山东农业科学	2017，49（3）：94-96

65	王培辉、王威振、谭晓雪、张敏、韩金赢	2	食用向日葵盆栽种植土壤优化的初步研究	现代园艺	2018 年第 10 期
66	吴琦杰	1	sIdentification and characterization of cherry genes in response to parthenocarpy induced by GA3 through transcriptome analysis	BMC Genetic	2019 年
67	顾超珩	1	硫代腺苷甲硫氨酸促进番茄百菌清讲解的生理机制	中国农业科学	2019，52（6）：1058-1065
68	车美美，袁依馨，赵一潼	2	局部供磷条件下苹果幼苗根系形态的适应性变化与磷素吸收	中国果树	2019（5）：16-19
69	姜乐璞，姚建，张兆梁，韩福 ，尚将 ，李玲	1	M9T337 自根砧嫁接不同苹果品种的植物学特性调查	园艺与种苗	2019，39（01）：32-34
70	芮麟	1	丽江市青贮饲用玉米高产栽培技术	农业科技通讯	2019.3：157-158
71	芮麟	1	不同品种及栽培措施对青贮饲用玉米产量的影响	农业科技通讯	2019.4：114-116
72	周昊然	1	Study on the microbial community in earthworm and soil under cadmium stress based on contour line analysis	Environmental Science and Pollution Research	2019 年第 26 卷第 20 期
73	马宇晨、赵玉梅、黄丹霖（园艺学院第三作者）、张梦晴、吴晓雨、王洁、陈雨、黄家保、段巧红	3	白菜 CesA 基因家族鉴定及表达模式分析	广西植物	2022 年
74	钱犇	1	生物促生剂在水和土壤治理中的应用分析	建材技术与应用	2021

75	陈雨、邓九州	1,2	白菜 DBB 基因家族的鉴定与表达分析	山东农业大学学报(自然科学版)	2021，52（2）：174-181
76	徐迎澳	1	桃砧木耐涝性研究进展	落叶果树	2021，53(6):38-43
77	赵文哲(园艺学院第一作者），陈修德，邓文鹏，吴琦杰，杜桂英，和华杰，王伟，周涛，肖伟，李玲	1	外源硅处理对草莓果实果胶物质降解的影响	植物生理学报	2021,57(10):1926-1936
78	赵文哲(园艺学院第一作者)，刘晓，杜桂英，邓文鹏，宋新卫，肖伟，李玲	1	干旱胁迫及复水对‘M9T337’苹果砧木苗生理特性的影响	山东农业科学	已接受
79	赵文哲(园艺学院第一作者)，刘晓， 姜珊， 陈敏，王旭旭， 刘金宝， 谭梅，王宇，李玲,高东升,陈修德,肖伟	1	干旱过程中‘M9T337’苹果砧木苗光合特性及 MdCP2 与 MdGLK1 互作分析	植物生理学报	2021,57(6):1337-1348
80	亓正聿、毕泽楷	1,2	果树耐盐机制研究进展	种子科技	2021 年 11 期：8-11
81	毕泽楷、亓正聿、金梦、李恩		ROP 信号途径调控花粉管极性生长的分子机理研究进展	分子植物育种	2021
82	贺强	1	阳荷植物组织培养技术的研究	信息科技	2021
83	Z. Zhao1,2, S.Yu1,2, X. Han1,2* and S. Yang3，赵子昕（园艺学院第一作者）	1	Influence of drought and dry-wet alternation on nitrogen transformation and low abundance microorganisms in tea garden soil	Journal of Environmental Biology	2022
84	张鸿锦	3	Identification of potential pathways associated with indole-3-butyric acid in	Functional & Integrative Genomics	2021

			citrus bud germination via transcriptomic analysis		
85	吴双敏　陈晨	1,2	三种成膜材料对“赤霞珠”枝条的保水效果研究	《中外葡萄与葡萄酒》编辑部	2021
86	芮麟	1	以“粮改饲”为主线，促进绿色供给侧结构性改革向纵向发展	农业科技通讯	2018.3：28-30
87	张楚乔	1	土地利用规划环境影响评价的相关问题分析	《商情》	2021,9(35):139-141
88	张楚乔	1	城市规划与土地利用规划衔接问题分析	《科学与财富》	2021,13(9):128-129
89	邹今朝	1	土地资源管理及其可持续利用研究	《区域治理》	2021,30(308):06
90	王洪佳	1	农田生态系统恢复力评价与调控机制	汉斯期刊	2021,10.12677/SD.2021.115071
91	柏逸飞	1	基于地学信息图谱的2000-2020年南京城市扩张特征对比分析	《城市建筑》杂志	2022，已接受

RESEARCH ARTICLE SUMMARY

PLANT SCIENCE

Horizontal gene transfer of *Fhb7* from fungus underlies *Fusarium* head blight resistance in wheat

Hongwei Wang*†, Silong Sun*, Wenyang Ge*, Lanfei Zhao*, Bingqian Hou*, Kai Wang*, Zhongfan Lyu*, Liyang Chen, Shoushen Xu, Jun Guo, Min Li, Peisen Su, Xuefeng Li, Guiping Wang, Cunyao Bo, Xiaojian Fang, Wenwen Zhuang, Xinxin Cheng, Jianwen Wu, Luhao Dong, Wuying Chen, Wen Li, Guilian Xiao, Jinxiao Zhao, Yongchao Hao, Ying Xu, Yu Gao, Wenjing Liu, Yanhe Liu, Huayan Yin, Jiazhu Li, Xiang Li, Yan Zhao, Xiaoqian Wang, Fei Ni, Xin Ma, Anfei Li, Steven S. Xu, Guihua Bai, Eviatar Nevo, Caixia Gao, Herbert Ohm, Lingrang Kong†

INTRODUCTION: *Fusarium* head blight (FHB) is a fungal disease that devastates global wheat production, with losses of billions of dollars annually. Unlike foliar diseases, FHB occurs directly on wheat spikes (inflorescences). The infection lowers grain yield and also causes the grain to be contaminated by mycotoxins produced by the *Fusarium* pathogen, thus imposing health threats to humans and livestock. Although plant breeders have improved wheat resistance to FHB, the lack of wheat strains with stable FHB resistance has limited progress.

RATIONALE: Many genetic loci in wheat affect FHB resistance but most only have minor effects; only a few exhibit a stable major effect on resistance. Wheat relatives in the Triticeae tribe carry resistant genes to different diseases including FHB and thus can be alternative sources of FHB resistance for wheat breeding. *Thinopyrum* wheatgrass has been used as a source of beneficial genes transferable to wheat by distant hybridization breeding since the 1930s. *Fhb7,* a gene transferred from *Thinopyrum* to wheat, showed a stable large effect on FHB resistance. However, the lack of a *Thinopyrum* reference genome hampered gene cloning and marker development, delaying the use of *Fhb7* in wheat breeding. Here, we cloned *Fhb7* using a reference assembly that we generated for *Th. elongatum* and characterized its resistance mechanisms and evolutionary history.

RESULTS: Using sequence data from *Th. elongatum*, we assembled the Triticeae E reference genome with 44,474 high-confidence genes annotated. Using this reference, we genetically mapped *Fhb7* and located it to a 245-kb genomic region. We determined a gene encoding a glutathione S-transferase (GST) as *Fhb7* by virus-induced gene silencing and evaluated mutants and transgenic plants. We discovered that Fhb7 detoxifies pathogen-produced trichothecene toxins by conjugating a glutathione (GSH) unit onto the epoxide moieties of type A and B trichothecenes. *Fhb7* GST homologs are absent in the plant kingdom, but one sequence showing ~97% identity with *Fhb7* was found in endophytic fungi of an *Epichloë* species that establishes symbiosis with temperate grasses. This result suggests that *Fhb7* might have been transferred from *Epichloë* to *Th. elongatum* through horizontal gene transfer. Finally, we demonstrated that *Fhb7*, when introgressed into diverse wheat backgrounds by distant hybridization, confers broad resistance to both FHB and crown rot without penalizing wheat yield. Our results suggest a source of *Fusarium* resistance for wheat improvement.

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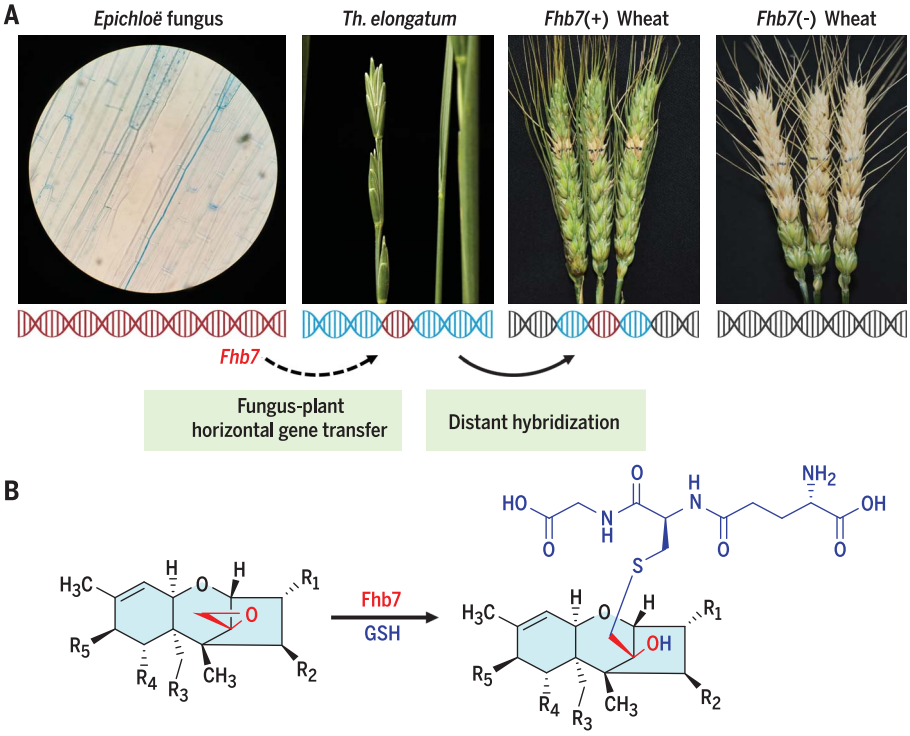
CONCLUSION: *Th. elongatum* carries biotic and abiotic resistance genes and is a useful resource for wheat breeding. The assembled *Th. elongatum* reference genome can aid identification and cloning of such genes for wheat improvement. Cloning of *Fhb7* revealed that it encodes a GST that can detoxify trichothecene toxins. Thus, *Fhb7* resistance differs from *Fhb1* resistance, which depends on a reduction of pathogen growth in spikes, although both confer durable resistance. The ability of *Fhb7* to detoxify multiple mycotoxins produced by various *Fusarium* species demonstrates its potential as a source of resistance to the various diseases for which *Fusarium* trichothecenes are virulence factors. The deployment of *Fhb7* in commercial wheat cultivars could alleviate both the food safety issue for consumers and the yield loss problem for growers. Sequence homologies between fungal and plant *Fhb7* suggested that horizontal gene transfer may help to shape plant genomes. ■



***Fhb7* confers FHB resistance by detoxifying trichothecenes.** (**A**) *Fhb7* in *Th. elongatum* genome likely came from an *Epichloë* fungus through horizontal gene transfer. *Fhb7* drives FHB resistance when introgressed from *Thinopyrum* into wheat. (**B**) *Fhb7* encodes a GST that detoxifies *Fusarium*-produced trichothecenes by conjugating GSH (blue) to the epoxy group (red). R_1 to R_5 refer to the variable groups in trichothecenes.

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RESEARCH ARTICLE

PLANT SCIENCE

Horizontal gene transfer of *Fhb7* from fungus underlies *Fusarium* head blight resistance in wheat

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Fusarium head blight (FHB), a fungal disease caused by *Fusarium* species that produce food toxins, currently devastates wheat production worldwide, yet few resistance resources have been discovered in wheat germplasm. Here, we cloned the FHB resistance gene *Fhb7* by assembling the genome of *Thinopyrum elongatum*, a species used in wheat distant hybridization breeding. *Fhb7* encodes a glutathione S-transferase (GST) and confers broad resistance to *Fusarium* species by detoxifying trichothecenes through de-epoxidation. *Fhb7* GST homologs are absent in plants, and our evidence supports that *Th. elongatum* has gained *Fhb7* through horizontal gene transfer (HGT) from an endophytic *Epichloë* species. *Fhb7* introgressions in wheat confers resistance to both FHB and crown rot in diverse wheat backgrounds without yield penalty, providing a solution for *Fusarium* resistance breeding.

Wheat (*Triticum aestivum* L.) is a leading source of calories for the human population (*1*). The prevalence and widespread outbreaks of the devastating *Fusarium* head blight (FHB) disease, exacerbated by recent changes in climate and certain cropping practices, has posed a threat for global wheat production and food safety. *Fusarium* species cause not only FHB in wheat, barley, and oat, but also crown rot in wheat and ear rot in maize. However, *F. graminearum* is the prominent pathogen of wheat FHB in China, the United States, Canada, Europe, and many other countries (*2*). *Fusarium* produces epoxy-sesquiterpenoid compounds known as trichothecenes, some examples of which are deoxynivalenol (DON), T-2 toxin, HT-2 toxin, and nivalenol (NIV), among others; these compounds are inhibitors of protein synthesis and virulence factors for pathogenicity (*2*). Trichothecene contamination in cereal grain results in immunotoxicity and cytotoxicity in humans and animals and thus has aroused public safety concerns (*3*). Despite global screening efforts examining tens of thousands of wheat accessions, a limited number of quantitative trait loci (QTLs) have been verified to confer a stable effect on FHB resistance (*4*). *Fhb1* on chromosome 3B is the only QTL that has been used in breeding programs worldwide. Although it has been cloned from different Chinese wheat sources, its molecular identity and resistance mechanisms remain equivocal (*5*–*8*).

Wheat relatives have proven to be alternative sources for improvement of resistance to both biotic and abiotic stresses in wheat (*9*). Distant hybridization, the practice of making crosses between two different species, genera, or higher-ranking taxa, makes it possible to transfer alien genes from Triticeae tribe relatives to wheat (*9*–*11*). Tall and intermediate wheatgrasses of the *Thinopyrum* genus (forage grasses) are sources of resistance to salinity, drought, and disease for wheat. Several disease resistance genes, including stem rust (e.g., *Sr24*, *Sr25*, *Sr26*, *Sr43*, *Sr44*, and *SrB*), leaf rust (*Lr19*, *Lr24*, *Lr29*, and *Lr38*), powdery mildew (*Pm40* and *Pm43*), barley yellow dwarf virus (*Bdv2* and *Bdv3*), and *Fusarium* head blight (*Fhb7*), have been introduced from *Thinopyrum* into wheat for resistance breeding (*10*, *12*–*16*).

Fhb7 is a QTL introduced from *Thinopyrum elongatum* and shows a similar effect on FHB resistance as *Fhb1*. *Th. elongatum* (syn. *Agropyron elongatum* or *Lophopyrum elongatum*), a grass of the Triticeae family with a diploid E genome ($2n = 2x = 14$), is native to Eurasia and is thought to be a genome donor species for various tetra-, hexa-, and even decaploid species in the *Thinopyrum* genus (*14*). The lack of a reference sequence for the E genome has impeded the process of cloning and the development of diagnostic markers for the deployment of *Fhb7* and other E genome–derived resistance genes. Here, we report the assembly of a reference genome for *Th. elongatum* and describe the cloning and biomolecular characterization of *Fhb7*. Using the newly assembled E genome reference, we identified a GST gene as a candidate for *Fhb7* by map-based cloning and confirmed its function in FHB resistance using transgenics. Fhb7 can detoxify trichothecenes by catalyzing the conjugation of a glutathione (GSH) unit onto their toxic epoxide moiety. *Fhb7*'s coding sequence has no obvious homology to any known sequence in the entire plant kingdom but shares 97% sequence identity with a species of endophytic fungus (*Epichloë aotearoae*) known to infect temperate grasses, which provides evidence that *Fhb7* in the *Th. elongatum* genome might be derived from the fungus through HGT. We demonstrate here that *Fhb7* confers resistance to both FHB and crown rot without yield penalty in wheat.

Results

Th. elongatum genome assembly and evolution

To sequence and assemble the genome of *Th. elongatum*, 1.1 Tb of high-quality sequence reads were generated from a series of libraries, which is about 236× coverage of the *Th. elongatum* genome (table S1). We initially assembled the short sequence reads using DeNovoMAGICTM3.0 software (NRGene) and then filled the gaps using ~145 Gb (~31×) PacBio SMRT reads. The initial assembly was finely tuned using high-quality paired-end polymerase chain reaction (PCR)–free reads. Two Bionano optical maps (based on enzymes BspQI and DLE1 data) were further used to extend the scaffolds (tables S2 and S3), which resulted in a 4.63-Gb assembly with a contig N50 size of 2.15 Mb and a scaffold N50 size of 73.24 Mb (Table 1).

To construct the pseudochromosomes, high-throughput chromosome conformation capture (Hi-C) data were used to categorize and order the assembled scaffolds (table S4). A total of 141 scaffolds were anchored and oriented onto seven pseudochromosomes, which account for 95% of the estimated genome size (4.78 Gb; fig. S1) and 98% of the assembled genome sequences (fig. S2). About 97.6% complete and 1.3% fragmented Embryophyta genes were detected in our assembly according to BUSCO [Benchmarking Universal Single-Copy Orthologs (*17*)], proportions comparable

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Table 1. Summary statistics for *Th. elongatum* genome assembly.

Assembly characteristics	Values
Estimated genome size	**4.78 Gb**
Total length of contigs	4.58 Gb
N50 length of contigs	2.15 Mb
Total number of contigs	12,262
Longest contigs	11.6 Mb
Total length of scaffolds	4.63 Gb
N50 length of scaffolds	73.24 Mb
Total number of scaffolds	783
Longest scaffolds	258.71 Mb
Total gap size	52.78 Mb
Total sequences anchored to the pseudochromosomes	4.54 Gb
Number of annotated high-confidence genes	44,474
Percentage of repeat sequences	81.29%
Complete BUSCOs	97.6%
Fragmented BUSCOs	1.3%
Missed BUSCOs	1.1%

to other *Triticum* genomes (table S5). The quality of the E genome assembly was validated by assessment of the long terminal repeat (LTR) completeness using LTR Assembly Index (LAI) software (*18*) (table S6), by genomic alignment with 61 randomly selected bacterial artificial chromosome (BAC) clones (fig. S3 and table S7), and by the consistency of our assembly with a high-density genetic map from a hexaploid *Thinopyrum* species (*19*) (fig. S4).

Repetitive elements are dispersed throughout the E genome, with ~81.29% of the *Th. elongatum* assembly being annotated as repetitive elements, including retrotransposons (62.39%), DNA transposons (17.83%), and unclassified elements (1.07%) (table S8 and table S9). Analysis of the Cereba and Quinta LTR retrotransposons supported that the centromere regions were appropriately assembled (fig. S5). The composition of different classes of repetitive DNA in the E genome was similar to those of the wheat A, B, or D subgenomes (fig. S6). No recent LTR burst was detected in the E or common wheat genomes (fig. S7), suggesting relatively stable genomes and helping to explain the success of distant hybridization breeding efforts using these materials. A total of 44,474 high-confidence protein-coding genes were predicted on the basis of a combination of methods [ab initio, protein homology based, and RNA-sequencing (RNA-seq) based], and 44,144 (99.3%) of the predicted genes were anchored onto the seven assembled pseudochromosomes (figs. S8 and S9 and tables S10 to S12).

Gene family analysis identified 32,048 orthologous genes between the E genome and the wheat A, B, or D genome or the barley genome (fig. S10). A synonymous substitution rate (K_s) value was calculated using a moving-average model with the ortholog dataset, which revealed similar K_s peak values between the E genome and the wheat subgenomes (E and A: 0.0645, E and B: 0.0645, E and D: 0.062), indicating a branching time for *Th. elongatum* and *Triticum* of ~4.77 to 4.96 million years ago when a nucleotide substitution rate of $6.5*10^{-9}$ was used (Fig. 1A) (*20*).

We also compared the E genome with other Triticeae genomes that have been used for distant hybridization based on a maximum likelihood tree built using single-copy genes from available Triticeae genome assemblies; the tree also incorporated transcript data for several diploid species, including the Triticeae R, Q, V, F, and Ns genomes (table S13). The three wheat subgenomes are more closely related to the E genome of *Th. elongatum* than they are to the R genome of rye, another species frequently used in wheat distant hybridization (Fig. 1A). A syntenic block analysis indicated genome-wide colinearity between the E genome and the A, B, or D genomes, which helps to explain the success of E-genome–based distant hybridization breeding in wheat (Fig. 1B and data S1). Substantial colinearity notwithstanding, we did identify 18 fragmental inversions between the E genome and the wheat subgenomes, with sizes ranging from 1.5 to 18 Mb, which is supported by both the Bionano maps and Hi-C data (fig. S11 and table S14).

Map-based cloning of the *Fusarium* resistance gene *Fhb7*

A total of 1897 resistance gene analogs (RGA) were annotated in the E genome (fig. S12 and table S15). An apparent RGA expansion, especially for CC-NBS-LRR (CNL), on the distal end of the long arm of chromosome 7E (7EL) is accompanied with the expansion of this genomic region (fig. S13 and table S16). Some of the alien resistance gene introgressions into wheat are located in this region, including *Lr19*, *Sr25*, *Bdv3*, and *Fhb7* (*10*, *13*, *14*).

Previously, we mapped the *Fhb7* to the distal end of the 7EL (based on recombination between 7E1 and 7E2 in a common wheat background) using a recombinant inbred line (RIL) population from a cross between an FHB-susceptible substitution line (7E1/7D) and an FHB-resistant substitution line (7E2/7D) (*13*, *21*). For further mapping of this gene, we developed a segregation population derived from BC_6F_1 with the same cross, in which FHB resistance was tracked as monogenic trait for validation of phenotypes. We also developed a population to promote 7E recombination by introducing the CS *ph1bph1b* locus (fig. S14). Because of the semidominant nature of *Fhb7*, the homozygous offspring of the recombinants were evaluated for FHB resistance. With analysis of 258 recombinants (between the *XBE45653* and *XsdauK67* markers) screened from 19,200 progeny of BC_6F_1 population, we confirmed that *Fhb7* is positioned between the *XSdauK79* and *XSdauK80* markers within an ~1.2-Mb region based on the E reference genome (Fig. 1C and fig. S15).

Analysis of the RNA-seq data of E reference genome from *Th. elongatum* spikes identified eight expressed genes in the *Fhb7* region (Fig. 1C and table S17). However, when conducting transcriptomics analysis of the parental lines of 7E1/7D (S) and 7E2/7D (R), we found that only two candidate genes (Tel7E01T1020600.1 and Tel7E01T1021800.1) were expressed in a manner specific to the 7E2 genome (the resistant parent) and E reference genome [which also confers FHB resistance (*12*, *22*)] (Fig. 1C and tables S18 and S19). BAC clones containing Tel7E01T1020600.1 were identified from the resistant donor line and new markers (*XsdauK86* and *XsdauK87*) derived from the BAC ends were developed to screen recombinants among 5760 progeny of the segregation population harboring the CS *ph1bph1b* locus (Fig. 1C, fig. S14, and table S20). Analysis of phenotypic data of the three key recombinants verified that *Fhb7* is located between the *XsdauK86* and *XsdauK88* markers, thereby delineating this locus to a 245-kb region containing a single expressed gene: Tel7E01T1020600.1 (Fig. 1C). This gene is present in the E reference genome and 7E2 genome but absent in the susceptible 7E1 genome based on analysis of genomics and transcriptomics data (table S19 and table S21).

Gene expression analysis using quantitative PCR indicated that Tel7E01T1020600.1 was constitutively expressed in all tissues examined, including root, leaf, shoot, and spike (fig. S16). Moreover, barley stripe mosaic virus (BSMV)–induced gene silencing of Tel7E01T1020600.1 in wheat leaves revealed that it conferred

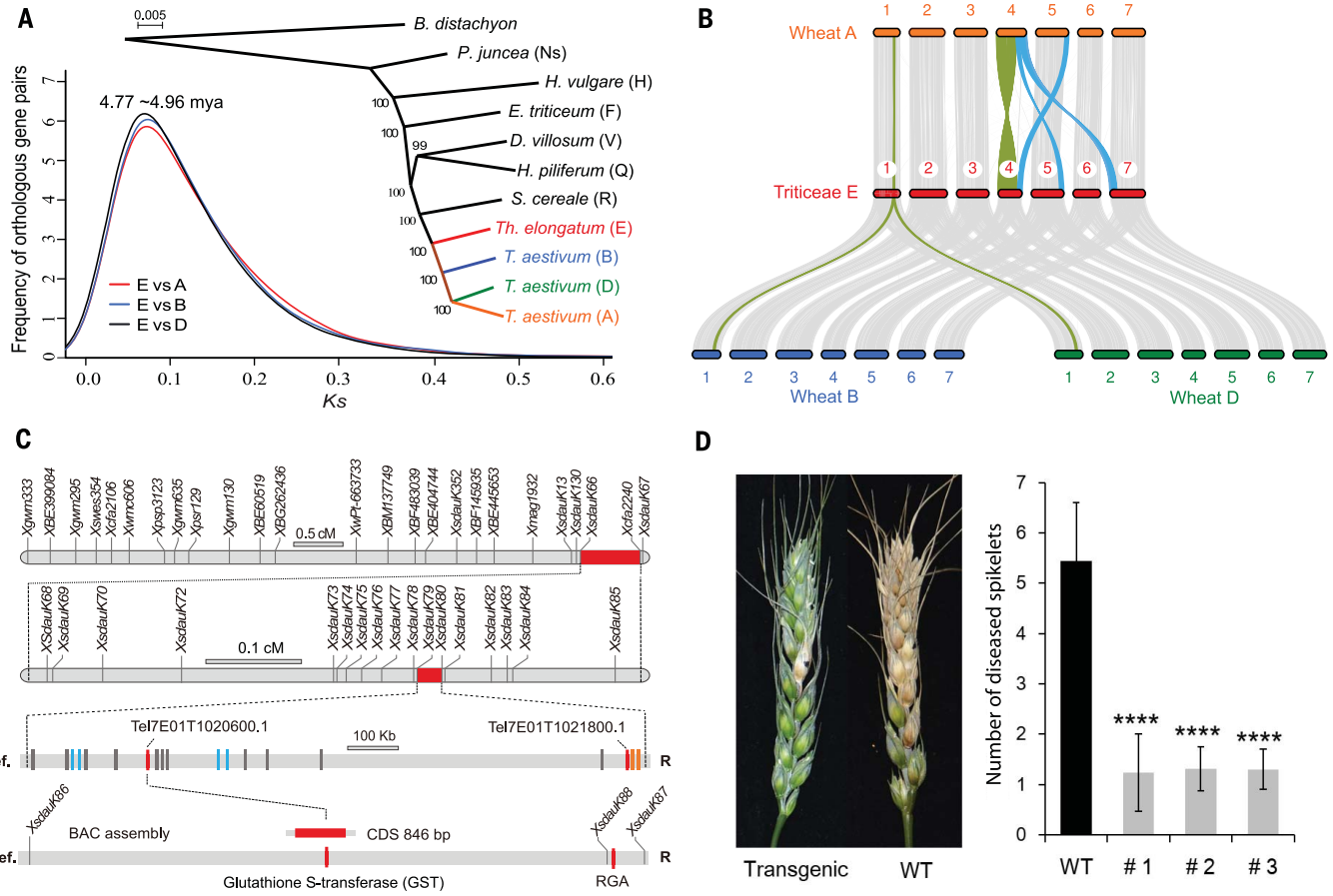

Fig. 1. Genome evolution of *Th. elongatum* and cloning of *Fhb7*. (**A**) Maximum likelihood phylogenetic tree of the genomes of Triticeae species and the Ks distributions of ortholog genes between the E genome and the wheat Chinese Spring A, B, and D subgenomes. mya, million years ago. (**B**) Syntenic blocks between the E genome and the three wheat subgenomes. The representative inversion fragment is indicated in green; chromosomal translocations for the wheat A subgenome compared with the E genome are also indicated in blue. (**C**) Map-based cloning of *Fhb7* at the distal region of chromosome 7E. Using the BC_6F_1 population derived from the cross between two wheat-*Thinopyrum* substitution lines, 7E1/7D and 7E2/7D, *Fhb7* was initially mapped to an interval between the markers *XsdauK79* and *XsdauK80* (~1.2 Mb on the E reference genome) (second bar from the top). The expressed genes are labeled as follows: gray refers to no expression in the E reference genome; blue refers to E reference genome–specific expression; orange refers to expression in the E reference, 7E1 and 7E2 genomes; red refers to expression in FHB-resistant donor genomes of 7E2 and E reference (third bar from the top). BAC clones containing Tel7E01T1020600.1 were identified from the substitution line 7E2/7D, based on which genetic markers (XsdauK86 and XsdauK87) were developed for recombinant screening of the CS *ph1bph1b* population. Finally, *Fhb7* was genetically confirmed within a 245-kb region between markers *XsdauK86* and *XsdauK88*, with only the candidate gene Tel7E01T1020600.1 encoding a GST [CDS is shown in red; untranslated region is shown in gray] (fourth bar from the top). (**D**) FHB was evaluated for wild-type (WT, KN199) and transgenic wheat KN199 expressing the native promoter and the 846-bp open reading frame of *Fhb7*. T_3 plants containing *Fhb7* from three different lines were evaluated for FHB resistance using single floret inoculation (*35*). The FHB was scored for at least five spikes per repeat, with at least three repeats for each transgenic line.

resistance to *F. graminearum*, supporting that this gene represents *Fhb7* (fig. S17). Sequence analysis of 22 ethyl methanesulfonate (EMS)–induced mutants identified five amino acids that were implicated in *Fhb7*'s FHB resistance–related function: S34F, T48I, A98V, A9V, and P106L (fig. S18 and data S2). Moreover, two stop-gain mutations at position 209 or 243 led to reduced resistance to *F. graminearum* (fig. S18 and data S2). To confirm Tel7E01T1020600.1 as *Fhb7*, we transgenically introduced a construct with the native promoter and the 846–base pair (bp) coding sequence of this gene into the FHB-disease–susceptible wheat cultivar KN199 and assessed three independent T_3-transgenic plants. The *Fusarium*-inoculated transgenic plants exhibited lower FHB symptom with substantially fewer diseased spikelets per spike than the control (Fig. 1D).

Evolutionary history and molecular function of *Fhb7*

Protein domain–based functional annotation predicted that *Fhb7* likely encodes a GST enzyme. A BLAST search of the *Fhb7* sequence against the National Center for Biotechnology Information (NCBI) GenBank database (*23*) did not find any homolog of *Fhb7* in the *Triticum* genus or in the entire plant kingdom. However, there is a homolog sharing 97% identity in the genome of *E. aotearoae* (Fig. 2A and fig. S19). A phylogenetic analysis of the *Fhb7* sequence revealed its distribution among *Epichloë* species, endophytic fungi of temperate grasses (Fig. 2A). Thus, the occurrence of the *Fhb7* gene in the *Th. elongatum* genome might be caused by fungus-to-plant HGT (FP-HGT) event. Because the *Fhb7* locus is present both in the diploid E genome of *Th. elongatum* and in 7E2 from decaploid *Th. ponticum*, this FP-HGT event apparently occurred after the divergence of the E genome from *Triticum* sp. but before the formation of the decaploid *Th. ponticum* (Fig. 2A).

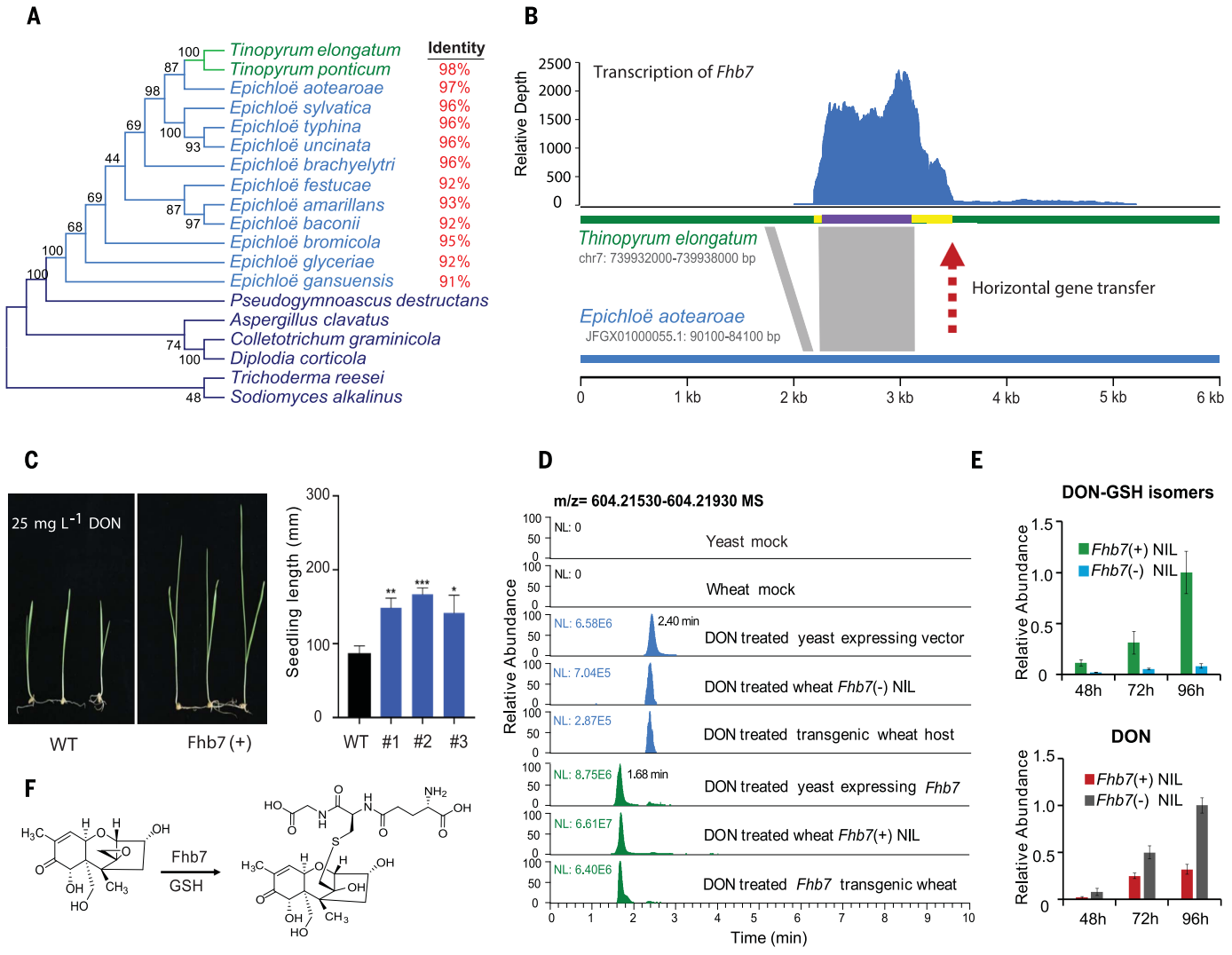

Fig. 2. *Fhb7* confers FHB resistance by detoxifying DON. (**A**) Maximum likelihood phylogenetic tree of the closest homologs of *Fhb7* from plants and fungi. The DNA sequence similarity with *Fhb7* is marked in red. (**B**) Horizontal gene transfer of *Fhb7*. The transcripts CDS (purple), and possible untranslated regions (yellow) of *Fhb7* are shown along chromosome 7E, and the sequence sharing high similarity with the *E. aotearoae* genome is presented as a gray block. The genomic fragment (897 bp) containing full CDS and partial untranslated region of *Fhb7* showed 97% identity between the two genomes. (**C**) DON tolerance of *Fhb7*-transgenic wheat. Seedlings (4 days old) were moved to a petri dish containing 25 mg L^{-1} DON and seedling length was evaluated 7 d after the DON treatment at room temperature. (**D**) Extracted ion chromatograms (EICs) at *m/z* 604.2173 revealing the presence of two DON-glutathione adducts. The *Fhb7* NIL, *Fhb7*-transgenic wheat, and *Fhb7*-transgenic yeast (*P. pastoris*) cultures were treated with 25 mg L^{-1} DON for 24 hours. A product that elutes at 1.68 min accumulated in Fhb7(+) samples and a known, nonenzymatically produced DON-glutathione adduct product that elutes at 2.4 min accumulated in the corresponding *Fhb7*(−) control samples. (**E**) Relative abundances of the de-epoxidated Fhb7-catalyzed DON-glutathione (green) adduct and the known nonenzymatic DON-glutathione adduct (blue) in spikes of *Fusarium*-challenged NIL plants contrasting in *Fhb7*. After inoculation of *F. graminearum* on spike glumes, the *Fhb7*(+) NIL accumulated a copious amount of de-epoxidated DON-glutathione adduct. By contrast, the DON substrate reduced the accumulation in *Fhb7*(+) NIL compared with that in *Fhb7*(−) NIL, as shown in the bottom bar chart. (**F**) Molecular structure of the de-epoxidated DON-glutathione adduct catalyzed by Fhb7.

The horizontal transfer of the *Fhb7* sequence did not occur as a part of a gene cluster (presuming that it is from *E. aotearoae* as the donor genome; this is the species harboring the closest identified homolog of *Fhb7*) (fig. S20). On the basis of sequence similarity, the sequence was transferred into the diploid E genome as a short fragment, including the 846-bp coding sequence for *Fhb7*, a 32-bp sequence before the start codon, and a 19-bp sequence after the stop codon (Fig. 2B). At the position 535 bp upstream of *Fhb7*'s start codon in the E genome, another 90-bp sequence shows high identity to a sequence in *E. aotearoae* (Fig. 2B), suggesting the possibility that a larger sequence was initially transferred to *Th. elongatum* but late mutations occurred in the transferred sequence. The insertion of the *Epichloë* genome fragment in the E genome was also identified in a BAC clone harboring *Fhb7* (Fig. 1C and data S3), confirming that the sequence is not an artifact from the genome assembly process.

Phylogenetic analysis of the GST superfamily showed that *Fhb7* belongs to the fungal GTE (glutathione transferase etherase–related) subfamily (fig. S21 and tables S22 and S23), wherein all members contain a LigE domain, but none of which has been functionally characterized to date (*24*). The *Fhb7* gene is conserved in *Epichloë* species and in multiple *Thinopyrum* species, emphasizing its role in protecting organisms from the cytotoxic damage caused by *Fusarium* species (Fig. 2A and fig. S20).

Gene expression analysis in a time course of *Fusarium* infection in *Th. elongatum* and the 7E2/7D substitution line (table S18) showed that the transcription levels of *Fhb7* were induced at 48 hours after infection (fig. S22).

Research in plant pathology about the progression of *F. graminearum* infection in wheat has established that the fungus starts to produce its DON mycotoxin, an inhibitor of protein synthesis that targets ribosomal machinery, by the 48-hour infection time point (*25*). We therefore conducted DON assays on wheat seedlings of the 7E2/7D substitution line. The results showed that the expression of *Fhb7* can be induced within 6 hours after DON treatment (fig. S22), suggesting that this putative GST enzyme may have a role in xenobiotic detoxification. To test this hypothesis, we conducted a growth inhibition assay by growing *Fhb7* near-isogenic lines (NILs) and *Fhb7*-transgenic wheat seedlings in media containing DON and found that the plants with *Fhb7* grew better (assessed as seedling length) than the plants without *Fhb7* (Fig. 2C and fig. S23). We also expressed *Fhb7* in yeast to test its growth on DON-containing media and found that both the *Fhb7*(+) and *Fhb7*(−) yeasts grew well in the absence of DON; however, only the *Fhb7*(+) yeast grew normally on the media containing 400 mg L^{-1} DON (fig. S24).

Further evidence for the involvement of *Fhb7* in detoxification was demonstrated by its direct use of DON as a substrate. We treated the seedlings of NILs, *Fhb7*-transgenic wheat, and *Fhb7*-expressing yeast cultures with DON, and found that the presence of *Fhb7* in wheat and yeast caused accumulation of a chromatographic peak at 1.68 min, but the accumulation was not detected in the corresponding control samples without *Fhb7* (Fig. 2D). This peak had a mass/charge (m/z) value of 604.2173 (± 3 ppm) under positive ion mode, which is equal to the value for the molecule comprising DON (296.1259), a glutathione group (307.0838), and a hydrogen atom (1.0078), therefore suggesting that *Fhb7* confers GST activity to form a glutathione adduct of DON (DON-GSH) (Fig. 2D and fig. S25).

Previous studies on FHB- and DON-associated chemistry (*26*, *27*) using nuclear magnetic resonance spectroscopy confirmed the nonenzymatic formation of a DON-GSH adduct that was formed through a reaction with the double bond at C10 on DON's first planar ring. This product was mainly detected in the DON-treated *Fhb7*(−) yeast cultures and *Fhb7*(−) wheat samples with the peak at 2.4 min (Fig. 2D and fig. S25). Although the two detected DON-GSH isomers had identical m/z values, tandem mass spectrometry with collision-induced dissociation experiments unequivocally supported that the *Fhb7*(+) samples produce a de-epoxidated DON-GSH adduct (figs. S25 to S28); that is, the GSH group added by Fhb7 is attached to the C13 carbon, which disrupts the epoxy group known to be critical in DON's toxicity (Fig. 2F) (*28*). Further, we used liquid chromatography–high-resolution mass spectrometry (LC-HRMS) to profile DON-treated spikes from 37 diverse wheat germplasm accessions and cultivars without Fhb7. We detected the DON-GSH (C10) peak at 2.4 min in all of these plants but did not detect the 1.68-min de-epoxidated DON-GSH (C13) adduct in any of them (fig. S29).

Fusarium species produce a series of trichothecene mycotoxins, including DON, 3-ADON, 15-ADON, T-2, HT-2, fusarenon-X, NIV, diacetoxyscirpenol, and others, the distribution of which varies among *Fusarium* chemotypes (*24*, *26*). Considering the common occurrence of epoxy groups in these trichothecene compounds, we hypothesized that Fhb7 may be able to detoxify trichothecenes other than DON. Indeed, LC-HRMS analysis of trichothecene-treated wheat samples revealed the presence of GSH adducts for all the trichothecenes that we tested in this study (figs. S30 to S37). In light of Fhb7's wide catalytic spectrum for these mycotoxins, we investigated whether it can confer resistance to other *Fusarium* chemotypes, including *F. pseudograminearum* for crown rot and *F. asiaticum*, a predominant FHB-causing strain in south China. Assays using detached wheat leaves showed that the *Fhb7*-transgenic plants exhibited smaller lesions than wild-type plants for all the tested *Fusarium* species (fig. S38). *F. pseudograminearum* was also inoculated on the base of wheat seedlings, and the results confirmed that the transgenic plants also exhibit improved crown rot resistance compared with the nontransgenic controls (fig. S39). These results further demonstrate how *Th. elongatum* benefits from *Fhb7* through the FP-HGT event, which protects plants from *Fusarium*-caused cytotoxic damage by detoxifying trichothecene through de-epoxidation (fig. S20).

Application of *Fhb7* in *Fusarium* resistance breeding

Considering *Fhb7*'s functionality, specifically in the enzymatic conversion of trichothecenes, we speculated that incorporating the *Fhb7* locus into wheat may confer resistance in different genetic backgrounds without affecting yield traits. Indeed, the translocation of a short fragment [with ~16% of the 7E long arm (*13*)] on wheat 7D resulted in wheat lines with broad resistance to both FHB and crown rot (Fig. 3, A to C). Detailed characterization of NILs (LX99 background) in field conditions showed no significant difference in agronomic yield traits (e.g., thousand grain weight, flag leaf length, etc.; Fig. 3, D and E). Obvious yield penalty caused by *Fhb7* resistance was also not detected when it was transferred into seven additional genetic backgrounds (Fig. 3F and fig. S40). These results demonstrated the advantages of *Fhb7*-mediated resistance over other QTLs, including high resistance to both FHB and crown rot and detoxifying DON without yield penalty, and thus highlighted the potential utility of the *Fhb7* locus in future wheat breeding for improved FHB resistance and good yield traits.

Discussion

Fusarium diseases are economically impactful because of their effects on the production of cereal crops. In this study, the successful cloning of *Fhb7* from the Triticeae E genome and characterization of its molecular mechanism advances the knowledge on the essential role of trichothecenes in the pathogenesis of *Fusarium*. We have demonstrated that *Fhb7* confers FHB resistance in diverse wheat genetic backgrounds without yield penalty and Fhb7 is able to biochemically detoxify trichothecene mycotoxins produced by multiple *Fusarium* species, which highlights the value of *Fhb7* in combating FHB and reducing DON contamination in wheat and other cereal crops through breeding.

The epoxides at the C12/13 of trichothecene mycotoxins are the key contributors to their toxicity. However, to date, genes or proteins with de-epoxidation function have not been identified (*3*). *Fusarium* species can reduce DON toxicity by adding an acetyl group on the hydroxyl group at C3 and C15, forming 3-ADON and 15-ADON, respectively; however, the reduction of cytotoxicity for these DON derivatives is modest in plant cells (*3*). In planta, glucosylation at C3 has been documented to detoxify DON by forming DON-3-glucoside (D3G), which is reversible in animals, causing release of DON during digestion (*29*). Here, beyond our identification of an FHB resistance gene, the broad detoxification spectrum of Fhb7 through de-epoxidation of trichothecenes suggests the potential utility of the GST enzyme in the biomedicine, feed, and food industries in addition to reducing DON content in wheat grain.

HGT, the transfer of genes between nonmating species, is thought to occur frequently in prokaryotes, but much less so in eukaryotes (*30*). There is accumulating evidence illustrating instances of HGT events involving bacteria or the organellar genomes of another plant as donor (*31*). For instance, two *Agrobacterium* genes were found to be inserted in the genome (with transfer DNA borders) of a cultivated sweet potato [*Ipomoea batatas* (L.) Lam.], revealing a naturally occurring transgenic food crop (*32*). However, there is little evidence for HGT events involving nuclear DNA transmission from fungi or other eukaryotes, and such transmission has been thought to be insignificant (*33*). Fundamentally, our results highlight the roles that FP-HGT has had in shaping plant genomes, which advances the

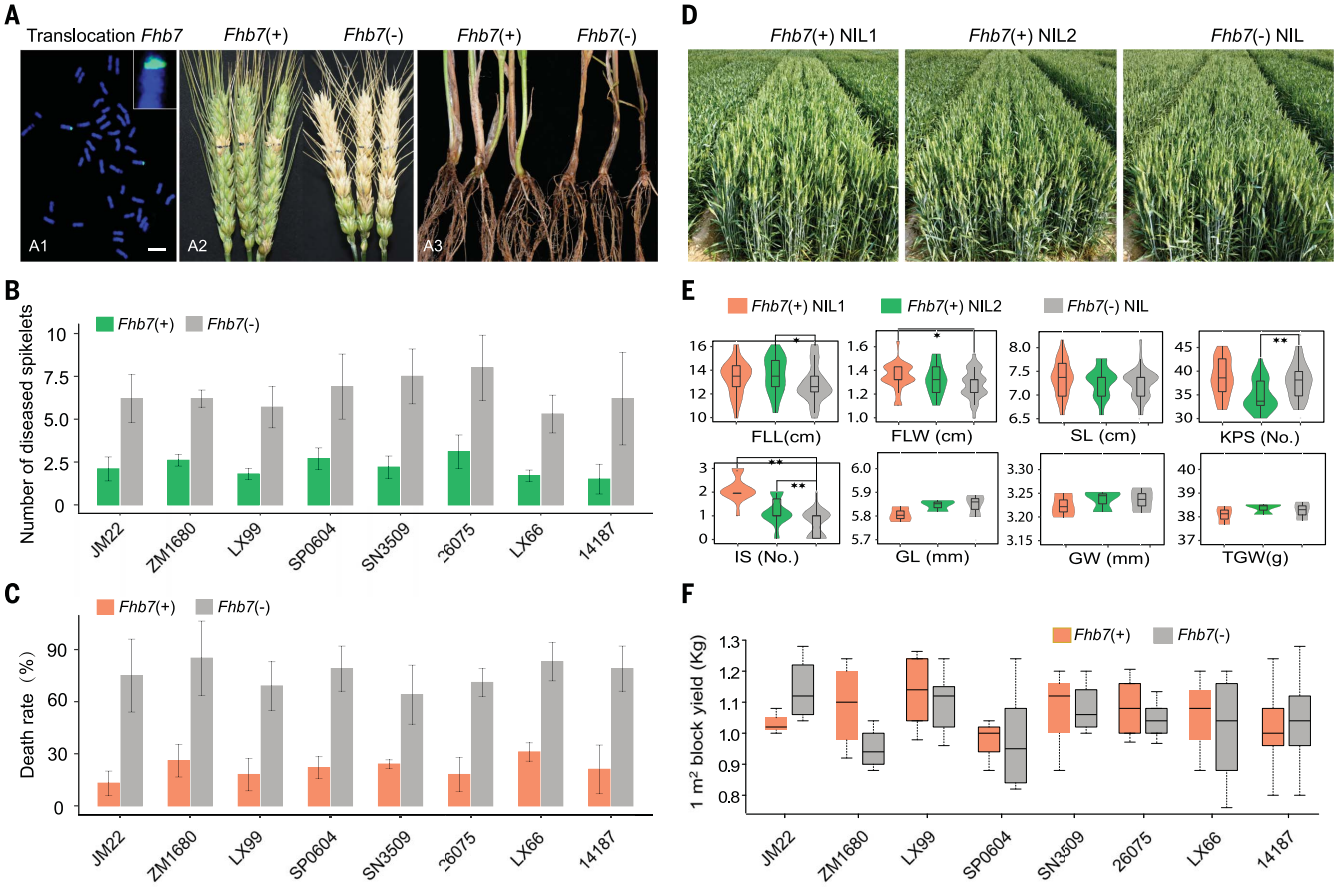

Fig. 3. Application prospects for *Fhb7* in wheat resistance breeding. (**A**) Genomic in situ hybridization analysis (left panel) showing a translocation of the distal region of 7E (containing *Fhb7*) from an E genome donor into wheat. Scale bar, 20 μm. Also shown are images of *Fusarium*-infected spikes (middle panel) and crown rot (right panel) of LX99 NILs contrasting in *Fhb7*. (**B**) FHB resistance of *Fhb7* in eight different wheat genetic backgrounds evaluated at 21 d after inoculation in field conditions. (**C**) Crown rot phenotypes were recorded as the death ratio after growth in soil containing *F. pseudograminearum* at 30 days postinfection. (**D**) Field plant photographs of two *Fhb7*(+) NILs and one *Fhb7*(−) NIL in the LX99 background. (**E**) Comparison of the yield traits among the two *Fhb7*(+) NILs and one *Fhb7*(−) NIL in the LX99 background evaluated in the 2017 field experiment. FLL, flag leaf length (cm); FLW, flag leaf width (cm); SL, spike length (cm); KPS, kernels per spike; IS, infertile spikelets; GL, grain length (mm); GW, grain width (mm); TGW, thousand grain weight (g). (**F**) Comparison of the grain yield among eight *Fhb7* translocation lines in different wheat genetic backgrounds. The grain yield was measured from a 1-m^2 plot in the 2017 and 2018 field experiments.

knowledge on disease resistance gene evolution and opens a new avenue for the identification of plant resistance genes.

The endophytic *Neotyphodium* and *Epichloë* fungi often form mutualistic symbiotic associations with forage grasses and offer hosts bioprotective benefits against pathogens and abiotic stresses, presumably owing to the fungus-mediated anabolism and catabolism of various natural product compounds (*34*). Here, we showed that the GST encoded by *Fhb7* is conserved in *Epichloë* species and is able to detoxify the trichothecene mycotoxins secreted by *Fusarium* species. Thus, transfer of this fungal gene into a plant genome could be beneficial to plants, perhaps even eliminating the need for the symbiotic association per se. The finding of *Fhb7*-mediated resistance to both FHB and crown rot diseases further emphasizes the importance of this HGT in benefiting the perennial *Th. elongatum*, which is perhaps reflected by constitutive expression of *Fhb7* in all examined tissues. However, the molecular machinery that enabled the FP-HGT of *Fhb7* and the nature of the promoter evolution underlying the expression of *Fhb7* remain to be elucidated.

Methods summary

The *Th. elongatum* genome was first sequenced by Illumina short-read sequencing and was de novo assembled using the software package DeNovoMAGICTM3.0. PacBio SMRT long reads were used to fill the gaps in the assembly and Bionano optical maps were then used to correct and extend the scaffold sequences. The assembly was anchored into seven pseudochromosomes using Hi-C data. The assembly was validated using independent BAC sequences, genetic maps of related species, and commonly used software programs. Genes, repetitive DNA, and other genomic features in the assembly were annotated to reveal the landscape of the species and to examine their relationship with wheat and other related species by in-depth comparative analyses. Genetic markers in the *Fhb7* region were developed by means of the reference genome sequence and used to screen recombinants for fine mapping to identify the *Fhb7* candidate gene. The candidate gene was functionally validated by virus-induced gene silencing, EMS-induced mutation, and transgenic approaches. FHB resistance was evaluated by inoculation of *Fusarium* conidial suspensions on wheat spikes, leaves, or crowns. LC-HRMS(/MS) analysis was used to infer the biochemical structure of trichothecene-glutathione adducts catalyzed by Fhb7. *Fhb7* was introgressed into diverse wheat backgrounds using distant hybridization and conventional breeding, and the presence of alien chromatin in wheat was validated by genomic in situ hybridization.

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SUPPLEMENTARY MATERIALS

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Transcriptomic changes in *Nicotiana benthamiana* plants inoculated with the wild-type or an attenuated mutant of *Tobacco vein banding mosaic virus*

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SUMMARY

Tobacco vein banding mosaic virus (TVBMV) is a potyvirus which mainly infects solanaceous crops. The helper component proteinase (HCpro) of a potyvirus is an RNA silencing suppressor protein and determines the severity of disease symptoms caused by different potyviruses, including TVBMV. It has been shown that substitution mutations introduced into the HCpro open reading frame (ORF) in a TVBMV infectious clone result in changes of Asp_{189} to Lys or Ile_{250}-Gln_{251} to Asp-Glu (Asp, aspartic acid; Gln, glutamine; Glu, glutamic acid; Ile, isoleucine). These amino acid changes eliminate the RNA silencing suppression activity of the mutant HCpro (HCm) and attenuate the disease symptoms caused by the mutant TVBMV (T-HCm) in *Nicotiana benthamiana* plants. Here, we used RNA-sequencing technology to compare gene expression in plants inoculated with the wild-type TVBMV (T-WT) with that in plants inoculated with T-HCm at 1, 2 and 10 days post-agroinfiltration (dpai). At 1 and 2 dpai, *N. benthamiana* genes related to the translation machinery were up-regulated, whereas genes related to lipid biosynthesis and metabolism or to responses to extracellular/external stimuli were down-regulated in leaves inoculated with T-WT or T-HCm. At 10 dpai, T-WT infection repressed photosynthesis-related genes. T-WT and T-HCm infections differentially perturbed the genes involved in the RNA silencing pathway. The salicylic acid and ethylene signalling pathways were induced, but the jasmonic acid signalling pathway was repressed after T-WT infection. Infections of T-WT and T-HCm also differentially regulated the genes involved in auxin signalling transduction, which is known to associate with the stunting phenotypes caused by TVBMV. These results illustrate the dynamic nature of TVBMV infection in *N. benthamiana* at the transcriptomic level.

Keywords: attenuated mutant virus, *Potyvirus*, RNA-seq, *Tobacco vein banding mosaic virus*, transcriptomics.

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INTRODUCTION

Viruses in the genus *Potyvirus* (family *Potyviridae*) account for approximately 30% of the known plant viruses, and cause great economic losses to agricultural production worldwide (Fauquet *et al.*, 2005). The potyvirus genome is a positive-sense, single-stranded RNA of approximately 10 kb in length. It contains two open reading frames (ORFs) that encode a large and a shorter polyprotein, respectively. The two polyproteins are then processed into 11 mature proteins by three viral-encoded proteinases (Chung *et al.*, 2008; Fauquet *et al.*, 2005; Geng *et al.*, 2015; Olspert *et al.*, 2015; Riechmann *et al.*, 1992; Vijayapalani *et al.*, 2012). From the N-terminus of the polyprotein, the 11 mature proteins are designated as the P1, helper component proteinase (HCpro), P3, P3N-PIPO, 6K1, cylindrical inclusion (CI), 6K2, viral genome-linked protein (VPg) of the nuclear inclusion protein a (NIa), proteinase domain of the NIa (NIa-Pro), nuclear inclusion protein b (NIb) and coat protein (CP), respectively.

RNA silencing is an adaptive antiviral defence process (Ding, 2010; Ding and Voinnet, 2007). To counteract this antiviral defence, plant viruses have evolved to produce various suppressors of RNA silencing (VRSs) (Burgyán and Havelda, 2011). Potyvirus-encoded HCpro is a well-studied VRS and is a multifunctional protein necessary for aphid transmission, virus movement, replication, symptom development and suppression of RNA silencing (Díaz-Pendón and Ding, 2008; Hasiów-Jaroszewska *et al.*, 2014; Maia *et al.*, 1996). Specific mutations in VRSs can attenuate the symptoms induced by plant viruses. For example, a change in cysteine (Cys) to tyrosine (Tyr) at the amino acid position 348 in the *Tobacco mosaic virus* 130-kDa protein results in an attenuated phenotype in infected plants and a reduction in RNA silencing suppression activity of this protein (Nishiguchi and Kobayashi, 2011). Similarly, substitutions of amino acids in the 2b protein of *Cucumber mosaic virus* (Dong *et al*, 2016; Nishiguchi and Kobayashi, 2011) or in the HCpro of *Clover yellow vein virus* or *Zucchini yellow mosaic virus* also result in symptom attenuations and reduction in RNA silencing suppression activities (Lin *et al.*, 2007; Shiboleth *et al.*, 2007). Compared with our knowledge of the attenuated viral pathogenicity and RNA silencing suppression

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Full Length Article

Isolation and Identification of Four Novel Biocontrol *Bacillus* Strains against Wheat Sharp Eyespot and their Growth-Promoting Effect on Wheat Seedling

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Abstract

Natural resistance of wheat plant to sharp eyespot disease is inadequate and new strategies are urgent desired to control this serious soil-borne disease. Biological control is an alternative and attractive approach to effectively reduce the utilization of chemicals in agriculture. In this study, four biocontrol bacterial strains exhibited strong antagonistic activities against *Rhizoctonia cerealis* were isolated from rhizosphere soil of infected wheat *via* a dual culture method. The strains of TA28, TA31, Z-5 and Z-7 were identified as *Bacillus methylotrophicus*, *B. subtilis*, *B. amyloliquefaciens* and *B. tequilensis*, respectively, according to morphological, physiological and biochemical characterizations and 16S rRNA gene sequencing. The feasible mechanism of impeding mycelial growth for these *Bacillus* strains was irregularly swollen-tipped and increasing compartment in hypha. The antifungal spectrum indicated that these four strains conferred significant antagonistic effects against many soil-borne pathogenic fungi, including *Fusarium oxysporum*, *Pythium aphanidermatum* and *Phytophthora sojae*. In addition, the bacterium suspension of each strain could actively promote wheat seedling growth. Therefore, the present work provides some new insights on potential candidates for biological control of wheat sharp eyespot and other common soil-borne plant diseases. © 2019 Friends Science Publishers

Keyword: Antagonistic experiment; *Bacillus* spp.; Biological control; Wheat sharp eyespot

Introduction

Wheat sharp eyespot, caused by *Rhizoctonia cerealis* anastomosis group D subgroup I (AG-DI), is a serious wheat stem-base disease in temperate regions worldwide (Li *et al.*, 2017). In infection process, the pathogenic fungus destructs stem and sheath of wheat plant, triggering to block the transportation of nutrients required for the wheat growth (Zhu *et al.*, 2015). Actually, sharp eyespot has been considered as one of the most economically important wheat diseases in China and exceeded approximately an annual loss of million ha in wheat production (Rong *et al.*, 2016).

The main control strategy against *R. cerealis*, in general, is fungicide seed coating and spraying at early jointing stage (Chen *et al.*, 2013). Unfortunately, the undesirable environmental and ecological consequences of increasing fungicide residues have seriously challenged the utilization of chemical agents in agriculture (Talhinhas *et al.*, 2018). Moreover, although cultivars resistant to sharp eyespot can be used to prevent this disease, few wheat cultivars possess moderate resistance and highly resistant cultivars are rare (Wu *et al.*, 2017). Therefore, an environmental-friendly approach has been desired to efficiently control this wildscale soil-borne disease.

Recently, more attention has been drawn to the biological control, which plays an important role in reducing potential environmental and health risks of chemical fungicides and provides an option for combating with soil-borne disease pathogens in crop (Postma *et al.*, 2003; Compant *et al.*, 2005). Thus, it is essential to explore biocontrol agents with high efficiency. Beneficial bacteria are capable of antagonizing pathogens by producing extracellular antifungal compounds and indirectly stimulating the self-defense system of host plants (Beneduzi *et al.*, 2012; Guardado-Valdivia *et al.*, 2018). The *Bacillus* species serve as one of the most realized biocontrol agents on account of a series of connected modes of action and formation of compression resistance with their superior survival ability at different environmental conditions (Setlow, 2014). This favorable feature contributing to antifungal activity is the secretion of active secondary metabolites, including low molecular weight volatile compounds and antibiotics (Manns *et al.*, 2012; Wang *et al.*, 2014a; Wu *et al.*, 2015). Additionally, as an important genus, *Bacillus* strains are able to produce multiple

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ORIGINAL PAPER

Insights into the Synergistic Biodegradation of Waste Papers Using a Combination of Thermostable Endoglucanase and Cellobiohydrolase from *Chaetomium thermophilum*

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Abstract
Enzymatic hydrolysis is considered an efficient and environmental strategy for the degradation of organic waste materials. Compared to mesophilic cellulases, thermostable cellulases with considerable activity are more advantageous in waste paper hydrolysis, particularly in terms of their participation in synergistic action. In this study, the synergistic effect of two different types of thermostable *Chaetomium thermophilum* cellulases, the endoglucanase CTendo45 and the cellobiohydrolase CtCel6, on five common kinds of waste papers was investigated. CtCel6 significantly enhanced the bioconversion process, and CTendo45 synergistically increased the degradation, with a maximum degree of synergistic effect of 1.67 when the mass ratio of CTendo45/CtCel6 was 5:3. The synergistic degradation products of each paper material were also determined. Additionally, the activities of CTendo45 and CtCel6 were found to be insensitive to various metals at 2 mM and 10 mM ion concentrations. This study gives an initial insight into a satisfactory synergistic effect of *C. thermophilum* thermostable cellulases for the hydrolysis of different paper materials, which provides a potential combination of enzymes for industrial applications, including environmentally friendly waste management and cellulosic ethanol production.

Keywords *Chaetomium thermophilum* · Thermostable cellulase · Synergistic reaction · Waste paper · Biodegradation · Metal ion-insensitivity

Introduction

Waste paper, which is the major portion of solid waste, is generated in large volumes by humans worldwide. Although direct disposal of such wastes to soil is the most common traditional management, it has a negative effect on the environment in that dangerous gases are released [1, 2]. Paper recycling affords an effective approach to reduce the environmental impact [3], and it has also been demonstrated that paper recycling increases sustainability in raw material use and has a positive economic effect [4, 5]. In contrast, it is undeniable that the harmful chemical substances would be accumulated in paper recycling process, for example, the migration of chemicals from packaging materials into foodstuffs would adversely affect human health [6, 7]. As a consequence, the development of innovative waste management solution seems highly attractive.

Paper is made of connected cellulose fibers, which can be biodegraded into glucose. Thus, as a safe and environmentally friendly alternative, enzymatic degradation of waste papers has attracted considerable attention because it can greatly reduce the amount of biomaterial waste [8]. Furthermore, the conversion of paper materials into biological products is a potential biotechnology procedure to reduce the demand for fossil fuels [9]. More than 80 billion liters of cellulosic bioethanol are generated from waste paper worldwide each year, which could effectively replace gasoline consumption and reduce greenhouse gas emissions [10].

Although the metabolic process of cellulolytic microorganisms contributes to paper biodeterioration [11], the predominant biochemical mechanism is enzymatic degradation

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Engineering the conserved and noncatalytic residues of a thermostable β-1,4-endoglucanase to improve specific activity and thermostability

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Endoglucanases are increasingly applied in agricultural and industrial applications as a key biocatalyst for cellulose biodegradation. However, the low performance in extreme conditions seriously challenges the enzyme's commercial utilization. To obtain endoglucanases with substantially improved activity and thermostability, structure-based rational design was carried out based on the *Chaetomium thermophilum* β-1,4-endoglucanase CTendo45. In this study, five mutant enzymes were constructed by substitution of conserved and noncatalytic residues using site-directed mutagenesis. Mutants were constitutively expressed in *Pichia pastoris*, purified, and ultimately tested for enzymatic characteristics. Two single mutants, Y30F and Y173F, increased the enzyme's specific activity 1.35- and 1.87-fold using carboxymethylcellulose sodium (CMC-Na) as a substrate, respectively. Furthermore, CTendo45 and mutants exhibited higher activity towards β-D-glucan than that of CMC-Na, and activities of Y173F and Y30F were also increased obviously against β-D-glucan. In addition, Y173F significantly improved the enzyme's heat resistance at 80 °C and 90 °C. More interestingly, the double mutant Y30F/Y173F obtained considerably higher stability at elevated temperatures but failed to inherit the increased catalytic efficiency of its single mutant counterparts. This work gives an initial insight into the biological function of conserved and noncatalytic residues of thermostable endoglucanases and proposes a feasible path for the improvement of enzyme redesign proposals.

Cellulose, the most abundant renewable carbon resource on earth, is generally considered a sustainable feedstock to replace fossil fuels for biochemical and biotechnological production[1]. As a strategy for cellulose utilization, enzymatic hydrolysis has been widely applied for practical applications[2–5]. Endoglucanase (EC 3.2.1.4), which randomly hydrolyses β-1,4-glucosidic bonds in amorphous regions of cellulose chains to catalyse the initial attack on the biopolymer, is a major catalyst for cellulose biodegradation[6,7].

The high cost of preparation and low performance in extreme reaction conditions have been perceived as major bottlenecks to industrial applications[8–10]. One effective approach to reduce enzyme-related costs is to enhance enzyme performance[11]. Thus, recent developments in enzyme production focus on the improvement of hydrolysis efficiency and specific tolerability, allowing the production of cheaper and stronger enzymes for industrial use[12]. Rational protein engineering is an efficient genetic approach to optimize properties through structural analysis and functional prediction[13,14]. Recently, more attention has been drawn to the underlying function of important residues, along with practicality of modifying conserved noncatalytic residues[15–17], to generate mutant enzymes with improved properties and to help elucidate structure-function relationships[18].

Generally, thermostable enzymes have excellent tolerance to various harsh conditions, including high salt concentrations and extreme pHs[19,20]. More importantly, in order to profoundly improve hydrolysis efficiency at high temperatures while simultaneously reducing microbial contamination in reaction processes, it is important for enzymes to be both thermoactive and thermostable[21,22]. Therefore, thermostable endoglucanases with excellent

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Characterization of a novel thermostable GH45 endoglucanase from *Chaetomium thermophilum* and its biodegradation of pectin

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A novel thermostable endoglucanase (CTendo45) encoding gene was cloned from *Chaetomium thermophilum* and heterologously expressed in *Pichia pastoris*. Sequence alignment indicated that the CTendo45 enzyme belonged to glycoside hydrolase family 45. The recombinant enzyme was purified by Ni^{2+} affinity chromatography, and its apparent molecular mass was estimated to be 32 kDa by SDS-PAGE. The purified enzyme displayed maximum activity at 70°C and pH 4. CTendo45 was stable at 60°C for 1 h, and residual activities of 78.9% and 65.6% were estimated after 1 h at 70°C and 80°C, respectively. Ca^{2+}, Zn^{2+}, Mg^{2+}, Cu^{2+} and Mn^{2+} were found to have beneficial effects on the enzyme activity to different degrees. The specific activity of purified CTendo45 was 1.52 IU mg^{-1} and the K_m value was 59.6 μg ml^{-1} with a sodium carboxymethyl cellulose substrate. Moreover, CTendo45 exhibited high hydrolysis activity towards pectin, and the hydrolysis products were mainly galacturonic acid oligosaccharides. CTendo45 is the first reported bifunctional enzyme in glycoside hydrolase family 45 from *C. thermophilum* that is able to hydrolyze both cellulose and pectin. The biochemical properties of this recombinant CTendo45 make it a potentially effective glycoside hydrolase for industrial applications.

[**Key words:** Glycoside hydrolase family 45; Endoglucanase; Heterologous expression; Thermostability; Bifunctional enzyme]

Cellulose is regarded as the most abundant renewable carbon resource on the planet. Endoglucanases (EC 3.2.1.4) are responsible for the endohydrolysis of β-1,4-glucosidic linkages in cellulose. For this reason, endoglucanases have attracted extensive attention in recent years, particularly in biotechnological and industrial applications, such as in the ethanol biorefinery and the feed industries (1). According to the classification of the carbohydrate-active enzymes database (CAZy), endoglucanases are assigned to 14 glycoside hydrolase (GH) families: GH5-GH10, GH12, GH26, GH44, GH45, GH48, GH51, GH74, and GH124 (2). GH45 endoglucanases, in particular, have attracted attention due to their low molecular weights and high activities. These properties make GH45 family enzymes important in many specialized bioindustrial applications. All of the members of the GH45 family are reported to be endoglucanases in CAZy, with no other hydrolytic activities. Most of the GH45 endoglucanases identified to date originate from fungal sources. At present, only five crystal structures of GH45 endoglucanases are available from different organisms: *Humicola grisea* var. *thermoidea* (PDB: 1HD5), *Humicola insolens* DSM 1800 (PDB: 3ENG) (3,4), *Melanocarpus albomyces* (PDB: 1OA7) (5), *Mytilus edulis* (PDB: 1WC2) and *Phanerochaete chrysosporium* K-3 (PDB: 3X2G) (6).

Chaetomium thermophilum, a thermophilic fungus of the phylum Ascomycota, has been used to identify novel glycoside hydrolases. In the past few decades, many enzymes from *C. thermophilum* have been characterized and commercialized, in particular, thermostable glycoside hydrolases. Generally, significant thermal stability at high temperatures is an important requirement for a commercial cellulase because thermostable enzymes can effectively improve the hydrolysis efficiency and reduce possible contamination in industrial processes (7). To satisfy the demand for thermostable enzyme production, heterologous expression has become the principal method in the enzyme preparation industry (8). Many enzymes from *C. thermophilum* have been heterologously expressed, such as a GH55 β-1,3-glucanase (9), a manganese superoxide dismutase (MnSOD) (10) and a GH6 cellobiohydrolase (11). However, although there are significant glycoside hydrolases in thermophilic *C. thermophilum*, GH45 endoglucanases have not been reported to date. In this study, a novel GH45 endoglucanase, CTendo45, was isolated and characterized. This is the first report that shows that a GH45 endoglucanase from *C. thermophilum* has the ability to hydrolyze pectin.

MATERIALS AND METHODS

Strain, vector, and materials The *C. thermophilum* strain was isolated from bovine feces at Tengchong (Yun'nan, China). The strain was deposited in the publicly accessible culture collection CGMCC (no. 3.17990), Beijing, China. *Escherichia coli* T1 (TransGen Bio, Beijing, China) was used for gene cloning. *Pichia pastoris* GS115 (Invitrogen, Carlsbad, CA, USA) was used for recombinant protein production. The *Pichia* secretory protein expression vector pPIC9K (Invitrogen) was used in the expression system. Primers were synthesized by Sangon Biotech (Shanghai, China).

Gene cloning and construction of the yeast expression system Total RNA was isolated from the mycelia of *C. thermophilum* using Trizol reagent (Invitrogen) according to the manufacturer's instructions. Reverse transcription was performed according to the RNA PCR Kit 3.0 instructions (Takara Bio, Shiga, Japan). The *CTendo45* gene (Genbank accession number KC441877), based on the genomic DNA of *C. thermophilum* (http://ct.bork.embl.de/), was amplified using the primers 5′-

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Full Length Article

A Novel Application Potential of GH6 Cellobiohydrolase CtCel6 from Thermophilic *Chaetomium thermophilum* for Gene Cloning, Heterologous Expression and Biological Characterization

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Abstract

Chaetomium thermophilum is a thermophilic fungus expressed a series of glycoside hydrolases. Genome sequence analysis of *C. thermophilium* revealed that *ctcel6* gene encoded a putative cellobiohydrolase which composed of 397 amino acid residues including a predicted signal peptide sequence. Ctcel6 gene was cloned, heterologously expressed in *Pichia pastoris* and purified by Ni^{2+} affinity chromatography. Sequence alignment indicated that CtCel6 enzyme belonged to glycoside hydrolase family 6 (GH6) and the molecular mass of purified recombinant enzyme CtCel6 was 42 kDa by SDS-PAGE analysis. Characterization of recombinant CtCel6 exhibited high hydrolysis activity and excellent thermostability. The optimum reaction temperature and pH was 70°C and pH 5, respectively. The bivalent metallic cations Mg^{2+} and Ca^{2+} significantly enhanced the activity of CtCel6. The specific activity of CtCel6 enzyme was 1.27 U/mg and K_m value was 0.38 mM on β-D-glucan. The substrate specificity and hydrolysis products insisted that CtCel6 was an exo-/endo-type cellobiohydrolase. The biochemical properties of recombinant CtCel6 made it potentially effective for bioconversion of biomass and had tremendous potential in industrial applications such as enzyme preparation industry and feed processing industry. © 2017 Friends Science Publishers

Keyword: Glycoside hydrolase family 6; Cellobiohydrolase; Heterologous expression; Thermostable enzyme; Bioconversion

Introduction

Lignocellulosic biomass, the most abundantly renewable and available carbohydrate resource on the earth, has been regarded as an available feedstock for biochemical and biotechnological applications to produce biofuels and chemicals (Margeot *et al.*, 2009). Since the global energy crisis and environmental pollution are intensifying, it has attracted extensive attention to the utilization of lignocellulosic biomass. Cellulose is the major composition of lignocellulosic biomass and the bioconversion of cellulose needs a combined effect of three classes of hydrolytic enzymes, including endoglucanases (EC 3.2.1.4), cellobiohyrolases (EC 3.2.1.176; EC 3.2.1.91) and β-glucosidases (EC 3.2.1.21) (Lynd *et al.*, 2002; Sánchez, 2009). Cellobiohyrolases are a group of glycoside hydrolases, which hydrolyzed oligosaccharides of assorted lengths generated by endoglucanases to cellobiose (Ragauskas *et al.*, 2006). According to the classification of the carbohydrate-active enzymes database (CAZy), cellobiohyrolases are assigned to 5 glycoside hydrolase families: GH5-7, GH9 and GH48 (Cantarel *et al.*, 2009). In particular, the cellobiohydrolases of GH6 are widely believed to act processively from the non-reducing terminal of cellulose chains to generate cellobiose. GH6 family members are mainly produced by bacterial and fungal sources, and the hydrolysis mechanism is inverting. GH6 family includes both endoglucanases and cellobiohydrolases, many GH6 endoglucanases have been reported, such as *Thermobifida fusca* Cel6A (Ali *et al.*, 2015), *Thermobifida halotolerans* GH6 endoglucanase (Yin *et al.*, 2015), and *Cellulosimicrobium funkei* CelL (Kim *et al.*, 2016). However, the study of GH6 cellobiohydrolases has been reported rarely.

Chaetomium thermophilum is a thermophilic fungus living in the high temperature environment up to 60°C belonged to the phylum Ascomycota. By now, many glycoside hydrolases have been isolated from *C. thermophilum*, such as a GH55 β-1,3-glucanase (Papageorgiou and Li, 2015), a β-glucosidase (Xu *et al.*, 2011) and a cellobiohydrolase II (Wang *et al.*, 2013). Generally, *C. thermophilum* glycoside hydrolases are thermostable and have a high optimal reaction temperature based on the previous researches. Thermostable enzymes have potential advantages in lignocelluloses conversation, on account of effectively improving hydrolysis efficiency and reducing the possible contamination at high temperature in industrial processes (Huy *et al.*, 2016).

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Characterization of a novel thermostable GH7 endoglucanase from *Chaetomium thermophilum* capable of xylan hydrolysis

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ABSTRACT

A new endoglucanase encoding gene (*ctendo7*) was cloned from the thermophilic fungus *Chaetomium thermophilum* and heterologously expressed in *Pichia pastoris*. The recombinant CTendo7 enzyme was purified by Ni^{2+} affinity chromatography and subsequently characterized. CTendo7 belongs to glycoside hydrolase family 7, and exhibited considerable activity against sodium carboxymethyl cellulose (CMC-Na) and xylan of 1.91 IU/mg and 3.05 IU/mg at the optimum reaction condition of 55 °C, pH 5.0, respectively. The purified enzyme displayed relatively good thermostability. The residual endoglucanase and xylanase activities were 74.3% and 66.2% after a 60 min pre-incubation at 70 °C. Additionally, Ag^{+}, Fe^{3+} and Cu^{2+} negatively affected the enzyme's activity, while the presence of 1 mM and 5 mM Mn^{2+} significantly enhanced both endoglucanase and xylanase activities. Generation of soluble oligosaccharides from lignocellulose is a critical step in bioethanol production, and it is noteworthy that CTendo7 produced cello-oligosaccharides and xylo-oligosaccharides from the continuous enzymatic saccharification of CMC-Na and xylan, respectively. This is the first detailed report on a novel bifunctional endoglucanase/xylanase enzyme from *C. thermophilum*. Furthermore, the excellent properties of CTendo7 distinguish it as a promising candidate for industrial lignocellulosic biomass conversion.

1. Introduction

Lignocellulose represents the most abundant renewable carbon source on the planet and is composed of three major biopolymeric substances, including cellulose (35–50%), hemicellulose (20–30%, mainly xylan) and lignin (20–30%) [1]. To initiate the production of biofuels and other value-added products from cellulosic biomass, bioconversion of lignocellulosic components into fermentable saccharides is necessary [2]. Endoglucanase (EC 3.2.1.4) plays a primary role in hydrolyzing cellulose and randomly cleaves β-1,4-glucosidic bonds in amorphous regions of cellulose chains to catalyze the initial attack on the biopolymer [3]. Similarly, xylanase (EC 3.2.1.8) acts as a xylanolytic enzyme that breaks down β-1,4-xylosidic linkages in the cellulose backbone and then decomposes the linear polysaccharide β-1,4-xylan, a main chain of hemicellulose, to produce xylo-oligosaccharides [4]. Moreover, the catalytic performance of endoglucanases against lignocellulose can be effectively enhanced by the addition of xylanases, since the deconstruction of hemicellulose layers between microfibers contributes to further improve the accessibility of cellulose to hydrolytic enzymes [5]. Due to the importance of the two enzymes in lignocellulose hydrolysis, a bifunctional endoglucanase/xylanase could be advantageous in biotechnological applications, particularly in the ethanol biorefinery and feed industries [6].

According to the classification of the Carbohydrate-Active Enzyme (CAZy) database, endoglucanases are widespread among 14 glycoside hydrolase (GH) families: GH5-GH10, GH12, GH26, GH44, GH45, GH48, GH51, GH74, and GH124 [7]. It was reported that xylanase-like activity is a common feature shared only by endoglucanases of the GH7 family [2], which can be ascribed to the structural homology between GH7 endoglucanases and xylanases that potentially originated from a diverged ancestral gene [8].

It is especially noteworthy that an integrated bio-refinery of lignocellulosic materials for ethanol production requires elevated temperatures [9]. In this regard, as ideal catalysts, thermostable lignocellulolytic enzymes with excellent activity are always preferred in ethanol bio-refinery, since they can provide additional advantages. Generally, thermostable enzymes have considerable tolerance to harsh conditions, including high salt concentrations and extreme pHs [10, 11]. More importantly, enzymes that are both thermoactive and

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Improving the thermostability of a thermostable endoglucanase from *Chaetomium thermophilum* by engineering the conserved noncatalytic residue and N-glycosylation site

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ABSTRACT

Endoglucanases provide an attractive avenue for the bioconversion of lignocellulosic materials into fermentable sugars to supply cellulosic feedstock for biofuels and other value-added chemicals. Thermostable endoglucanases with high catalytic activity are preferred in practical processes. To improve the thermostability and activity of the thermostable β-1,4-endoglucanase CTendo45 isolated from the thermophilic fungus *Chaetomium thermophilum*, structure-based rational design was performed by using site-directed mutagenesis. When inactivated mutation of the unique N-glycosylation sequon (N88-E89-T90) was implemented and the conserved Y173 residue was substituted with phenylalanine, a double mutant T90A/Y173F demonstrated enzymatic activity that dramatically increased 2.12- and 1.82-fold towards CMC-Na and β-D-glucan, respectively. Additionally, T90A/Y173F exhibited extraordinary heat endurance after 300 min of incubation at elevated temperatures. This study provides a valid approach to the improvement of enzyme redesign protocols and the properties of this endoglucanase mutant distinguish it as an excellent candidate enzyme for industrial biomass conversion.

1. Introduction

Lignocellulosic biomass is the earth's most abundant utilizable organic resource with great potential for the sustainable production of chemicals and biofuels [1,2]. Cellulose serves as a major component (approximately 35–50%) of lignocellulosic biomass, followed by hemicellulose (20–30%) and lignin (20–30%) [3]. Endoglucanase (EC 3.2.1.4), which is a critical set of cellulases for lignocellulose deconstruction, randomly cleaves the internal β-1,4-glucosidic bonds in cellulose fibers and triggers the initial catalytic attack on biopolymer chains in amorphous regions [4].

From a practical point of view, the inadequate performance and instability of commercial cellulases under extreme reaction conditions is now considered as the main obstacle to their effective application in industrial and agricultural fields which results in extending the reaction period and raising the production cost [5]. Therefore, for modern cellulase preparation research and development, the improvement of enzyme specific tolerability and catalytic efficiency is essential [6,7]. To date, structure-based rational engineering is an effective approach to enhancing enzymatic properties, which is implemented based on the modification of some crucial residues and domains in terms of the enzyme's structure-function relationship [8,9]. In addition, more attention has been focused on the optimization of conserved residues in the active site architecture, which play important roles in modulating enzyme structure and catalytic properties [10,11].

Elevating the reaction temperature is generally conducive to substantially increasing the hydrolysis rate and preventing microbial contamination in the realistic biorefinery of lignocellulosic saccharification [12]. Thus, thermostability is a valuable property for cellulases in practice. Utilizing potent thermostable cellulases endowed with pronounced activity at high temperatures is beneficial for accelerating the catalytic process, shortening the reaction period and reducing the enzyme dosage [2,13]. N-glycosylation, which represents a ubiquitous post-translational modification in eucaryon, is capable of regulating the thermal stability and hydrolysis activity of cellulases [14–17]. In the case of a glycoprotein, the carbohydrate unit is covalently connected to the specific asparagine residue within the sequon Asn-Xaa-Ser/Thr (where Xaa cannot be Pro) [18].

In our previous work, rational design of a thermostable GH45 β-1,4-endoglucanase CTendo45 from *Chaetomium thermophilum* was performed to further enhance the thermostability by substituting the conserved noncatalytic Y173 residue with F in the active site architecture using site-directed mutagenesis [19]. Additionally, when the unique N-glycosylation sequon in CTendo45 was modified, a T90A variant exhibited superior characteristics of high temperature tolerance [17]. In this study, a double mutant T90A/Y173F was generated and exhibited enhanced thermostability compared with that of its single counterparts, providing a promising biocatalyst for diverse biotechnological

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Identification and Characterization of a Novel Hyperthermostable Bifunctional Cellobiohydrolase-Xylanase Enzyme for Synergistic Effect With Commercial Cellulase on Pretreated Wheat Straw Degradation

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The novel cellobiohydrolase gene *ctcel7* was identified from *Chaetomium thermophilum*, and its recombinant protein CtCel7, a member of glycoside hydrolase family 7, was heterologously expressed in *Pichia pastoris* and biochemically characterized. Compared with commercial hydrolases, purified CtCel7 exhibited superior bifunctional cellobiohydrolase and xylanase activities against microcrystalline cellulose and xylan, respectively, under optimal conditions of 60°C and pH 4.0. Moreover, CtCel7 displayed remarkable thermostability with over 90% residual activity after heat (60°C) treatment for 180 min. CtCel7 was insensitive to most detected cations and reagents and preferentially cleaved the β-1,4-glycosidic bond to generate oligosaccharides through the continuous saccharification of lignocellulosic substrates, which are crucial for various practical applications. Notably, the hydrolysis effect of a commercial cellulase cocktail on pretreated wheat straw was substantively improved by its combination with CtCel7. Taken together, these excellent properties distinguish CtCel7 as a robust candidate for the biotechnological production of biofuels and biobased chemicals.

Keywords: bifunctional, thermostable, cellobiohydrolase, xylanase, synergism, pretreated corn stover

INTRODUCTION

Lignocellulosic biomass is the most abundant renewable resource in nature and represents a promising feedstock for the agricultural, biochemical and biofuel industries (Liu et al., 2019). Cellulose and hemicellulose are the primary components of lignocellulosic biomass with mass ratios of 35–50% and 20–30%, respectively (Penttilä et al., 2018). Due to the structural complexity of lignocellulosic biomass, its efficient enzymatic depolymerization necessitates the synergistic actions of a diverse set of glycoside hydrolases (GHs), particularly cellulase and xylanase. Because multifunctional glycoside hydrolases with excellent activity are favorable for carbohydrate conversion (Cao et al., 2018; Wang et al., 2019), much effort has been devoted to the bioprospecting of these potent catalysts to allow their application in industrial fields. Although many multifunctional GHs have been identified (Yang et al., 2017; Tan et al., 2018; Liu et al., 2019), an inherent enzymatic feature of significant thermostability, which is preferred for

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A Conserved Glycoside Hydrolase Family 7 Cellobiohydrolase PsGH7a of *Phytophthora sojae* Is Required for Full Virulence on Soybean

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Phytopathogens deploy glycoside hydrolases (GHs) to disintegrate plant cell walls for nutrition and invasion. However, the pathogenic mechanisms of the majority of GHs in virulence remain unknown, especially in oomycetes. In this study, a *Phytophthora sojae* gene encodes a GH7 family cellobiohydrolase, named *PsGH7a*, was identified. *PsGH7a* was highly induced during the cyst germination and infection stages. PsGH7a is conserved in oomycetes, and shares a high amino acid sequence identity (>85%) within *Phytophthora* genus. The recombinant PsGH7a catalyzes the hydrolysis of β-1,4-glucan and avicel, which represent the major components of cellulose in plant cell wall. The mutation of catalytic residue Glu236 to alanine resulted in a lower catalytic activity. In addition, the PsGH7a promotes *Phytophthora* invasion, while the mutant can not. Notably, PsGH7a protein triggers hypersensitive cell death in diverse plants. PsGH7a knockout mutants were generated via CRISPR/Cas9 system, to investigate its biological function. Compared to wild-type strain P6497, the mutants showed reduced virulence on susceptible soybean, indicates PsGH7a is indispensable to *P. sojae* virulence.

Keywords: ***Phytophthora sojae*, glycoside hydrolase 7, virulence, soybean, cellobiohydrolase**

INTRODUCTION

The battle between plants and microbes is the product of million-years of co-evolution. The front line of the plant defense is numerous physical barriers such as the cell walls, waxes and hairs (Hématy et al., 2009). A primary challenge for microbial pathogens is to penetrate the formidable and dynamic barrier of plant cell walls, which are constructed of cellulose, hemicellulose, pectin, and joined by complex distinct connection types (Somerville et al., 2004; Vorwerk et al., 2004; Wang et al., 2019).

Plant pathogens produce cell wall degrading enzymes (CWDEs) as part of their arsenal for nutrition and plant invasion (Faure, 2002; Martinez et al., 2004; Hashimoto et al., 2007; Hématy et al., 2009; Bakunina et al., 2013; van Wyk et al., 2017; Pluvinage et al., 2019). Phytopathogenic fungi and oomycetes are unique microbial pathogens that being able to break the intact physical surfaces of host plants (Soanes et al., 2007). Many plant-pathogenic fungi secrete a range of CWDEs to degrade the host cell wall, such as glycoside hydrolases, polysaccharide lyases, and esterases,

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RESEARCH Open Access

Improvement of the catalytic activity and thermostability of a hyperthermostable endoglucanase by optimizing N-glycosylation sites

Chao Han*, Qunqing Wang, Yanxu Sun, Ruirui Yang, Mengyu Liu, Siqi Wang, Yifan Liu, Lifan Zhou and Duochuan Li*

Abstract

Background: Endoglucanase has been extensively employed in industrial processes as a key biocatalyst for lignocellulosic biomass degradation. Thermostable endoglucanases with high catalytic activity at elevated temperatures are preferred in industrial use. To improve the activity and thermostability, site-directed mutagenesis was conducted to modify the N-glycosylation sites of the thermostable β-1,4-endoglucanase CTendo45 from *Chaetomium thermophilum*.

Results: In this study, structure-based rational design was performed based on the modification of N-glycosylation sites in CTendo45. Eight single mutants and one double mutant were constructed and successfully expressed in *Pichia pastoris*. When the unique N-glycosylation site of N88 was eliminated, a T90A variant was active, and its specific activity towards CMC-Na and β-D-glucan was increased 1.85- and 1.64-fold, respectively. The mutant R67S with an additional N-glycosylation site of N65 showed a distinct enhancement in catalytic efficiency. Moreover, T90A and R67S were endowed with extraordinary heat endurance after 200 min of incubation at different temperatures ranging from 30 to 90 °C. Likewise, the half-lives ($t_{1/2}$) indicated that T90A and R67S exhibited improved enzyme thermostability at 80 °C and 90 °C. Notably, the double-mutant T90A/R67S possessed better hydrolysis activity and thermal stability than its single-mutant counterparts and the wild type.

Conclusions: This study provides initial insight into the biochemical function of N-glycosylation in thermostable endoglucanases. Moreover, the design approach to the optimization of N-glycosylation sites presents an effective and feasible strategy to improve enzymatic activity and thermostability.

Keywords: Endoglucanase, N-glycosylation site, Thermostability, Specific activity

Background

Plant polysaccharide depolymerization by synergistic enzyme cocktails is crucial for the production of lignocellulosic biofuels [1]. These biomass-degrading enzymes are primarily composed of a diverse set of glycoside hydrolases (GHs) that efficiently catalyze the conversion of lignocellulose to glucose [2]. Endoglucanase (EC 3.2.1.4), which randomly deconstructs the internal β-1,4-glucosidic linkages in amorphous regions of biopolymer fibers to trigger an initial catalytic attack on cellulose chains, is an essential biocatalyst for cellulose degradation [3].

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Enhancement of catalytic activity and thermostability of a thermostable cellobiohydrolase from *Chaetomium thermophilum* by site-directed mutagenesis

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ABSTRACT

Enzymatic saccharification of lignocellulosic biomass is increasingly applied in agricultural and industrial applications. Nevertheless, low performance in the extreme environment severely prevents the utilization of commercial enzyme preparations. To obtain cellobiohydrolases with improved catalytic activity and thermostability, structure-based rational design was performed based on a thermostable cellobiohydrolase CtCel6 from *Chaetomium thermophilum*. In the present study, four conserved and noncatalytic residue substitutions were generated via site-directed mutagenesis. Mutations were heterologously expressed in yeast *Pichia pastoris*, purified, and ultimately assayed for enzymatic characteristics. The mutant Y119F increased the catalytic activity 1.82-, 1.65- and 1.43-fold against β-D-glucan, phosphoric acid swollen cellulose (PASC) and carboxymethylcellulose sodium (CMC-Na), respectively. In addition, S131 W effectively enhanced the enzyme's heat resistance to elevated temperatures. The half-life ($t_{1/2}$) of this mutant enzyme was increased 1.42- and 2.40-fold at 80 °C and 90 °C, respectively, compared to the wild-type. This study offers initial insight into the biological function of the conserved and noncatalytic residues of thermostable cellobiohydrolases and provides a valid approach to the improvement of enzyme redesign proposal.

1. Introduction

Cellulose is the main composition of lignocellulosic biomass and consists of β-1,4-linked D-glucopyranosides to form adjacent linear-chain molecular polymers [1]. One crucial determinant to restrict saccharification efficiency is the recalcitrance of crystalline region in cellulose substrates [2]. Cellobiohydrolase (EC 3.2.1.91), which acts processively on the end of exposed cellulosic polysaccharide to release cellobiose and cellooligosaccharide, is an essential biocatalyst for cellulose decomposition [3], especially for the native cellulosic material that contains a great proportion of crystalline polymers [4].

The high cost of commercial enzymes and low performance in extreme conditions are considered as major obstacles to industrial applications [5]. Thus, there is a significant impetus to improve enzymatic hydrolysis efficiency and specific tolerability for the recent development of cellulase preparation [6]. As an efficient genetic approach to optimize properties, rational protein engineering contributes to generating mutations with enhanced properties and simultaneously elucidates the enzyme's structure-function relationship [7,8]. Recently, more attention has been drawn to the potential functional residues, which play important roles in modulating enzyme structure and catalytic properties, especially these conserved noncatalytic residues [9,10].

Thermoactive and thermostable cellulases, in general, have favorable effects on the resistance of adverse conditions, including high salt concentrations and extreme pHs. In particular, enzymes with such excellent properties are desired to effectively enhance hydrolysis efficiency at elevated temperatures while simultaneously reducing microbial contamination in industrial processes [11]. Therefore, it is essential to explore thermoactive enzymes with considerable thermostability. *Chaetomium thermophilum* produces multiple thermostable cellulases with high efficiency [12], such as a β-1,4-endoglucanase CTendo45 [7] and two cellobiohydrolases CtCel6 [13] and CtCBH1 [14]. According to the classification of the Carbohydrate-Active Enzyme (CAZy) database [15], cellobiohydrolases are mainly assigned into two glycoside hydrolase families (GH6 and GH7). Besides, the GH6 cellobiohydrolase is extensively considered to act processively from the non-reducing terminal of

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Selection of organosilicone surfactants for tank-mixed pesticides considering the balance between synergistic effects on pests and environmental risks

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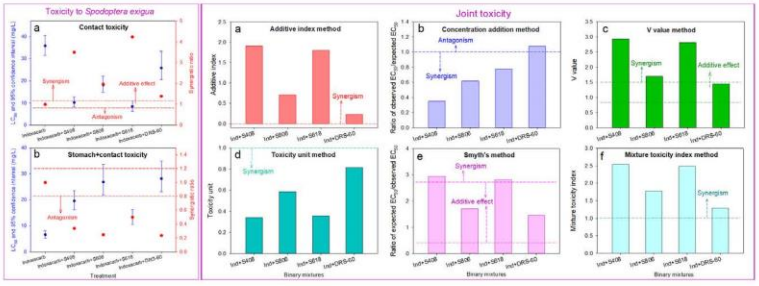
HIGHLIGHTS

- Organosilicone were synergistic on the contact toxicity of indoxacarb.
- Organosilicone were highly toxic to *D. magna* but less toxic to *B. rerio*.
- Additive index, concentration addition and toxicity unit were robust.
- V-value and equilibrium curve were conservative in the joint toxicity evaluation.

GRAPHICAL ABSTRACT



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ABSTRACT

In this study, the bioactivities of binary mixtures of organosilicone surfactants and indoxacarb against two Lepidopteran pests were investigated along with their environmental risks. All of the tested organosilicone surfactants had obvious synergistic effects on the contact toxicity of indoxacarb against *Spodoptera exigua* and *Agrotis ipsilon*. However, all of the organosilicone surfactants exhibited certain antagonism for indoxacarb against *S. exigua* in terms of stomach & contact toxicity; both Silwet-408 and Silwet-806 exhibited additivity against *A. ipsilon*, whereas Silwet-618 and Silwet-DRS-60 exhibited synergism and slight antagonism, respectively. All of the tested chemicals were highly toxic to *Daphnia magna*, among which Silwet-DRS-60 had the lowest acute toxicity (EC_{50} of 94.91 μg/L). However, these chemicals were less toxic to *Brachydanio rerio*. Silwet-DRS-60 had a low toxicity to *B. rerio*, while Silwet-408, Silwet-806 and Silwet-618 were moderately toxic to *B. rerio*. For the joint toxicity evaluation of organosilicone surfactants and indoxacarb to *D. magna* and *B. rerio*, the additive index method, concentration addition method and toxicity unit method were robust in judging synergism or antagonism, whereas other methods were more conservative; the V-value method and equilibrium curve method exhibited high robustness and viability in evaluating the combined effects of binary mixtures. Overall, we

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frontiers in Chemistry

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Easily Tunable Membrane Thickness of Microcapsules by Using a Coordination Assembly on the Liquid-Liquid Interface

Bei-xing Li[1,2], *Xiao-xu Li*[1], *Yang Liu*[1], *Da-xia Zhang*[1,2], *Jin Lin*[2], *Wei Mu*[2] *and Feng Liu*[1,2]*

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A model solvent, 1,3,5-trimethylbenzene, was encapsulated using coordination assembly between metal ions and tannic acid to reveal the deposition of coordination complexes on the liquid-liquid interface. The deposition was confirmed by zeta potential, energy dispersive spectroscopy and X-ray photoelectron spectroscopy. Scanning electron microscopy and transmission electron microscopy were integrated to characterize the microcapsules (MCs). According to atomic force microscopy height analysis, membrane thickness of the MCs increased linearly with sequential deposition. For MCs prepared using the Fe^{3+}-TA system, the average membrane thicknesses of MCs prepared with 2, 4, 6, and 8 deposition cycles were determined as 31.3 ± 4.6, 92.4 ± 15.0, 175.4 ± 22.1, and 254.8 ± 24.0 nm, respectively. Dissolution test showed that the release profiles of all the four tested MCs followed Higuchi kinetics. Membrane thicknesses of MCs prepared using the Ca^{2+}-TA system were much smaller. We can easily tune the membrane thickness of the MCs by adjusting metal ions or deposition cycles according to the application requirements. The convenient tunability of the membrane thickness can enable an extensive use of this coordination assembly strategy in a broad range of applications.

Keywords: microcapsule, polyphenol, metal ion, deposition, coordination assembly, membrane thickness

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INTRODUCTION

Microencapsulation technology is a promising approach that has been widely reported to protect sensitive core materials, including but not limited to chemicals and living biomaterials (Parthasarathy and Martin, 1994; Anderson and Shive, 2012; Tong et al., 2012; Li B.-X. et al., 2017). The past few decades have seen significant advances of microencapsulation in drug delivery (Wang et al., 2006; De Koker et al., 2012; Simoes et al., 2015; Jia et al., 2017; Li H. et al., 2017), material industry (White et al., 2001; Su and Schlangen, 2012; Jamekhorshid et al., 2014), biomaterials (Parthasarathy and Martin, 1994; Zhi and Haynie, 2006; Kurayama et al., 2012; Ekanem et al., 2017; Zhao et al., 2017), agrochemicals (Li et al., 2016, 2018; Liu et al., 2016; Liang et al., 2017), food industry (Xu et al., 2013), and other fields. There are numerous encapsulation methods focusing on *in situ* polymerization (Su et al., 2013; Zuo et al., 2014), interfacial polymerization (Wagh et al., 2009), spray drying (Zhang and Zhong, 2013), solvent

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Alcohol ethoxylates significantly synergize pesticides than alkylphenol ethoxylates considering bioactivity against three pests and joint toxicity to *Daphnia magna*

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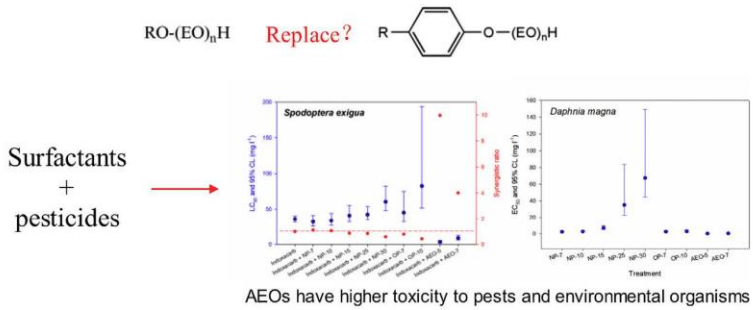
HIGHLIGHTS

- AEOs had higher synergistic effects on pesticides than APEOs.
- Additive index, toxicity unit, V value and isobologram methods were integrated.
- Synergistic ratios and toxicities of AEOs and APEOs decreased with the EO numbers.
- AEOs may be potential alternatives for APEOs in agricultural production system.

GRAPHICAL ABSTRACT



AEOs have higher toxicity to pests and environmental organisms

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ABSTRACT

Seeking alternatives for alkylphenol ethoxylates (APEOs) have been a heavily researched topic in the surfactant industry and agricultural systems. In this study, the combined effects of different ethoxylates and pesticides on the bioactivity against three pests and toxicological risks to *Daphnia magna* were investigated. Results showed that alcohol ethoxylates (AEOs) had higher synergistic effects on the bioactivity of pesticides against *Spodoptera exigua*, *Agrotis ipsilon* and *Aphis citricola* than did APEOs. In terms of the joint toxicity of the ethoxylates and pesticides to *D. magna*, additive index method, toxicity unit method, V value method and isobologram method were used in the tests. All of these methods indicated that the joint effects of APEOs + acetamiprid and APEOs + indoxacarb upon *D. magna* turned from synergism to antagonism with the increasing EO (ethylene oxide) numbers. Those of AEOs exhibited similar trends. Overall, AEOs may be potential alternatives for APEOs in agriculture as they synergize pesticides against three pests significantly more than do APEOs. However, further research should investigate the compounds' environmental risks to aquatic organisms because the AEOs were highly toxic to *D. magna*.

1. Introduction

Alkylphenol ethoxylates (APEOs) are highly consumed surfactants in the textile, cosmetic, agrochemicals and other domains owing to

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Article

Identification of Anthocyanin Composition and Functional Analysis of an Anthocyanin Activator in *Solanum nigrum* Fruits

Shaoli Wang [1,†], Zhaohui Chu [1,†], Mingxing Ren [2], Ru Jia [1], Changbao Zhao [1], Dan Fei [3], Hao Su [1], Xiaoqi Fan [1], Xiaotian Zhang [1], Yang Li [1], Yingzi Wang [4] and Xinhua Ding [1,*]

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Abstract: *Solanum nigrum* fruits have been conventionally used in beverages due to their nutritional substances such as minerals, vitamins, amino acids, proteins, sugars, polyphenols, and anthocyanins. The characterization of components and regulatory mechanism of anthocyanins in *S. nigrum* fruits have rarely been reported. In this study, we determined that the peel and flesh of *S. nigrum* fruits shared similar HPLC profiles but different contents and total antioxidant activities for anthocyanins. After an efficient purification method, mainly including extraction with pH 1.0 distilled water and then desorption with pH 1.0 95% ethanol after a DM-130 resin adsorption step to obtain more pure anthocyanin extracts, the purity of anthocyanins extracted from *S. nigrum* fruits reached 56.1%. Moreover, eight anthocyanins from *S. nigrum* fruit were identified with HPLC-MS/MS for the first time. A typical R2R3-MYB transcription factor gene, *SnMYB*, was also cloned for the first time by rapid amplification of cDNA ends (RACE)-PCR from *S. nigrum*. Moreover, the contents of anthocyanins were shown to correlate well ($r = 0.93$) with the expression levels of *SnMYB* gene during the fruit's developmental stages. Most significantly, *SnMYB* gene successfully produced high anthocyanin content (1.03 mg/g) when *SnMYB* gene was transiently expressed in tobacco leaves. Taken together, *S. nigrum* fruits are a promising resource for anthocyanin extraction, and *SnMYB* gene is an activator that positively regulates anthocyanin biosynthesis in *S. nigrum*.

Keywords: *Solanum nigrum*; anthocyanin; HPLC-MS/MS; antioxidant capacity; SnMYB transcription factor

1. Introduction

As a group of natural pigments, anthocyanins are water-soluble and provide many flowers and fruits with their purple, blue, and red colors, which promotes pollination and seed distribution [1]. Natural anthocyanins have the ability to protect plants from biotic and abiotic stress [2]. For example, anthocyanins can provide plants with increased resistance to some fungal diseases and insect damage [3,4]. Furthermore, anthocyanins are capable of protecting plants from cold damage and UV

DISEASE NOTES

OPEN

First Report of *Tobacco vein banding mosaic virus* Infecting Sesame in China

H. Y. Wang, H. H. Gong, Z. Y. Yan, W. Tang, T. S. Zhu, M. Zhao, and X. D. Li

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TOOLS SHARE

Sesame (*Sesamum indicum*) is one of the major economic crops in the world and is mainly cultivated in Asia and Africa. In the summer of 2016, we conducted a series of field surveys in Zhangqiu, Shandong Province, China, to investigate the viruses infecting sesame. Sesame plants (cv. Zhongzhi No. 11) showing symptoms of mosaic, etiolation, shoestring, and puckering were found in several fields at incidences of 20 to 35%.Young leaves of five symptomatic plants were collected for total RNA extraction using Trizol reagent (Invitrogen, Carlsbad, CA). Equal amounts of total RNAs were pooled for small RNA deep-sequencing (SRDS) analysis (Illumina HisEquation 2500) to detect potential viruses. SRDS on the pooled RNA resulted in 26,181,653 reads. Clear reads of 21 to 24 nucleotides (nt) were assembled with Trinity v2.06 and used to search homologous sequences in the NCBI nucleotide database using the BLAST tool. BLAST searches identified three potyviruses, *Watermelon mosaic virus* (WMV), *Bean common mosaic virus* (BCMV), and *Tobacco vein banding mosaic virus* (TVBMV). The percentages of the small RNA reads corresponding to WMV, BCMV, and TVBMV were 0.43, 13.48, and 1.23%, respectively. WMV and BCMV have already been reported to infect sesame in China. The contigs mapping to TVBMV were 9,240 nt, covering 96.5% of its complete genome. To further confirm the existence of TVBMV in sesame samples, the 3′-terminal region of TVBMV genome was amplified by RT-PCR using primers specific to potyviruses (Chen et al. 2001). A fragment of ~1.6 kb was amplified from three of the 12 plants tested. The resultant consensus sequences of 1,624 nt (GenBank accession no. KY247146, isolate name TVBMV-ZhangQiu) included 624 nt encoding partial NIb, 816 nt encoding CP, and 184 nt belonging to the 3′-UTR. The 3′-terminal genomic sequence of TVBMV-ZhangQiu shared identities of 90.64 to 98.09% at the nucleotide level with other TVBMV isolates available in GenBank, while the CP-encoding sequence shared identities of 90.04 to 98.52% at the nucleotide level and 95.93 to 100.0% at the amino acid level with other TVBMV isolates. TVBMV-ZhangQiu formed a clade with isolate GSZNS (GU904069) from Gansu, China, and was clustered to the CPMC group (Zhang et al. 2011) in the phylogenetic analysis. In Western blot analysis, all the three PCR-positive plants showed positive reaction with antiserum against TVBMV (Lan et al. 2007). TVBMV has been reported to infect tobacco, potato, *Datura stramonium*, wild eggplant (*Solanum torvum*), and tomato in China (Geng et al. 2014; Roggero et al. 2000; Zhang et al. 2011). To the best of our knowledge, this is the first report that TVBMV can naturally infect sesame in China. Our results show that TVBMV is a potential threat to sesame production and provide new clues for the sustainable control of sesame viral diseases.

DISEASE NOTES

OPEN

First Report of *Papaya ringspot virus* Associated With a Ringspot Disease of Zucchini in Northern China

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Papaya ringspot virus (PRSV) is a member of the genus *Potyvirus* (family Potyviridae) and causes great economic losses in agriculture (Bateson et al. 2002). So far, PRSV has been reported to infect *Carica papaya*, *Citrullus vulgaris*, *Cucumis sativus*, *Cucurbita maxima*, *Cucurbita pepo*, *Cucurbita moschata*, *Luffa acutangula*, *Trichosanthes anguina*, and *Sicyos angulatus* in the southern (subtropical) Chinese provinces of Guangdong, Guangxi, Hainan, Fujian, Taiwan, and Yunnan (Huang et al. 2008; Liao et al. 2005). However, PRSV has not been detected in northern China. In August 2016, 30 to 50% zucchini (*Cucurbita pepo*; cv. Zaoqing No. 1) plants in two farms of ~10 ha in Jinan, Shandong Province, showed mosaic and distortion on leaves and ringspot on fruits, which caused huge damage to the yield and commercial value. Twelve samples were detected with DAS-ELISA using antibody against PRSV (ADGEN, Scotland, U.K.). The absorbance values of blank, negative, and positive controls were 0.119, 0.199, and 3.099, respectively, while those for the 12 samples varied from 0.649 to 3.631, indicating that all of them were infected with PRSV. To further confirm the existence of PRSV, primers PRSV-9061-F (5′-GCTCCATATGTGTCTGAGGTTG-3′), which was identical to nucleotides (nt) 9,061 to 9,082 of PRSV isolate CI (accession no. AY027810), and PRSV-10241-R (5′-CCTCACTGTAAAATAGAAGCGG-3′), which was complementary to nt 10,241 to 10,220, was designed. Reverse transcription was conducted using HiScript II reverse transcription (Vazyme, Nanjing, China) with primer PRSV10241-R. A specific fragment of ~1.2 kb was PCR-amplified using LA *Taq* DNA polymerase (Takara, DaLian, China) with primers PRSV10241-R and PRSV9061-F. The products of two independent PCR were sequenced by Biosune, Shanghai, China. The sequences was 1,058 nt in length, included 143-nt nuclear inclusion b protein (NIb) coding region, 867 nt coat protein (CP) gene, and 48 nt 3′-untranslated region (UTR). BLAST analysis showed that the virus isolate (here designated as PRSV-Jinan, GenBank accession no. KX9048/9) showed an identity of 98% at nt level with Hanoi 1, a PRSV isolate from Vietnam (JN822231). In the phylogenetic tree constructed with the CP gene using software MEGA 7.0 (Kumar et al. 2016), PRSV-Jinan formed a close branch with isolate Hanoi 1. To our knowledge, this is the first report of PRSV naturally occurring in zucchini in a northern Chinese province with a temperate climate. These findings highlight the importance of developing a suitable strategy for controlling the spread of viral diseases of zucchini in northern China.

SCIENTIFIC REPORTS

OPEN

Tobacco vein banding mosaic virus 6K2 Protein Hijacks NbPsbO1 for Virus Replication

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Chloroplast-bound vesicles are key components in viral replication complexes (VRCs) of potyviruses. The potyviral VRCs are induced by the second 6 kDa protein (6K2) and contain at least viral RNA and nuclear inclusion protein b. To date, no chloroplast protein has been identified to interact with 6K2 and involve in potyvirus replication. In this paper, we showed that the Photosystem II oxygen evolution complex protein of *Nicotiana benthamiana* (NbPsbO1) was a chloroplast protein interacting with 6K2 of *Tobacco vein banding mosaic virus* (TVBMV; genus *Potyvirus*) and present in the VRCs. The first 6 kDa protein (6K1) was recruited to VRCs by 6K2 but had no interaction with NbPSbO1. Knockdown of *NbPsbO1* gene expression in *N. benthamiana* plants through virus-induced gene silencing significantly decreased the accumulation levels of TVBMV and another potyvirus *Potato virus Y*, but not *Potato virus X* of genus *Potexvirus*. Amino acid substitutions in 6K2 that disrupted its interaction with NbPsbO1 also affected the replication of TVBMV. NbPsbP1 and NbPsbQ1, two other components of the Photosystem II oxygen evolution complex had no interaction with 6K2 and no effect on TVBMV replication. To conclude, 6K2 recruits 6K1 to VRCs and hijacks chloroplast protein NbPsbO1 to regulate potyvirus replication.

To achieve successful infection in plant, positive-stranded RNA viruses utilize host cellular membranes to form viral replication complexes (VRCs)[1–4]. Formation of VRCs can occur on various organelle membranes including endosome, endoplasmic reticulum (ER), golgi membrane, chloroplast membrane, mitochondria membrane, peroxisome and plasma membrane[1–3,5]. VRCs were reported to contain viral RNA, viral replication-associated proteins and host proteins like RNA-modifying enzymes, protein chaperones, ESCRT proteins, translation factors and proteins involved in lipid biosynthesis[6,7]. Identification of new host factor(s) necessary for virus replication will further advance our understanding of virus replication and the development of novel and sustainable antiviral strategies for agriculture.

Chloroplast is the metabolic energy manufacture in plant. It is also known to have an important role in plant virus replication. For example, *Turnip yellow mosaic virus* 140 K protein contains methyltransferase, proteinase and NTPase/helicase motifs and targets chloroplast envelope. This 140 K protein can recruit the 66 K viral RNA-dependent RNA polymerase (RdRp) to chloroplast periphery[8,9]. *Alfalfa mosaic virus* (AMV) and *Barely stripe mosaic virus* (family *Bromoviridae*) also target chloroplast for their replication[10–12]. Several chloroplast proteins have now been identified to interact with viral proteins during virus infection in plant. The replicase of *Tobacco mosaic virus* (TMV) was shown to interact with Rubisco activase (RCA) and silencing *RCA* gene expression through virus-induced gene silencing (VIGS) enhanced TMV accumulation in cells[13]. The movement protein of *Tomato mosaic virus* (ToMV) interacted with Rubisco small subunit (RbCS) and knockdown of *RbCS* expression enhanced ToMV infection in virus-inoculated plant[14].

Photosystem II (PS II) was reported to function as a light-driven, water-plastoquinone oxidoreductase. In addition, the PS II contains several extrinsic oxygen evolution complex (OEC) proteins that are known to have key roles in stabilizing the active manganese site[15,16]. OEC proteins are also known to interact with viral proteins. For example, the PS II OEC protein PsbP was shown to interact with AMV coat protein and *Rice stripe virus* (RSV) disease-specific protein. Transient over-expression of *PsbP* in *N. benthamiana* leaves led to a significant reduction of AMV accumulation. Similarly, over-expression of *PsbP* in rice plant through stable transformation strongly

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套作糯玉米对西兰花连作田土壤微生物量及酶活性的影响

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摘 要 本文研究了套作糯玉米(*Zea mays* L. *sinensis* Kulesh)及其秸秆还田的种植模式下土壤生物活性的变化,以明确套作模式和秸秆还田减缓西兰花连作田连作障碍的效果。以西兰花单作为对照,设置西兰花套作糯玉米且糯玉米秸秆还田-西兰花(B/MR-B)、西兰花套作糯玉米且糯玉米秸秆不还田-西兰花(B/M-B)、西兰花-西兰花(B-B),共3个处理,分别测定土壤微生物生物量碳(MBC)、氮(MBN)以及脲酶、蔗糖酶、中性磷酸酶、过氧化氢酶活性。结果表明:套作并秸秆还田有效提升MBC和MBN,玉米和西兰花共生阶段后期MBC和MBN较高;套作并秸秆还田可有效提高土壤脲酶、中性磷酸酶和蔗糖酶活性,促进有机养分的转化和有效化;秸秆还田前期胁迫作物分泌过氧化氢,导致土壤过氧化氢酶活性升高,套作处理可有效减缓土壤中过氧化氢的累积。相关分析表明,MBC与脲酶、蔗糖酶、中性磷酸酶、过氧化氢酶显著相关或极显著正相关,MBN与脲酶、蔗糖酶、中性磷酸酶极显著正相关。套作并秸秆还田可改善西兰花连作田土壤生态环境,在一定程度上有效缓解西兰花连作障碍。

关键词 套作;秸秆还田;连作障碍;土壤微生物生物量碳;土壤微生物生物量氮;土壤酶活性

Effects of intercropping with waxy maize on soil microbial biomass and enzyme activity of continuous broccoli field.

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Abstract: Using continuous cropping as a contrast, the effects of different intercropping patterns of waxy maize and maize straw returning on soil microbial biomass and enzyme activities were studied. The aim was to know how intercropping and maize straw returning benefit for solving continuous cropping obstacle problems in vegetable fields. Three treatments were used, including relay-intercropping mode of broccoli and waxy maize with waxy maize straw returning (B/MR-B), relay-intercropping mode of broccoli and waxy maize without waxy maize straw returning (B/M-B), broccoli continuous cropping (B-B). The activities of soil urease, sucrase, neutral phosphatase and catalase, and soil microbial biomass carbon (MBC) and nitrogen (MBN) were analyzed. Relay-intercropping with straw returning increased MBC and MBN, especially during the later co-existence period of maize and broccoli. Moreover, the activities of urease, neutral phosphatase and invertase were increased, and the transformation of organic nutrients in the soil was improved. Hydrogen peroxide was secreted by crop roots in the early stage of residues incorporation, which increased the activity of catalase. The intercropping system may have the potential to de-

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耕种模式对三江平原黑土细菌多样性的影响

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摘 要:本试验使用 Illumina MiSeq 测序平台,选取我国东北三江平原地区的大田黑土、菜园黑土和森林黑土3种样本,对不同耕种黑土表层细菌群落结构进行分析。结果显示:大田土、菜园土和森林土的微生物群落存在明显差异。大田土的群落丰富度明显低于菜园土和森林土,后两者相似度较高。大田土的 OTU 数比菜园土和森林土分别少 13.3% 和 7.6%。菜园土的微生物群落组成种类最为丰富,森林土中硝化螺旋菌门含量最高,大田土微生物群落含有的外源性物质降解相关基因相对丰度较高。可见,三江平原大田黑土的退化和土壤微生物群落的改变有关,调节土壤微生物群落结构将有助于东北大田黑土的修复。

关键词:细菌群落多样性;高通量测序;黑土地;耕种模式

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Effects of Cultivation Mode on Bacterial Diversity of Black Soil in Sanjiang Plain

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Abstract The bacterial community structure of field, vegetable garden and forest black soil in Sanjiang Plain in the northeast of China were analyzed using Illumina MiSeq sequencing platform under different cultivation modes. The results showed that the microbial community of field, vegetable garden and forest soil existed obvious differences. The community richness of field soil was significantly lower than that of vegetable garden and forest soil, and the latter two were similar. Compared with vegetable garden and forest soil, the OTU number of field soil reduced by 13.3% and 7.6% respectively. The microbial community composition of vegetable garden soil was the most abundant. The bacteria of Nitrospirae in forest soil held the highest level. The microbial community of field soil contained more abundant degradation genes of exogenous substances. Obviously, the black soil degradation in Sanjiang Plain was related to the changes of soil microbial community, and regulating the structure of soil microbial community was beneficial to the repair of field black soil.

Key words Bacterial community diversity; High-throughput sequencing; Black land; Cultivation mode

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基于DNDC模型的秸秆还田量与氮肥的耦合效应对夏玉米农田N_2O排放的影响研究

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摘 要:N_2O是一种重要的温室气体,农田已成为其重要排放来源。为明确秸秆还田量与氮肥的耦合效应对夏玉米农田N_2O排放的影响,本试验利用农田实测N_2O排放数据对DNDC模型进行验证,并利用该模型研究了秸秆还田量与氮肥施用量对N_2O排放的耦合效应。结果表明,秸秆还田与氮肥均有利于N_2O排放,玉米大喇叭口期与乳熟期增量最大;秸秆还田量和氮肥对N_2O排放的影响有叠加作用,且随着施氮量的增加,叠加作用越明显;在高氮条件(N30)下每增加1 875 kg/hm^{2}的秸秆还田量,N_2O排放量增加0.033 kg/hm^2,是低氮条件下的1.65倍。本研究结果可为黄淮海地区夏玉米农田N_2O减排措施的研究提供科学依据,具有一定的实践价值。

关键词:DNDC模型;N_2O;秸秆还田;氮肥;夏玉米

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Study on Coupling Effect of Straw Returning and Nitrogen Fertilizer on N_2O Emission in Summer Corn Field Based on DNDC Model

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Abstract N_2O is an important greenhouse gas. Farmland has become its main source. To make clear the coupling effect of straw returning and nitrogen fertilizer on N_2O emission in summer corn farmland, the DNDC model was verified according to the comparison of N_2O measured data and simulated data, and the coupling effect was studied. The results showed that straw returning and nitrogen fertilizer both had active effect on N_2O emission especially at bellmouthed and milk stages. Straw returning and nitrogen fertilizer also had a superimposition effect on N_2O emission, and the effect increased along with the increase of nitrogen application amount. Under the condition of high nitrogen (N30), for every increase of 1 875 kg/hm^2 straw returning, the N_2O emission increased by 0.033 kg/hm^2 on the average, which was 1.65 times that under low nitrogen condition. This study provided scientific bases for the study of N_2O emission reduction measures in summer corn field of Huanghuaihai region, which had certain practical value.

Key words DNDC model; N_2O; Straw returning; Nitrogen fertilizer; Summer corn

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Research on Production Conditions of High-grade Wine from Northern Blueberry

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Abstract In order to develop the best brewing condition for high-grade fruit wine from northern blueberry, on the basis of reviewing literatures about blueberry wine manufacturing process, this study put forward a new technological process, including raw materials-juicing-addition of auxiliary materials including pectinase-primary fermentation with yeast-post-fermentation-hot and cold treatment-sterilization-packaging-finished product, and sensory indexes, physicochemical indexes and hygienic indexes of the product were inspected according to corresponding national standards and industry standards. The results showed that for northern blueberry pulp, the optimal addition amount of yeast was 1.1 g/L, the fermentation temperature was 22 ℃, and the addition amounts of pectinase and sulfurous acid were 0.3 ml/kg and 100 ppm, respectively; the alcohol degree of the finished product was adjusted to 15.6°; and alternated cold and heat treatment used instead of conventional clarifying agent for removing colloid-like impurities resulted in the brewed product with good wine fragrance, taste and color.

Key words Blueberry; Juicing; Pectinase; Yeast; Primary fermentation; Post-fermentation; Cold and heat treatment

Blueberry has a scientific name of *Vaccinium spp.*, and the main variety cultivated and processed in China currently is originated from North America with good quality, sour and sweet tastes, and rich types, and suitable for wide region [1–2]. Blueberry processing is mainly conducted in the areas of Daxing'an and Xiaoxing'an Mountains and Changbai Mountains[3]. Blueberry processing is a resource-dependent type industry, the annual production of wild blueberry in whole China is less than the processing amount of one company, which is also the main reason for smaller scale of blueberry deep processing enterprises, and therefore, more fresh fruit is imported into domestic market in winter when no fruit is produced, mainly from Chile and Argentina [4–5]. The processing and brewing of blueberry wine is developing rapidly though it is started later than fruit juice. The brewing process of wine requires more fruit as raw materials, so that blueberry wine has higher nutritional components, as well as certain nourishing and health-caring effects[7–8]. However, there are few brands and little quantity, which could not satisfy increasing material and cultural needs of the people, and especially, there have been fewer studies on processing conditions of blueberry wine [9–11]. In view of this, this study investigated the processing conditions of high-grade blueberry wine taking the situation of blueberry planting industry in Taian into consideration with the support of College Students' Innovative Planning Project of Tiaan City.

Materials and Methods

Materials

Following materials were used: organic blueberry purchased from Shunten Agriculture and Forestry Technology Development Co., Ltd.; Y-2-3 type liquid pectin purchased from

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北方蓝莓高档果酒制作工艺条件的研究

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摘 要 为探讨北方蓝莓高档果酒最佳的酿造工艺, 在查阅了蓝莓酒制造工艺文献的基础上, 提出了新的工艺流程, 包括原料-榨汁-加入果胶酶等辅料-主发酵过程加入酵母-后发酵-冷热变温处理-杀菌-包装-成品, 并依据国家标准和行业标准对产品的感官指标, 理化指标, 卫生指标进行检验, 结果表明, 北方蓝莓果浆最佳酵母添加量为 1.1 g/L, 发酵温度 22 ℃, 果胶酶添加量为 0.3 ml/kg, 亚硫酸 100 ppm, 酒成品的酒精度调至 15.6°, 冷热交替代替了过去的用澄清剂去除胶体杂质, 酿造成品酒醇香、口感好、色泽好。

关键词 蓝莓; 榨汁; 果胶酶; 酵母; 主发酵; 后发酵; 冷热处理

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Effects of Seaweed Bio-organic Fertilizer on Growth and Yield of Winter Wheat

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Abstract **[Objective]** This study aimed to investigate the effects of seaweed bio-organic fertilizer on yield and quality of winter wheat. **[Method]** Seaweed bio-organic fertilizer was applied to leaves of winter wheat according to the dose of 45 kg/hm^2 from jointing stage to maturing stage, and plant height, dry matter accumulation, flag leaf photosynthetic characteristics and grain yield of winter wheat were investigated. **[Result]** Foliar spraying of seaweed bio-organic fertilizer showed little effect on plant height of winter wheat, thickened stems, promoted dry matter accumulation, increased flag leaf photosynthetic rate by 3.16%, and increased yield of winter wheat by 6.85%. **[Conclusion]** Foliar spraying of seaweed bio-organic fertilizer promoted the intelligent growth, thickened the stems, improved the lodging resistance, significantly increased the panicle weight per plant, and increased the bulk density of winter wheat, as well as improving the physical quality of wheat grain. In addition, foliar spraying of seaweed bio-organic fertilizer promoted the synthesis of chlorophyll and mitigated the decomposition of chlorophyll in winter wheat. Under the background of fertilizer-pesticide double reduction, the test results and data of this study can be promoted in the wheat-growing areas of Shandong Province and even whole China.

Key words Seaweed bio-organic fertilizer; Winter wheat; Dry matter accumulation; Yield; Photosynthetic rate

Wheat is one of the world´s major food crops, and is the staple food of more than 1/3 of the world´s population. Stabilizing production area and improving yield, quality and efficiency are the development trends of wheat production in the world[1]. The annual planting area of wheat in Shandong Province is 5.20×10^6 hm^2, and it is the most important food crop in Shandong Province. There are many problems in wheat production process. Excessive application of chemical fertilizers and pesticides leads to wheat farmland production, and restricts the sustainable development of wheat production. Currently, the average application amount of fertilizer in China´s land exceeds 400 kg/hm^2, which is much higher than the standard of 225 kg/hm^2 adopted by United States and the European[2]. Fertilizer residue in the soil is quite serious. Acid rain, sulfur and heavy metals produce pollution to the soil, crop and water, resulting in pesticide toxicity in biological products. In Shandong Province in 2012, the annual application amount of chemical fertilizer was 4.74×10^6 t, which was higher than the national average, and the effective irrigation area was 4.99×10^6 hm^2[3]. To address the negative effects of chemical fertilizers on agricultural products, the application amount of chemical fertilizers must be reduced, and the application amount of microbial fertilizers and bio-organic fertilizers should be increased to fundamentally solve the pollution of chemical fertilizers and to

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海藻生物有机肥对冬小麦的生长和产量的影响

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摘 要 [目的]探讨海藻生物有机肥对冬小麦的产量和品质造成的影响程度。[方法]采取大田小区试验方法,在冬小麦返青期后拔节-成熟期,叶面喷施海藻生物有机肥3次,共45 kg/hm^2,调查冬小麦的株高、干物质积累、旗叶光合特性和籽粒产量等。[结果]喷施海藻生物有机肥对冬小麦的株高影响较小,促进茎秆粗壮、干物质积累量增加,旗叶光合速率增加了3.16%,小麦产量增加6.85%。[结论]叶面喷施海藻生物有机肥可促进冬小麦智能化生长,增粗抗倒伏,显著提高了单株穗重的同时,增加了容重,提高小麦籽粒的物理品质;有利于叶片叶绿素的合成和缓解叶绿素的分解。在目前国家提出的化肥和农药双减战略形势下,该试验结果和数据可在全省和全国小麦种植区推广应用。

关键词 海藻生物有机肥;冬小麦;干物质积累;产量;光合速率

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不同耕作方式与氮肥类型对夏玉米光合性能的影响

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摘 要: 为了探讨不同耕作方式与氮肥类型对夏玉米光合性能的影响及其作用机制, 本试验以玉米杂交种郑单 958 为供试材料, 通过快速叶绿素荧光诱导动力学曲线(OJIP)及 820 nm 光吸收等技术, 深入研究了玉米花后叶片叶绿素含量、含氮量、气体交换参数、光系统 II (PSII)、光系统 I (PSI)及二者间的协调性。两年研究结果表明, 与常规尿素相比, 施用控释尿素均可显著提高玉米花后穗位叶叶绿素含量、净光合速率(P_n)及后期气孔导度(G_s), 明显改善光系统间协调性。与旋耕相比, 深松可进一步加强施氮对玉米叶片光合性能的促进作用。控释尿素结合深松可显著提高叶绿素含量及P_n, 明显改善叶片 PSII 反应中心供体侧和受体侧性能, 增强电子由 PSII 向 PSI 的传递, 使花后叶片 PSII 与 PSI 间协调性显著增加, 有利于产量形成期光合性能稳定。光合性能的提高显著增加了穗粒数和单株籽粒产量, 最终提高玉米产量。因此, 深松与控释尿素结合可有效地协调 PSII 与 PSI, 提高夏玉米光合性能, 促进玉米增产。

关键词: 深松; 控释尿素; 光合性能; 光系统; PSI-PSII 协调性

Effects of Tillage Methods and Nitrogen Fertilizer Types on Photosynthetic Performance of Summer Maize

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Abstract: In order to understand the effects of different tillage methods and nitrogen fertilizer types on photosynthetic performances and their mechanism, the characteristics of photosystem II (PSII), photosystem I (PSI) and the coordination between them in ear leaves of maize (cultivar, Zhengdan 958) were studied by using fast chlorophyll fluorescence-induction kinetics and 820 nm light-absorption curves. Two-year field experiment indicated that, compared with normal urea, controlled-release urea significantly increased the chlorophyll content, net photosynthetic rate (P_n) and stomatal conductance (G_s) of ear leaves after anthesis, and significantly improved the coordination between PSII and PSI. Compared with rotary tillage, subsoiling significantly increased the photosynthetic performances in ear leaves during reproductive stage. Subsoiling method combined with controlled-release urea application could significantly increase chlorophyll content, improve the performances of electron donor and acceptor sides of electron transport chain in PSII reaction center, and enhance the distribution of electron transported from PSII to PSI as well. Consequently, the coordination between PSII and PSI after anthesis was significantly improved, which is conducive to the stability of photosynthetic performances during maize reproductive stage. The improvement of photosynthetic performances

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2016年山东省玉米种植成本收益分析

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摘　要：基于相关调研，分析了2016年山东省小农经营与规模经营种植玉米的成本与收益情况，指出在不考虑土地成本的前提下，玉米种植的规模经营比小农经营有更多收益空间；当前玉米等粮食收储价格下跌的背景下，土地成本限制了玉米规模化种植的发展。同时，提出通过提高机械化水平、应用先进适用技术、调整玉米品种的种植结构、完善相关政策等措施来促进山东省玉米种植节本增效。

关键词：玉米种植；成本收益分析；机械化；适度规模经营；山东省

Cost-benefits Analysis of Maize Planting in Shandong Province in 2016

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Abstract: Based on related researches, the authors analyzed maize planting cost-benefits situation of petty-farmer management and scale management in Shandong province in 2016, and pointed out that the scale management of maize planting had more income space than petty-farmer management without considering the land cost, while the land cost limited the development of large-scale maize planting under the background of the purchasing and storage prices of maize and other grain falling down. At the same time, the authors also put forward some suggestions on promoting cost saving and efficiency increase of maize planting in Shandong province, such as raising the level of mechanization, using advanced and applicable technology, adjusting planting structure of maize varieties, improving relevant policies, and other measures.

Key words: maize planting; cost-benefits analysis; mechanization; appropriate scale management; Shandong province

0　引言

玉米种植收益的高低对农户种植玉米的积极性有较大影响，也是玉米种植业能否持续发展的基础。许多学者都对玉米生产的成本和收益做过相关研究，如，陈甜等研究了1998—2009年山东省玉米种植成本收益变动趋势，认为山东省玉米种植总成本增幅呈显著扩大趋势，但其净利润呈现更为快速的增长趋势[1]；范少玲对中美玉米种植成本与收益进行了比较研究，认为玉米价格和单产对我国玉米种植净利润产生了正向影响，而肥料费、其他物资服务费、人工成本和土地成本则阻碍了我国玉米种植收益的增加[2]。但这些研究大多建立在高玉米收储价格的前提下，一些结论已与当前玉米生产情况不相契合。2016年以来，玉米收储价格大幅降低，与玉米生产成本息息相关的生产资料价格、人工成本也发生了较大变化。同时，2016年我国正式提出在东北三省及内蒙古地区实行玉米“市场化收购”加“补贴”新机制，要求玉米价格

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根瘤菌、超声和 2,4-D 处理对小麦幼苗生长的影响

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摘 要: 以小麦种子和幼苗作为试材，通过超声、根瘤菌制剂处理小麦种子探究不同处理方式对于小麦根长生长发育的影响，通过 2,4-D、根瘤菌制剂处理小麦幼苗探究不同处理方式对于小麦幼苗叶绿素等生物指标的影响。结果表明：单纯超声处理和超声与根瘤菌制剂复合处理对小麦根长都有显著的促进作用；单纯根瘤菌制剂处理和 2,4-D 与根瘤菌制剂复合处理对小麦幼苗叶绿素含量的提升和植株生长态势都有着显著促进作用。

关键词: 根瘤菌; 超声; 2,4-D; 小麦
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The Influence of Rhizobium, Ultrasonic and 2,4-D Treatment on Wheat Seedling Growth

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Abstract: Using wheat seeds and seedlings as test materials, the effects of different treatments on the growth and development of wheat roots were investigated by ultrasonic and rhizobium treatment. The effects of different treatments on chlorophyll and other biological indexes of wheat seedlings were investigated by using 2C4-D and rhizobium treatment. The results showed that the ultrasonic and ultrasonic combined rhizobium treatment has a significant effect on wheat root length. The treatment of pure rhizobia and the combination of 24-D and rhizobia could promote the chlorophyll content and wheat seedlings growth.

Keywords: Rhizobium; ultrasonic; 2,4-D; wheat

根瘤菌对双子叶植物生长的促进作用的研究成果颇丰，而许多重要的粮食作物为单子叶植物，如何将根瘤菌利用在单子叶植物上是一个非常有意义的问题，诸多研究人员在这方面做了大量的探索。提升粮食作物的种子活力对于农业生产有重要的意义，一些物理方法如超声波处理可以提高绿豆和掌叶大黄种子的发芽率[1,2]，通过超声波和赤霉素结合处理川西獐牙菜种子可以提高种子萌发能力[3]，说明通过多种因素共同处理植物种子可以影响种子活力及植物的发育。

植物生长状况优良与否很大程度上依赖于根系对土壤养分和水分的吸收能力。土壤中的养分需要经过根系的吸收转化才能进入植物体内，为植物所利用，而植物吸收营养元素能力的强弱很大程度上取决于根系构型。如何改变植物根系构型，或间接增加其生物产量成为促进植物生长的有效手段。超声处理对水稻对其延展根系或提高其营养器官质量等具有显著促进作用[4,5]。超声处理小麦种子可以提高其萌发率[6,7]。根瘤菌在禾本科植物中的定殖和促进作用[8]，以及水稻内生菌的发现[9]，促进了非豆科植物固氮的研究[10-14]。产量的增加、固氮机理和微生物与植物共同作用等方面的研究的进一步的深入，使人们提高了对植物与微生物的相互协调生态圈的认知。本研究利用超声和根瘤菌单独或组合处理小麦种子，以及根瘤菌和 2,4-D 单独或组合处理小麦幼苗，探讨不同因素对小麦幼苗的促进作用，以期为小麦种子处理提供借鉴。

1 材料与方法

1.1 试验材料

山融 3 号（由山东大学夏光敏教授实验室提供）；根瘤菌剂（购买于克劳沃公司）。

1.2 试验设计及测定指标

1.2.1 *超声和根瘤菌处理吸胀种子* 取饱满的山融 3 号小麦种子 200 粒，进行种子表面消毒，暗处萌

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适期晚播对不同密度冬小麦产量和茎秆抗倒性能的调控效应

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摘 要:本试验在宽幅播种条件下,以大穗型冬小麦品种'泰农18'为材料,设计常规播期10月10日和适期晚播10月20日2个播期处理,各播期下分别设置225、375、525万株/hm^2 3个种植密度,研究了播期和密度互作对冬小麦产量及产量形成、茎秆重心高度、基部节间机械强度和茎秆抗倒指数的影响,以期明确适期晚播对密植高产冬小麦产量和茎秆抗倒性能的调控效应。研究结果表明:播期和种植密度显著影响冬小麦的产量和抗倒性能。各播期间适度密植(375万株/hm^2)可通过提高穗数获得高产,且在两播期间无显著差异。适度晚播显著提高各密度冬小麦抗倒指数;在晚播条件下,随种植密度增加,小麦重心高度提高的幅度、基部节间机械强度降低的幅度显著低于常规播期,从而使得适期晚播密植小麦的抗倒能力仍可维持在较高水平。适度密植小麦可通过适期晚播平衡其产量和抗倒能力,本试验条件下,采用375万株/hm^{2}的种植密度,在10月20日播种,可协同实现高产、稳产。

关键词:晚播;种植密度;冬小麦;产量;抗倒性能
中图分类号:S512.11 **文献标志码**:A **论文编号**:casb18070043

Effect of Late Sowing on Lodging Resistance and Grain Yield of Winter Wheat at Different Densities

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Abstract: Regular sowing date (October 10) and moderate late sowing date (October 20) were designed in this study using a large-spike winter wheat variety ("Tainong 18") as the experimental material. Furthermore, on each sowing date, three planting densities were designed at 2.25, 3.75 and 5.25 million plants/hm^2 respectively. The interactive effects of sowing date and planting density and their interactions on the grain yield, culm height at the center of gravity (CHCG), culm mechanical strength of the second base internode (CMS) and the culm lodging resistance index (CLRI) of winter wheat were studied to clarify the regulation of late sowing on grain yield and culm lodging resistance of high-yield winter wheat. The results showed that the interactive effect of sowing date and planting density significantly influenced the yield and lodging resistance of winter wheat. Properly increasing planting density to 3.75 million plants/hm^2 could achieve high yield by improving the number of spikes during each sowing period, and there was no significant difference between the two sowing dates. Moderate late sowing increased the CLRI of the winter wheat enormously in various planting densities. Under the late sowing condition, with the increase of planting density, the improvement of the CHCG and the reduction of the CMS were obviously lower than those of the conventional sowing date. Therefore, the

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氮肥水平对强筋小麦产量和氮素利用的影响

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摘 要:为明确强筋小麦产量与效率相协同的最优施氮量,试验选用'济麦20'和'洲元9369'2个优质强筋小麦品种为试验材料,设置0、120、180、240、300 kg N/hm^{2}等5个施氮水平,用烘干法和凯氏定氮法分别测定小麦成熟期干物质量积累和含氮量,用以计算小麦氮素积累及氮素利用相关指标。结果表明,随氮肥投入量的增加,小麦产量呈现先升高后降低的变化趋势,其中'济麦20'在N180和N240下达最高产量7.28 t/hm^{2}和7.26 t/hm^2,其较高的产量主要源于相对平衡的产量构成因素以及较高的干物质积累量(平均18.54 t/hm^2);'洲元9369'在N180下产量最高达7.75 t/hm^2,其较高的产量主要源于较高的单位面积穗数(970.65万/hm^2)、穗粒数(30.83粒)、较高的干物质积累量(20.77 t/hm^2)和收获指数(37.33%)。虽然氮肥偏生产力随着氮肥施用量的增加逐渐下降,但两品种的氮肥回收效率、氮肥农学利用率和氮肥生理利用效率均可在N180条件下达到最高值,其中,'济麦20'最高值分别为62.67%、5.71 kg/kg、9.11 kg/kg,'洲元9369'的最高值分别为63.65%、7.33 kg/kg、11.55 kg/kg。综合产量水平和氮素利用相关指标,本区域强筋小麦生产中产量与氮素利用效率相协同的施氮量为180 kg/hm^2。

关键词:强筋小麦;产量;氮素利用;氮肥水平;产量效率协同
中图分类号:S512.1+1 **文献标志码**:A **论文编号**:casb18090100

Nitrogen Fertilizer Level: Effects on Yield and Nitrogen Utilization of Strong Gluten Wheat

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Abstract: To clarify the optimal nitrogen fertilizer application which could simultaneously achieve high grain yield and high nitrogen use efficiency (*NUE*), the two winter wheat cultivars with strong gluten, 'Jimai 20' and 'Zhouyuan 9369', were used as materials. Five nitrogen levels included 0, 120, 180, 240, 300 kg N/hm^2 were set. The dry matter accumulation and nitrogen content were measured through drying method and Kjeldahl method, and the relevant *NUE* indices were calculated subsequently. The results showed that with the increase of the nitrogen input, the yield increased first and then declined. 'Jimai 20' had the highest yield at N180 (7.28 t/hm^2) and N240 (7.26 t/hm^2), mainly due to the balanced yield components and higher dry matter accumulation (averaged at 18.54 t/hm^2). While 'Zhouyuan 9369' reached the highest yield (7.75 t/hm^2) at N180, mainly resulting from higher spikes per unit area (970.65×10^4/hm^2), kernels per spike (30.83 kernels/spike), dry matter accumulation (20.77 t/hm^2) and harvest index (37.33%). Although the *PFP*、markedly reduced along with the improved nitrogen level, the *RE*、*AE*、and *PE*、reached the highest value (62.67%, 5.71 kg/kg, 9.11 kg/kg for 'Jimai 20' and 63.65%, 7.33 kg/kg, 11.55 kg/kg for 'Zhouyuan 9369',

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Multi-environments and multi-models association mapping identified candidate genes of lint percentage and seed index in *Gossypium hirsutum* L.

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Abstract Upland cotton (*Gossypium hirsutum* L.) accounts most of the natural fiber production worldwide. Lint percentage (LP) and seed index (SI) are important components of cotton fiber yield, which is a constant breeding goal of cotton. So, the loci underpinning LP and SI should be extensively dissected. Here, one single-locus and four multi-locus genome-wide association study (GWAS) models were employed to detect candidate loci for lint percentage and seed index under seven environments with 196 upland cotton accessions and 41,815 single nucleotide polymorphism (SNP) markers. Totally, 39 and 45 significant quantitative trait locus (QTL) were identified in at least two environments or two models, including 24 previously reported QTLs and six pleiotropic QTLs. Referred to the genome and gene expression database of TM-1, 614 candidate genes were detected for lint percentage and seed index, including 103 genes preferentially expressed in fiber or ovule. The gene Gh_A10G0378, functioned in potassium ion transport, was considered to be related to lint percentage. Collectively, the associated markers and promising genes detected herein will help to elucidate the genetic architecture of lint percentage and facilitate fiber yield improvement in cotton.

Keywords Genome-wide association study · Single-locus and multi-locus models · SNP · Lint percentage · Seed index · Cotton

Huixian Xing, Yanchao Yuan and Haijun Zhang contributed equally to this work.

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Introduction

Upland cotton (*Gossypium hirsutum* L.) is an important natural fiber and cash crop around the world, accounting for 95% of total fiber production (Fang et al. 2017). High fiber yield is a constant theme of cotton breeding and cultivation. Lint percentage (LP, the ratio of fiber weight to seed cotton weight) and seed index (SI, the weight of 100 seeds) are important components of fiber yield. And LP and SI, which are closely related to fiber yield improvement, are also the critical economic indexes for cotton cultivar (Du et al. 2018; Huang et al. 2017; Song et al. 2019). So, dissecting genetic variation and genes underlying LP and SI is therefore essential.

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白花丹参和紫花丹参叶绿素荧光日变化比较研究

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摘要：为从叶绿素荧光的角度阐明紫花丹参产量高于白花丹参的原因，探讨紫花丹参和白花丹参产量和叶绿素荧光差异内在关系，在山东农业大学农学实验站，利用便携脉冲调制式荧光仪在丹参快速生长期测定叶片的叶绿素荧光参数，并对其荧光特性进行综合评价。结果表明，紫花丹参的最大荧光、PSII 最大光化学效率、潜在光化学效率、实际光化学效率、PSII 光化学猝灭显著高于白花丹参，表明紫花丹参光合电子传递效率、光合能力高于白花丹参。由此推测，紫花丹参产量高于白花丹参的重要原因是紫花丹参对光能的利用率高于白花丹参。

关键词：丹参；叶绿素荧光；光合性能
中图分类号：S567.5+3.05 **文献标识码：**A **文章编号：**1001-4721（2019）01-0008-04

Comparisons of Chloropyll Fluorescence Parameters of *Salvia Miltiorrhiza* Bunge and *Salvia Miltiorrhiza* Bunge var alba C.Y.

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Abstract: To clarify *Salvia miltiorrhiza* Bunge from the perspective of chlorophyll fluorescence yield higher than *S.miltiorrhiza* Bunge var alba C.Y, explore *S.miltiorrhiza* Bunge , *S.miltiorrhiza* Bunge var alba C.Y. and chlorophyll fluorescence differences inherent relationship, in the shandong agricultural university agronomy experiment stations use portable pulse modulation fluorometer in *S.miltiorrhiza* growing up fast determination of chlorophyll fluorescence parameters of leaves, and the comprehensive evaluation of fluorescence properties.Results show that *S.miltiorrhiza* Bunge fluorescence,the largest PSII photochemical efficiency, the largest potential photochemical efficiency, the actual photochemical efficiency, PSII photochemical quenching is significantly higher than *S.miltiorrhiza* Bunge var alba C.Y., showed that *S.miltiorrhiza* Bunge photosynthetic electron transport efficiency, photosynthetic capacity is higher than *S.miltiorrhiza* Bunge var alba C.Y.. Therefore, it was speculated that the important reason for the higher yield of radix *S.miltiorrhiza* was that the utilization rate of light energy of *S.miltiorrhiza* Bunge was higher than that of *S.miltiorrhiza* Bunge var alba C.Y..

Key words: *Salvia.miltiorrhiza* Bunge;Chlorophyll fluorescence ;photosynthetic performance

丹参（*Salvia miltiorrhiza* Bunge）根入药，是治疗心血管疾病的常用中药。丹参广泛用于治疗冠心病、高血脂、高血压、心肌梗塞、脑梗塞、脉管炎等疾病，疗效显著，因此丹参市场需求量不断增加。为提高丹参的产量和品质，研究人员对丹参栽培技术进行深入研究。作物的光合性能与其生长和生产力水平密切相关，而叶绿素荧光与光合作用紧密相关，植物的叶绿素荧光参数反映植物光合作用与环境之间的关系，能灵敏地反映植物光合生理特性，具有“内在性”的特点[1]。植物光合能力的差异可以通过一系列光合和荧光指标综合反映[2]。贺立红等[3]研究银杏荧光变化发现，品种间叶绿素荧光参数存在显著差异。金银花[4]、厚朴[5]、女贞[6]等药用植物同样存在不同品种间叶绿素荧光参数间的差异，并将研究结果用于高光效药用植物品种

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间作大葱对桔梗根系分泌物的影响

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摘要:为探究间作大葱有效消减桔梗连作障碍的机理,本试验选取桔梗连作5年的地块进行研究,设置桔梗单作、大葱单作和桔梗大葱间作行比3:2、4:2、5:2共5个处理,利用原位取土法收集不同处理的根际土,并进行根系分泌物的提取与GC-MS分析。结果表明,单作桔梗和桔梗大葱间作根系分泌物中2,2′-亚甲基双-(4-甲基-6-叔丁基苯酚)相对含量高达23.51%~39.63%,而且桔梗大葱间作根系分泌物中该物质相对含量显著高于单作桔梗和单作大葱;间作大葱提高桔梗根系分泌物中角鲨烯、β-谷甾醇、岩皂甾醇等抗胁迫物质和苯并噻唑、2-甲硫基苯并噻唑这类杀菌抑菌物质的相对含量,同时提高桔梗根系分泌物中喹啉、二苄胺、N-(1,3-二甲基丁基)-N′-苯基对苯二胺等碱性物质的相对含量,降低三十六烷、N,N-二甲基十二酰胺、苯乙烯、十五烷、十七烷相对含量,有效改善间作土壤微环境。综上所述,间作大葱改变了间作体系中桔梗根系分泌物的组成成分和相对含量,增加了桔梗根系分泌物中生长促进型化感物质的相对含量,降低抑制型化感物质的相对含量,改善间作桔梗根际微环境,有效消减桔梗连作障碍。

关键词:桔梗;大葱;间作;单作;根系分泌物

中图分类号:S344.2 **文献标识号:**A **文章编号:**1001-4942(2019)11-0068-06

Effects of Intercropping *Allium fistulosum* on Root Exudates of *Platycodon grandiflorum*

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Abstract To clarify the alleviating mechanism of continuous cropping obstacle of *Platycodon grandiflorum* intercropped with *Allium fistulosum*, the plot of continuous cropping for 5 years was selected in this experiment. There were five treatments: *P. grandiflorum* monoculture, *A. fistulosum* monoculture, and intercropping of *P. grandiflorum* with *A. fistulosum* at row ratio of 3:2, 4:2 and 5:2. The rhizosphere soil of different treatments was collected by *in situ* soil sampling method, and the root exudates were extracted and analyzed by GC-MS. The results showed that the relative content of 2, 2′-methylene bis-(4-methyl-6-tert-butylphenol) in root exudates of *P. grandiflorum* was 23.51%~39.63%, and that was significantly higher in the intercropped *P. grandiflorum* than that in monoculture. The relative contents of squalene, beta-sitosterol, saponin sterol, benzothiazole and 2-methylthiobenzothiazole in intercropped *P. grandiflorum* increased, and the relative contents of quinoline, dibenzylamine, N-(1,3-dimethylbutyl)-N′-phenylp-phenylene-diamine also increased, while the relative contents of hexatriacontane, N, N-dimethyldodecamide, styrene, pentadecane and heptadecane decreased, which effectively changed the soil microenvironment of intercrop-

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研究报告
Research Report

一个新的丹参 CY*P450* 的克隆和生物信息学分析

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摘 要 从丹参(*Salvia miltiorrhiza* Bunge)中克隆 1 个新细胞色素 *P450* 基因 *SmCYP71D411.1*，采用生物信息学的方法，对该基因进行序列特征和组织表达特性分析。提取丹参根部总 RNA 后反转录为 cDNA，克隆得到 *SmCYP71D411.1* 基因全长，构建克隆重组质粒。利用生物信息分析预测蛋白质性质、结构。并利用实时荧光定量 PCR 进行了组织表达特异性分析。*SmCYP71D411.1* cDNA 全长 1 539 bp，编码 498 个氨基酸，相对分子质量 56 541.42 Da，理论等电点 7.67。亚细胞定位于叶绿体类囊体膜、细胞质、叶绿体基质等。*SmCYP71D411.1* 在根周皮中的表达量最高。本研究克隆得到一个新基因 *SmCYP71D411.1*，并进行了生物信息学分析，为进一步验证该基因的功能提供了一定的基础。

关键词 丹参(*Salvia miltiorrhiza*), 细胞色素 *P450* 基因, 基因克隆, 生物信息学分析, 荧光定量 PCR

Cloning and Bioinformatics Analysis of a New *Salvia miltiorrhiza* CY*P450*

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Abstract To clone a new cytochrome *P450* gene *SmCYP71D411.1* from *Salvia miltiorrhiza* Bunge and analyze the sequence and tissue expression characteristics of the gene using bioinformatics. After extracting the total RNA from *Salvia miltiorrhiza* root, it was reversed to cDNA, and the total length of the *SmCYP71D411.1* gene was obtained through transcription group database screening. The open reading box and translated amino acid sequence were obtained using NCBI ORF finder. The primer was designed for PCR amplification and the Kron recombinant plasmid was constructed. Using biological information analysis to predict protein properties and structures. Tissue expression specificity was analyzed by fluorescence quantitative PCR. The *SmCYP71D411.1* cDNA obtained by cloning had a total length of 1 539 bp, encoding 498 amino acids, relative molecular mass of 56 541.42 Da, and theoretical isoelectric point of 7.67. Subcellular localization was found in thylakoid membrane, cytoplasm chloroplast matrix of chloroplast, etc. The expression of *SmCYP71D411.1* was highest in the rind. A new gene, *SmCYP71D411.1*, was obtained by cloning, and bioinformatics analysis was carried out, which provides a foundation for further verification of the gene's function.

Keywords *Salvia miltiorrhiza*, Cytochrome *P450* gene, Gene cloning, Bioinformatics analysis, Fluorescent quantitative PCR

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增施磷肥提高弱光环境中夏大豆叶片光合能力及产量

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摘 要: 为研究增施磷肥对弱光环境中夏大豆光合能力的调控作用, 本研究以齐黄 34 为供试品种, 设置全生育期正常光照(L1)、花后弱光(L2) 2 个光照处理, 不施磷肥(P0)、常规施磷(P1)、增施磷肥(P2) 3 个磷肥处理, 通过测定叶片气体交换和叶绿素荧光, 系统分析了花后叶片光合能力和产量要素的变化。2 年结果表明, 花后弱光处理后大豆产量显著降低, 平均产量较正常光照组降低 61.4%。正常光照环境中, P2 比 P0 和 P1 处理的 2 年平均产量分别高 8.4%和 3.2%, 而弱光环境中, P2 较 P0、P1 处理分别增产 21.7%与 12.2%, 表明在弱光环境下增施磷肥增产效果更明显。弱光处理后大豆叶片叶面积、比叶面积和叶绿素 *a*、叶绿素 *b* 含量显著增加, 增施磷肥进一步扩大其增幅, 同时叶片净光合速率与气孔导度明显降低, 胞间 CO_2 浓度变化趋势与之相反, 证明弱光处理后同化能力的降低不是由于气孔限制。增施磷肥会提高光合速率和气孔导度, 在弱光条件下效果更显著。增施磷肥会显著降低叶片叶绿素荧光诱导曲线中的 J 点和 K 点相对荧光, 提高叶片光系统 II 电子传递性能, 在弱光环境下作用比正常照光下更明显。弱光环境下增施磷肥可提升叶片光合电子传递活性, 缓解弱光下叶片光合速率降低, 提高大豆植株干物质积累, 进而提高产量。

关键词: 大豆; 磷肥; 弱光; 光合能力; 产量

Increasing phosphate fertilizer application to improve photosynthetic capacity and yield of summer soybean in weak light environment

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Abstract: In order to study the effect of phosphate fertilizer application on the photosynthetic characteristics of summer soybean in weak light environment, two light treatments [normal light (L1) and weak light (L2)] with three phosphate fertilizer treatments including non-phosphate fertilizer application (P0), conventional phosphate fertilizer application (P1), and excessive-phosphate fertilizer application (P2) in each light treatment were set up to measure the gas exchange, chlorophyll a fluorescence differences of photosynthetic performance as well as the yield and its components in Qihuang 34. The yield reduced significantly in weak light treatment, with an average of 61.4% in two years lower than that under the normal light. The 2-year average yield of P2 was 8.4% and 3.2% higher than that of P0 and P1 respectively under the normal light, but 21.7% and 12.2% higher than P0 and P1 in

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Rice

ORIGINAL ARTICLE Open Access

OsHsfB4d Binds the Promoter and Regulates the Expression of *OsHsp18.0-CI* to Resistant Against *Xanthomonas Oryzae*

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Abstract

Background: Bacterial leaf streak (BLS) and bacterial blight (BB) are two major prevalent and devastating rice bacterial diseases caused by the Gram-negative bacteria of *Xanthomonas oryzae* pv. *oryzicola* (Xoc) and *Xanthomonas oryzae* pv. *oryzae* (Xoo), respectively. Previously, we identified a defence-related (*DR*) gene encoding a small heat shock protein, OsHsp18.0-CI, that positively regulates BLS and BB resistance in rice.

Results: To reveal the regulatory mechanism of the *OsHsp18.0-CI* response to Xoc and Xoo, we characterized the class B heat shock factor (Hsf), OsHsfB4d, through transcriptional analysis and a transgenic study. *OsHsfB4d* is upregulated post inoculation by either the Xoc strain RS105 or Xoo strain PXO99a in Zhonghua 11 (wild type, ZH11) as well as in *OsHsp18.0-CI* overexpressing rice plants. Transient expression of *OsHsfB4d* can activate the expression of green fluorescent protein (GFP) and luciferase (Luc) via the *OsHsp18.0-CI* promoter. Rice plants overexpressing *OsHsfB4d* exhibited enhanced resistance to RS105 and PXO99a as well as increased expression of *OsHsp18.0-CI* and pathogenesis-related genes. Furthermore, we found that OsHsfB4d directly binds to a DNA fragment carrying the only perfect heat shock element (HSE) in the promoter of *OsHsp18.0-CI*.

Conclusion: Overall, we reveal that OsHsfB4d, a class B Hsf, acts as a positive regulator of *OsHsp18.0-CI* to mediate BLS and BB resistance in rice.

Keywords: Defense response, Heat shock factor, Heat shock protein, *Oryzae sativa*, *Xanthomonas oryzae*

Background

Rice is an important staple crop worldwide that represents 40% of total grain output and nearly 60% of global food consumption (Sharma et al. 2012). However, it has been shown to suffer more than 70 diseases caused by fungi, bacteria, viruses and nematodes during rice growth (Niño-Liu et al. 2006; Ke et al. 2017). There are two major bacterial diseases, bacterial blight (BB) and bacterial leaf streak (BLS), caused by the gram-negative bacteria *Xanthomonas oryzae* that frequently occur in rice. BB is caused by *Xanthomonas oryzae* pv. *oryzae*, which enters into the rice leaf through hydathodes or wounds and colonizes in the xylem vessels. But BLS is caused by *Xanthomonas oryzae* pv. *oryziaola* (Xoc), which penetrates into the leaf through stomata or wounds and colonizes the intercellular space of leaf tissue and finally results in water-soaked stripe lesions (Niño-Liu et al. 2006; Ke et al. 2017; Ju et al. 2017). Currently, BB is well studied for host resistance. Over 40 major resistance genes and 30 defense-related (DR) genes were identified to control the race-specific or spectrum resistance to Xoo isolates (Ke et al. 2017; Ju et al. 2017). BLS is becoming a major concern due to its high prevalence and seriously affecting the yield and quality of rice production (Xu et al. 2010; Zhang et al. 2015). To date, only the *Xo1* locus, which encodes a putative receptor, has been reported to

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丹参水肥一体化种植对杂草生物量的影响

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摘要:试验探究了膜下水肥一体化种植方式和传统露天大水沟灌种植方式对丹参田间杂草生长的影响。结果表明,丹参水肥一体化种植的杂草生物量、杂草密度明显低于传统种植方式,其中生物量降低51.48%。该种植方式可减轻草害,节本增效。

关键词:丹参;膜下水肥一体化;杂草生物量;杂草密度

中图分类号:S567.5+30.53 **文献标识号**:A **文章编号**:1001-4942(2020)08-0096-04

Influence of Fertigation Integrated Cultivation of *Salvia miltiorrhiza* on Weed Biomass

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Abstract The experiment explored the influence of fertigation integrated cultivation under film and traditional cultivation by open ditch irrigation on the growth of weeds in the field of *Salvia miltiorrhiza*. The results showed that the density and biomass of weed were obviously lower under fertigation integrated cultivation compared with traditional cultivation, and the biomass decreased by 51.48%. Fertigation integrated cultivation could not only reduce the grass damage, but also save the cost and increase the efficiency.

Keywords *Salvia miltiorrhiza*; Fertigation integrated cultivation under film; Biomass of weeds; Weed density

丹参是我国传统大宗中药材,也是我国重要的经济作物,市场需求量大,具有活血调经、祛瘀生新的功效。山东省是我国丹参的主要产地,传统种植常采用大水沟灌方式,造成一定的水肥资源浪费,同时导致杂草丛生,与丹参争夺水、肥、光、热等自然资源[1,2]。在中药栽培过程中,杂草是影响中药材产量、品质及经济效益的重要因素,通常可以通过喷施除草剂防除,但目前丹参田没有可供选择的登记的除草剂产品,且中药材种植对农药化肥的施用有严格规定。降低田间杂草基数和生物量可以显著降低危害程度,减少劳动力投入,提高经济效益。

膜下水肥一体化栽培能减少水肥流失,控制土壤温度和湿度[3],达到增产稳产、提高经济效益的目的[4],同时还可以有效抑制杂草生长[5],在国内外不同作物上得到广泛应用。目前关于丹参膜下水肥一体化栽培技术研究还未见报道。本研究调查分析了大水沟灌和膜下水肥一体化种植方式下的杂草生物量及密度差异,探究丹参水肥一体化对杂草生物量的影响,为丹参种植过程中杂草防控提供依据。

1 材料与方法

1.1 试验材料

丹参为山东省主栽地方品种;地膜采用聚乙烯黑色薄膜;肥料为尿素(含N 46.4%)、过磷酸

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研究报告
Research Report

粗山羊草响应盐胁迫转录组分析

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摘　要　利用转录组测序(RNA-Seq)技术对粗山羊草'AL8/78'在 200 mmol/L NaCl 人工模拟盐胁迫 0 h 和 96 h 进行了转录组分析。结果表明：盐胁迫处理下上调和下调表达的差异表达基因(DEGs)分别为 546 个和 876 个；GO 富集分析发现，DEGs 主要集中在代谢过程、细胞过程、细胞连接、催化活性等生物过程；KEGG 富集分析发现，DEGs 主要富集在苯丙酮生物合成、类黄酮生物合成、植物激素信号转导等信号通路。通过对 DEGs 进行转录因子注释，共注释到 41 个转录因子，主要包括 WRKY、MYB、bHLH、NAC 和 HSP 等类型。利用实时荧光定量 PCR (qRT-PCR)对 DEGs 进行了表达模式验证，发现其与 RNA-Seq 测序结果一致，证明了 RNA-Seq 结果的准确性。本研究为挖掘粗山羊草耐盐基因提供了理论基础。

关键词　粗山羊草(*Aegilops tauschii*), 盐胁迫, 转录组

Transcriptome Analysis of *Aegilops tauschii* in Response to Salt Stress

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Abstract　Transcriptomic analysis of crude goatgrass 'AL8/78' was performed using RNA-Seq technology in 200 mmol/L NaCl, which simulated salt stress for 0 h and 96 h artificially. The results showed that 546 genes were up-regulated and 876 genes were down-regulated under salt stress. GO enrichment analysis showed that DEGs were mainly involved in biological processes like metabolic process, cellular process, cellular junction and catalytic activity. KEGG enrichment analysis showed that DEGs were mainly involved in pathways such as phenylalanine metabolism, flavonoid biosynthesis and plant hormone signal transduction. 41 transcription factors were annotated in all DEGs including WRKY, MYB, bHLH, NAC and HSP. The expression pattern of DEGs were identified by qRT-PCR, which demonstrated the accuracy of RNA-Seq as the change of DEGs were the same between RNA-Seq and qRT-PCR. This research will provide theoretical basis for gene identification of *Aegilops tauschii* resistance to salt stress.

Keywords　*Aegilops tauschii*, Salt stress, Transcriptome

全球气候变化、不合理的化肥施用和不科学的灌溉方法，使全球土壤盐渍化程度越来越严重。中国的盐碱地面积也在逐年上升(范金, 2015)。在盐碱土中生长的植物，水分吸收减弱、生长缓慢；长时间受

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花生种皮颜色研究进展

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摘要:花生(*Arachis hypogaea* L.)种皮由珠被发育而来,分三层,外表皮是一层厚壁细胞,中间层为若干层薄壁细胞,内表皮为一层薄壁细胞。种皮色素物质主要分布在1~2层表皮细胞内。种皮颜色是决定花生商品和保健价值的重要性状。本文主要对花生种皮颜色类型、不同颜色种皮的营养功效、种皮发育进程中的色泽变化和色素沉积、种皮颜色的遗传以及相关基因定位等方面研究进展进行综述,并对其未来研究进行了展望。

关键词:花生;种皮颜色;色素

中图分类号:S565.2 **文献标识号**:A **文章编号**:1001-4942(2022)01-0152-05

Research Progress on Testa Color of Peanut

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Abstract Peanut (*Arachis hypogaea* L.) testa is developed from the pearl, which is divided into three layers. The outer epidermis is a layer of thick-walled cells, the middle layer is a number of thin-walled cells, and the inner epidermis is a layer of thin-walled cells. The pigment substances in the testa are mainly distributed in the 1~2 layers of epidermal cells. Testa color is an important trait that determines the commodity and health value of peanut. The research progresses of peanut testa color types, nutritional efficacy of different colorful testa, color changes and pigment deposition during testa development, inheritance and genetic mapping of testa color were summerized in this paper, and the future research were also prospected.

Keywords Peanut; Testa color; Pigment

花生又名长生果,属豆科植物。花生是我国主要的油料作物和经济作物之一,也是重要的特色出口农产品[1],是促进农业可持续发展的主要农产品之一[2]。随着人民生活水平的提高,花生用作加工食品和外贸出口的比例在逐步增加,人们对特有的种皮色泽和特有的营养成分需求也日益增多[3]。我国花生种质资源丰富,籽仁种皮颜色分白、粉、红、紫、黑等多种类型。种皮色素物质主要分布在1~2层表皮细胞内。花生种皮除了含有多种营养成分外,还富含黄酮素类化合物、原花色素、白藜芦醇、花青素等多种生理活性成分,具有清除自由基、抗癌、降血压等作用。近几年,国内外学者对于花生种皮颜色的形成机制,尤其是色素积累基因的研究已有多次报道。

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授粉后降雨对玉米杂交种结实以及籽粒性状的影响

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摘要:本试验以郑单958为材料,研究授粉后不同间隔时间降雨(用人工喷水模拟降雨)对玉米产量以及籽粒性状的影响。结果表明,授粉后降雨显著降低玉米产量,相较于对照分别降65.82%(0 h)、54.36%(1 h)、42.07%(2 h)、35.75%(6 h)、29.49%(12 h)和12.03%(24 h),显著降低穗长和穗粗。较对照,授粉后降雨果穗结实性降低,百粒重提高,籽粒蛋白质相对含量提高3.4个百分点(0 h),籽粒淀粉相对含量降低3.35个百分点(0 h)。可见授粉后降雨对玉米产量以及籽粒性状产生显著影响,尽管降雨使籽粒蛋白质相对含量升高,但因总结实率、出籽率降低进而造成经济损失。

关键词:授粉后降雨;玉米;果穗结实;产量;籽粒品质

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Influence of Rainfall After Pollination on Seed and Kernel Traits of Maize Hybrid

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Abstract The effects of rainfall with different intervals (simulating rainfall with artificial water spray) after pollination on maize yield and grain traits were studied by using Zhengdan 958 as experimental material. The results showed that rainfall after pollination significantly reduced the yield, ear length and ear diameter of maize hybrid, and the yield was decreased by 65.82% (0 h), 54.36% (1 h), 42.07% (2 h), 35.75% (6 h), 29.49% (12 h) and 12.03% (24 h) compared with the control. Compared with the control, the ear fruiting decreased, the hundred grain weight increased, the relative content of grain protein increased by 3.4 percentage points (0 h), and the relative content of grain starch decreased by 3.35 percentage points (0 h) with rainfall after pollination. It could be seen that rainfall after pollination had significant effects on maize yield and grain traits. Although rainfall increased the relative content of protein, it caused economic losses due to the decrease of seed setting rate and kernel rate.

Keywords Rainfall after pollination; Maize; Ear fruiting; Yield; Grain quality

随着全球气候的变化,极端天气的出现也变得更加频繁,对农业生产产生极大影响。由于受到干旱的影响,秘鲁每周的香蕉出口下降25%[1],美国加州的油梨生产也受到干旱的影

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Germination characteristics of maize seeds with high and low-vigour levels in response to on-farm seed priming

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Abstract

On-farm seed priming, which refers to soaking seeds in water, is a pre-sowing seed enhancement technique in agriculture production. This study aimed to investigate the effects of on-farm priming on the germination performance of maize seeds with different vigour levels. Two maize hybrids and four inbred lines were used as experimental materials. Soaking treatment significantly decreased the final germination percentage of low-vigour seeds and had no beneficial effects on high-vigour seeds. With longer soaking times, the reduction in germination increased and there was a positive, negative or neutral effect of water soaking on the germination index. The responses of seeds to on-farm priming varied among the different hybrid / inbred lines used and depending on the length of the treatment. Reducing imbibition damage by PEG soaking had little impact on the poor germination induced by soaking in water. Removal of the seed pericarp alleviated the negative effects of soaking on germination. This study has demonstrated, for the first time, the differential effects of water soaking on maize seeds with different vigour levels. Our results suggested that all influencing factors, such as such as seed initial physiological quality and the technological process, need to be considered to realize the full potential of priming.

Keywords: germination index, germination percentage, maize, seed priming, seed vigour, soaking treatment

Introduction

Maize (*Zea mays* L.) is one of the most important crops used for human food and animal feed worldwide. High-quality seeds lead to good harvests. Seed vigour, an important index used to evaluate seed quality, determines the rate and uniformity of seed germination and seedling growth (ISTA, 2014). High-vigour seeds that exhibit rapid and uniform seed germination and vigorous field seedling emergence are vital to achieve high crop yields to meet the rising global demand for food (Rosegrant *et al*., 1995; Finch-Savage and Bassel, 2015). Maize is a well-spaced crop and, in the absence of tillering, emergence failure means reduced yield. Thus, high vigour maize seeds are of great significance to

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有机肥施用及合理密植提高黄淮海地区夏大豆光系统性能与籽粒产量

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摘要:【目的】氮素合理投入与作物合理增密种植之间的协调关系被普遍认为是挖掘作物增产潜力的重要措施之一。研究无机有机氮肥配合施用和不同种植密度条件下夏大豆光系统性能及产量差异，进而提出最佳施氮形式和密度组合模式，为黄淮海地区夏大豆的高产高效优质生产提供理论基础及科学依据。**【方法】**田间试验于2018—2020 年在山东农业大学农学实验站进行，供试夏大豆品种为‘齐黄 34’ (QH34)。采用完全随机区组设计，试验设置 4 个密度水平，分别为 90000 株/hm^2 (D1)、120000 株/hm^2 (D2)、150000 株/hm^2 (D3)、180000 株/hm^2 (D4)，D1 仅于 2018 年种植，D4 仅于 2019 和 2020 年种植。设置不施氮肥对照 (N0) 和 3 个等氮量氮肥处理：单施尿素 (U)、单施腐熟鸡粪 (M)、尿素与鸡粪氮各占 50% (UM)。测定各处理夏大豆产量及产量构成因素，花后叶片含氮量，净光合速率 (P_n) 及叶绿素荧光诱导动力学曲线 (OJIP 曲线)，分析大豆功能叶片 (主茎倒四叶) 光系统Ⅱ (PSⅡ) 性能，PSⅡ对单位氮素的利用差异，以及 P_n 和 PSⅡ，P_n/SLN 和单位氮素对 PSⅡ的贡献能力之间的相关性。**【结果】**施氮肥显著提高了大豆产量，且 4 个密度水平下，M 和 UM 处理的大豆产量均显著高于 U 处理，UM 处理在 2018 年 D1、D2、D3 密度的大豆产量均显著高于 M 处理，UM 和 M 处理在 2019 年高密度处理 (D4) 差异不显著，在 2020 年 D2、D3、D4 密度下均无显著差异。从产量分析，密度为 D2 或 D3 更有利于黄淮海地区大豆的生产。在相同密度条件下，施氮肥可以显著提高叶片 P_n、PSⅡ的供体侧 (W_k)、受体侧 (V_j) 和 PSⅡ对光能的吸收 (PI_{ABS})、捕获 (φ_{Po})、能量转化 (φ_{Eo}) 及电子传递活性 (Ψ_o)；有效提高单位氮素对 PSⅡ的贡献能力 (φ_{Po}/SLN、W_k/SLN、V_j/SLN、φ_{Eo}/SLN、Ψ_o/SLN)。2018 年各处理大豆的光合效能表现为 UM > M > U，随着种植年份的增加，UM 和 M 处理之间差异逐渐缩小，到 2020 年时二者之间差异不显著。在同一肥料条件下，密度处理之间光合效能指标无显著差异。P_n 和 PSⅡ，P_n/SLN 和单位氮素对 PSⅡ的贡献能力均呈显著正相关，且施氮肥显著提高了 P_n 和 PSⅡ的相关性，以 UM 处理效果最好。而在相同肥料处理条件下，随着密度的增加，P_n 和 PSⅡ的相关性降低但无显著差异。施氮肥后 PSⅡ性能的改善是 P_n 和大豆产量提高的主要原因。**【结论】**在黄淮海地区，稳定的有机肥投入与中高种植密度的结合更有利于大豆的高产高效优质。夏大豆多年连续种植模式下，可将夏大豆密度提高到 150000 株/hm^2 (D3)，重视有机肥氮的投入，在开始施肥的第 1～2 年，以一半尿素一半鸡粪为佳，之后改为单施腐熟鸡粪即可满足大豆的高产需求。

关键词: 夏大豆；有机无机肥配施；密度；光合特性；光系统Ⅱ性能；氮素；产量

Improving photosynthetic performance and yield of summer soybean by organic fertilizer application and increasing plant density

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中药渣制作平菇栽培种试验

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摘 要 将中药渣与传统食用菌栽培料按不同比例混合,设计不同的配方,并与青岛市常用制作平菇栽培种培养料进行比较,试验揭示中药渣可以部分替代棉籽壳用于平菇栽培种的制作,从而大幅度降低平菇制种成本。采用中药渣60%、玉米芯34%、麸皮6%配方培养基培养平菇栽培种,发菌速度、菌丝长势等均好于棉籽壳94%、麸皮6%的常用配方。

关键词 中药渣 平菇栽培种 配方
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中药行业随着国家中药产业政策的大力扶持和人民健康意识的普遍提升而快速发展,同时药渣问题也越来越突出。据报道,在以消耗中药和天然药物资源性原料进行中药制药、配方颗粒等深加工产业化过程中,全国每年产生的药渣等固体废弃物及副产物高达3500万t。中药渣传统处理方法主要是堆放、焚烧或掩埋,不仅污染环境,而且造成巨大的资源浪费。如何将中药渣资源化利用,使其变废为宝,成为众多科技工作者的研究热点。

中药渣中含有氮、磷、钾、硫、铁、镁、粗蛋白、粗纤维、有机碳及少量的维生素等营养成分。通过对青岛市中药企业药渣的外在形态、残留养分和碳氮比等理化性质分析,认为中药渣适宜制作平菇栽培种的培养基。笔者将中药渣与传统培养料按不同比例混合,设计不同的配方,并与青岛市常用制作平菇栽培种培养料进行比较,探索利用中药渣制作平菇栽培种的适宜配方,为中药渣资源化利用开辟新领域。现将试验结果总结如下。

1 材料与方法

1.1 供试材料

平菇菌株,早秋625引自江苏天达食用菌研究所。该品种是青岛市菇农广泛栽培菌株,属广温型,抗杂菌能力较强。

原种:自制麦粒原种。

主料:棉籽壳(市售)、混合药渣(青岛华钟制药股份有限公司提供)、玉米芯(市售)。辅料:麸皮(市售)。

1.2 供试配方

根据平菇菌丝体生长所需的碳氮比(C/N),设计6种培养基配方(表1),配方①作为对照,每个培养基配方制作10瓶,重复3次。

表 1 平菇栽培种培养基供试配方

配方	棉籽壳/%	药渣/%	玉米芯/%	麸皮/%	C/N
①	94	0	0	6	28.56
②	34	60	0	6	31.32
③	20	54	20	6	36.06
④	0	50	44	6	40.90
⑤	0	60	34	6	33.27
⑥	0	50	50	0	41.56

1.3 试验方法

1.3.1 供试栽培种培养基的制作

选择干净的水泥地面,将湿药渣摊开晒干,待试主料放入干燥箱内烘干,烘干温度设定为60 ℃,时间24 h。各配方的主料和辅料按编号顺序备好,每个配方投料量为7 kg。加水(料水比为1∶1.3)充分搅拌均匀,装入750 mL标准蘑菇瓶内,上紧下松,每瓶装湿料450 g,用直径1 cm的木棒在料中间打孔,用聚丙烯塑料薄膜封口,封口后进行高压灭菌,采用1.47×10^5 Pa的压力灭菌1.5 h。当压力降为常压后,放汽出锅。待蘑菇瓶冷却后,即可进行接种操作。

1.3.2 接种与观测

将适龄麦粒原种倒入无菌皿中,充分混合均匀(保证菌龄在整体上的一致性),每次取15~20粒小麦原种接种到供试蘑菇瓶中,在温度为22 ℃的洁净培养室中发菌培养。期间,从封料面开始,每隔24 h观察测量一次菌丝在瓶中发育伸展距离,记录菌丝

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·生物学与生态学·

海南省永兴岛革螨新纪录
(蜱螨亚纲:中气门目)

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摘要:为全面调查我国海南省三沙市永兴岛的革螨生物多样性,于2017年采集永兴岛周边土壤环境中的革螨。经形态分类学鉴定,海南省革螨新纪录有4种,分别是厉螨科的剑毛帕厉螨[*Stratiolaelaps scimitus* (Womersley, 1956)]、巍山裸厉螨(*Gymnolaelaps weishanensis* Gu & Guo, 1997)、力氏广厉螨(*Cosmolaelaps hrdyi* Samšiňák, 1961)和寄螨科的温氏寄螨(*Parasitus wentinghuani* Ma, 1995),标本保存于山东农业大学系统与应用蜱螨学实验室。

关键词:中气门目;革螨;海南省新纪录;永兴岛;三沙市
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Newly recorded species of gamasid mites in Yongxing Island in Hainan province, China (Acari: Mesostigmata)

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Abstract: In order to investigate the biodiversity of gamasid mites in Yongxing Island of Sansha in Hainan province, China, gamasid mite specimens were collected in the soil around Yongxing Island in 2017. According to morphological taxonomy, there were four newly recorded species of gamasid mites in Hainan province, i.e., *Stratiolaelaps scimitus* (Womersley, 1956), *Gymnolaelaps weishanensis* (Gu & Guo, 1997), *Cosmolaelaps hrdyi* (Samšiňák, 1961), and *Parasitus wentinghuani* (Ma, 1995). The specimens are deposited in Laboratory of Systematic and Applied Acarology, Shandong Agricultural University.

Key words: Mesostigmata; Gamasid mite; Newly recorded species of Hainan province; Yongxing Island; Sansha

曾有人对我国海南省及南海附属岛屿的病媒生物进行过多次调查,最早的报道见于1980年顾以铭和王菊生[1]在现海南省白沙、乐东、保亭和崖县检获寄生革螨8种。20世纪90年代,刘金华等[2]在海南省三亚、通什市检获寄生革螨6种;黄佳亮等[3]在海南省南部山区检获寄生革螨5种;龙芝美等[4]报道了南海地区寄生革螨7种。21世纪初,斯武等[5]在中海石油南海西部基地检获寄生革螨2种;王爱民等[6]随后又在该基地检获寄生革螨2种;2009年田珍灶和金道超[7]在海南省的小黄蝠(*Scotophilus temminckii* Horsfield, 1824)体表发现寄生革螨1新种;林坚贞等[8-14]在中国自由生活革螨调查报告中,先后记录了海南省革螨12种;杨明磊等[15]2016年调查了西沙永兴岛和石岛鼠形动物体外寄生虫,其中革螨占93.55%,但未报道革螨种类信息。截至2016年底,共报道我国海南省及南海附属岛屿的革螨7科13属24种。2017年本研究采集了三沙市永兴岛周边土壤环境中的革螨,经形态分类学鉴定,发现革螨

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李晨雨, 臧传江, 朱少杰, 等. 新烟碱类杀虫剂氟吡呋喃酮的研究开发现状与展望[J]. 农药, 2018, 57(11): 785-788.

新烟碱类杀虫剂氟吡呋喃酮的研究开发现状与展望

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摘要: 常规新烟碱类杀虫剂如吡虫啉、噻虫嗪等对蜜蜂等传粉昆虫存在一定环境风险, 拜耳作物科学公司最新开发的新烟碱类杀虫剂氟吡呋喃酮可有效防治刺吸式害虫, 并对传粉昆虫及其他环境非靶标生物低毒。从氟吡呋喃酮的理化性质、分子结构、作用机制、制剂产品、田间防效、环境生物安全性及其分析方法等方面, 综合阐述了该药剂的研究进展, 并对其进行了研究方向展望, 分析了其应用前景。

关键词: 新烟碱类杀虫剂; 氟吡呋喃酮; 作用机制; 环境安全性
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Research Status and Prospect of Flupyradifurone, a New Neonicotinoid Insecticide

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Abstract: The conventional neonicotinoid insecticides (e.g. imidacloprid and thiamethoxam) have certain environmental risks for the pollinating insects such as bees. Newly developed neonicotinoid insecticide, flupyradifurone, can effectively manage sucking insects and it also has low toxicity to pollinating insects and other non-target creatures in the environment. The physicochemical properties, molecular structure, mode of action, preparation products, field efficacy, environmental security and analysis methods of flupyradifurone are comprehensively elaborated, along with its application prospect.

Key words: neonicotinoid insecticide; flupyradifurone; mode of action; environmental security

随着高毒农药在全球市场的退出, 新烟碱类杀虫剂逐渐成为防治刺吸式害虫、小型鳞翅目和鞘翅目害虫最有效的一类杀虫剂[1]。但近年来, 研究者们发现部分新烟碱类杀虫剂(如吡虫啉、噻虫嗪、噻虫胺、呋虫胺等)对蜜蜂的急性毒性为高毒或剧毒, 且具有亚致死效应; 有效成分及代谢物在花粉和花蜜中残留, 导致蜜蜂种群减少, 存活与繁殖能力降低[2-3]; 此外, 吡虫啉、啶虫脒、烯啶虫胺、噻虫胺、噻虫啉等还对土壤生态系统代表生物蚯蚓高毒[4]。鉴于此, 多个国家和组织对部分新烟碱类杀虫剂的环境风险进行再评价或限制使用, 甚至禁用[5]。拜耳作物科学公司针对部分新烟碱类杀虫剂的高蜂毒问题, 开发了一种新型新烟碱类杀虫剂——氟吡呋喃酮(flupyradifurone), 该药剂可高选择性地作用于多种刺吸式口器害虫, 速效性好、持效期长, 且与常规新烟碱类杀虫剂无交互抗性, 其最突出的特点是对蜜蜂等传粉昆虫低毒[6-7]。现从理化性质、分子结构、作用机制、剂型、田间防效、对蜜蜂安全性及其分析方法等方面综述了氟吡呋喃酮的研究进展, 以期指导该药剂的合理使用。

1 氟吡呋喃酮的理化性质

氟吡呋喃酮, 4-[(6-氯-3-吡啶基甲基)-(2,2-二氟乙基)-氨基]-呋喃-2-(5*H*)-酮, 英文通用名为flupyradifurone, 商品名为Altus和Sivanto Prime。其CAS号为951659-40-8, ChemSpider ID为26050805, 分子式为$C_{12}H_{11}ClF_2N_2O_2$, 分子量288.678道尔顿, 化学结构式见图1a。氟吡呋喃酮纯品为白色至米黄色固体粉末, 几乎无味, 熔点72~74 ℃, 不易燃, 蒸气压为9.1×10^{-4} MPa(20 ℃), 比重1.43。20 ℃下, 氟吡呋喃酮在水中溶解度为3.2 g/L (pH值为4), 3.0 g/L(pH值为7)[8]; 在甲苯中溶解度为3.7 g/L; 易溶于乙酸乙酯和甲醇。其最大紫外吸收波长为259 nm。

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赤霉素对烟草幼苗生长发育及其生理生化特性的影响

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摘要:在霍格兰营养液中添加6个不同浓度(0、0.1、0.2、0.4、0.8、1.6 mg·L^{-1})的植物生长调节剂赤霉素(GA_3),研究其对烟苗生长及生理生化特性的影响。结果表明:随着赤霉素浓度的增加,烟草幼苗的生物量和根系指标呈先增加后降低趋势。具体来看,0.2 mg·L^{-1}处理较其它处理提升根系活力,并显著增加烟苗地上部干重和根干重,增幅分别达到7.26%~127.27%、8.54%~174.73%;而0.8、1.6 mg·L^{-1}赤霉素处理较对照降低烟草幼苗生物量和根系活力。随着赤霉素浓度的增加,烟苗根系各项指标呈先增加后减少趋势,各处理间差异显著:0.2 mg·L^{-1}赤霉素处理较其它处理显著增加烟苗根总长度、根表面积、根体积、根平均直径、根尖数,增幅分别达到4.46%~57.83%、2.34%~51.97%、14.44%~168.35%、14.59%~109.78%、17.73%~121.85%。此外,0.2 mg·L^{-1}赤霉素处理能提高烟苗叶片SOD、POD和CAT活性,降低MDA含量,增加叶片SPAD值,提升光合特性。在该试验条件下,烟草漂浮育苗营养液中添加的赤霉素最佳浓度为0.2 mg·L^{-1}。

关键词:烟草;漂浮育苗;赤霉素;幼苗生长;生理生化

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Effects of Gibberellin on Growth and Development and Physiological and Biochemical Characteristics of Tobacco Seedlings

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Abstract Plant growth regulators gibberellins with six different concentrations (0, 0.1, 0.2, 0.4, 0.8, 1.6 mg·L^{-1}) were added into Hoagland nutrient solution to study their effects on the growth and physiological and biochemical characteristics of tobacco seedlings. The results showed that with the increase of gibberellin concentration, the biomass and root index of tobacco seedlings increased firstly and then decreased. Compared with the other treatments, the treatment with 0.2 mg·L^{-1} gibberellin increased root vitality, and significantly increased shoot dry weight and root dry weight of the tobacco seedlings with the corresponding increments of 7.26%~127.27% and 8.54%~174.73%, respectively. Compared with the control group, 0.8 and 1.6 mg·L^{-1} gibberellin treatments reduced the biomass and root activity of tobacco seedlings. With the

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双孢蘑菇液体培养基筛选及培养条件优化

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摘 要: 目前国内双孢蘑菇生产多采用固体菌种进行栽培,但固体菌种在生产过程中存在生长周期长、效率低等问题。因此,本文采用正交试验方法,以双孢蘑菇菌丝体的生物量为测量指标,优化液体菌种的培养基配方及其培养条件,促进菌丝生长,在短期内使菌丝体的生物量达到最大,弥补了固体菌种在生产中的不足。试验结果显示,双孢蘑菇的最佳培养基配方为玉米淀粉 3%,蔗糖 2%,酵母粉 0.5%,磷酸二氢钾 0.15%,硫酸镁 0.2%;最适培养条件为温度 23 ℃,pH 5,转速 200 r/min,250 mL 三角瓶中的最适装液量为 150 mL。

关键词: 双孢蘑菇; 液体菌种; 菌丝体生物量; 正交试验

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Optimization of the Screening and Culture Conditions of the Liquefied Spawn of *Agaricus bisporus*

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Abstract: At present, the solid spawn is widely used in the cultivation of *Agaricus bisporus* in China. However, the growth cycle of solid spawn is very long and the efficient is low. Therefore, the orthogonal test method was used in this paper, taking the biomass of mycelium of *Agaricus bisporus* as the measurement index, the medium formulation and culture conditions of liquefied spawn were optimized to promote the growth of mycelium, the biomass of mycelium was maximized in a short time, this made up for the deficiency of solid spawn in production. The results illustrated the best medium formula was composed of 3% corn starch, 2% sucrose, 0.5% yeast powder, 0.15% potassium dihydrogen phosphate and 0.2% magnesium sulfate. The optimum culture conditions were as follows: temperature 23 ℃, pH 5, rotational speed 200 r / min, the optimal volume of liquid in 250ml triangular bottle 150 mL.

Keywords: *Agaricus bisporus*; liquefied spawn ; mycelial biomass; orthogonal experiment

双孢蘑菇(*Agaricus bisporus*)隶属于真菌界(Fungi)、担子菌门(Basidiomycota)、伞菌纲(Agaricomycetes)、伞菌目(Agaricales)、伞菌科(Agaricaceae)、蘑菇属(*Agaricus*)[1],富含多种氨基酸和维生素,是一种高蛋白、低脂肪的营养保健食品[2-4]。栽培品种主要包括白色、奶油色和棕色三个品系,其中白色品系栽培面积最大[5-9]。

食用菌的液体发酵技术是在抗生素发酵技术基础上发展起来的。液体菌种相较于固体菌种具有生产周期短、菌龄一致、接种简便、成本低等特点[10,11],并且液体菌种的生产效率高,易进行自动化控制,产品质量稳定,产品易于提取和精制,使得产量和质量都明显高于采用固体菌种栽培的生产模式,经济效益显著。

我国双孢蘑菇产业将逐渐向标准化、工厂化和周年化进行发展[12,13]。但目前的栽培模式主要采用固体菌种栽培模式,其生产周期长,生物学转化率低,需投入大量的人力、物力[14]。而液体菌种具有工艺简便、菌种纯、周期短、成本低、操作简单等优点。因此,本试验对液体菌种培养基及培养条件进行优化,为双孢蘑菇液体菌种的生产和应用提供技术参考,以期实现双孢蘑菇产业低碳经济增长的目的。

1 材料和方法

1.1 供试菌株

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红平菇液体发酵培养基优化

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摘　要：采用液体摇瓶培养法，通过单因素试验和正交试验，以菌丝体生物量为主要指标，对红平菇液体发酵培养基配方进行优化。结果表明：红平菇液体发酵最适宜的培养基配方为葡萄糖 20 g/L，酵母粉 15 g/L，KH_2PO_4 2 g/L，$MgSO_4 \cdot 7H_2O$ 0.5 g/L。进一步对正交试验结果进行验证表明该配方确实为较优配方。

关键词：红平菇；液体培养基；碳源；氮源；优化

Optimization of Fermentation Medium for Liquid Spawn of *Pleurotus djamor*

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Abstract: The liquid fermentation medium of *Pleurotus djamor* in submerged culture was studied according to the index of mycelium dry weight by single factor experiment and orthogonal experiment. The result showed that the optimum fermentation medium for liquid spawn of *Pleurotus djamor* was glucose 20 g/L, yeast extract 15 g/L, KH_2PO_4 2 g/L, $MgSO_4 \cdot 7H_2O$ 0.5 g/L, and we verify it through experiment.

Key words: *Pleurotus djamor*; liquid medium; carbon source; nitrogen source; optimization

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孟丽，宋超群，吴宝杰，等. 红平菇液体发酵培养基优化[J].食品研究与开发，2018，39(6)：169-171
MENG Li, SONG Chaoqun, WU Baojie, et al. Optimization of Fermentation Medium for Liquid Spawn of *Pleurotus djamor*[J]. Food Research and Development, 2018, 39(6): 169-171

红平菇（*Pleurotus djamor*）又名桃红平菇、红侧耳，属真菌门、层菌纲、伞菌目、侧耳科、侧耳属[1]。红平菇的子实体呈粉红色，色泽鲜艳。此外，红平菇味道鲜美，富含蛋白质、氨基酸、矿质元素和维生素，且有抗肿瘤、抗菌和增强机体免疫力的功效[2-9]，因此是一种兼具食用和观赏价值的珍稀蕈菌，具较高的商业开发前景。

食用菌液体菌种具有周期短、菌龄一致，发菌整齐，成本低等优点，因此在食用菌生产中具有广阔的应用前景[10-14]。目前关于红平菇的研究主要集中在固体栽培配方优化及其活性成分方面[15-19]，关于其液体菌种方面的报道较少。为此本研究采用液体摇瓶培养法，以菌丝体干重为指标，以碳氮源为主要因素对红平菇液体发酵培养基配方进行优化，旨在为红平菇液体发酵生产提供理论依据。

1　材料与方法

1.1　材料

1.1.1　菌种

红平菇菌种由山东农业大学菌物实验室提供。

1.1.2　培养基

PDA 综合培养基：马铃薯 200 g/L，葡萄糖 20 g/L，琼脂 20 g/L，$MgSO_4 \cdot 7H_2O$ 1 g/L，KH_2PO_4 1 g/L；液体培养基：葡萄糖 20 g/L，蛋白胨 10 g/L，$MgSO_4 \cdot 7H_2O$ 1 g/L，KH_2PO_4 1 g/L，pH 自然，0.1 MPa，121 ℃灭菌 30 min。

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甘蔗花叶病毒2个山东分离物的全基因组序列分析

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摘要:甘蔗花叶病毒(*Sugarcane mosaic virus*,SCMV)是引起我国玉米矮花叶病的主要病毒。本文从山东泰安采集到2个表现矮花叶症状玉米样品的分离物(命名为DWK1和DWK2),通过RT-PCR扩增全基因组片段并测定了其序列(GenBank登录号分别为KU171814和KU171815)。序列分析结果表明,DWK1和DWK2基因组全长分别为9 575和9 576个核苷酸(nucleotides,nt),开放阅读框均为9 192 nt,编码3 063个氨基酸的多聚蛋白。DWK1和DWK2的全基因组核苷酸一致率为81.7%,DWK1与山西分离物SX(AY569692)的核苷酸一致率最高,为90.9%;DWK2与河北分离物BD8(JN021933)核苷酸一致率最高,达99.4%。二者在系统进化树中分别被聚类到Ⅰ组和Ⅳ组。重组分析发现,DWK1是HN(AF494510)、Guangdong(AJ310105)和BD8 3个分离物的重组体。选择压力分析表明,SCMV 11个蛋白的d_N/d_S值都小于1,均处于负选择,但在P1、P3和CP中存在正选择位点。本研究结果可为甘蔗花叶病毒株系的监测及防控提供理论指导。

关键词:全基因组序列;系统发育分析;重组分析;甘蔗花叶病毒

Complete genomic sequence and analysis of two *Sugarcane mosaic virus* isolates from Shandong Province, China

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Abstract: *Sugarcane mosaic virus* (SCMV) is the major virus causing maize dwarf mosaic disease in China. We collected two maize samples showing dwarf mosaic symptoms from Tai'an, Shandong province. The causing pathogens were designated as DWK1 and DWK2, respectively. RT-PCR was performed to obtain the complete genomic sequences of the two isolates, and sequence analysis showed that the full-length genome of DWK1 and DWK2 (GenBank accession numbers KU171814 and KU171815) were 9 575 and 9 576 nucleotides (nt), respectively, which contain an open reading frame of 9 192 nt in length, encoding a putative polyprotein of 3 063 amino acids. The complete genome of DWK1 and DWK2 shared an identity of 81.7% at the nt level. DWK1 had the highest identity of 90.9% with isolate SX (AY569692) from Shanxi province, while DWK2 had the highest identity of 99.4% with isolate BD8 (JN021933) from Baoding of Hebei province. Phylogenetic tree was constructed based on the complete genomic sequences of different SCMV isolates, showing that DWK1 and DWK2 were clustered into groups I and IV, respectively. Recombination analysis indicated that DWK1 was a recombinant of the HN (AF494510), Guangdong (AJ310105) and BD8 isolates. The d_N/d_S values of all proteins were < 1.0, indicating that all the 10 proteins of SCMV were under negative selection. However, amino acids under positive selection were detected in P1, P3 and CP. Our results provide valuable information for the

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取食行为对丽蝇蛹集金小蜂雌雄成虫体内可培养真菌的影响*

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摘 要 **【目的】** 本研究旨在阐明取食行为对丽蝇蛹集金小蜂 *Nasonia vitripennis*（Walker）雌雄虫体内可培养真菌群落结构的影响，对其体内可培养真菌进行多样性研究。**【方法】** 采用不同浓度、不同培养基通过分离培养法研究丽蝇蛹集金小蜂体内真菌，将培养所得真菌采用形态和分子方法（ITS 基因序列）进行鉴定。**【结果】** 取食前后丽蝇蛹集金小蜂雌、雄虫体内共分离得到 49 种真菌，隶属子囊菌门，盘菌亚门的座囊菌纲、散囊菌纲、锤舌菌纲、子囊菌纲，共 10 个属，其中以链格孢属（*Alternaria*）、枝孢属（*Cladosporium*）、青霉属（*Penicillium*）为优势属。**【结论】** 丽蝇蛹集金小蜂体内可培养真菌物种数目，雄虫多于雌虫，取食后多于取食前。本研究结果说明食物、性别是丽蝇蛹集金小蜂体内可培养真菌群落结构产生差异的重要因素。

关键词 丽蝇蛹集金小蜂，可培养真菌，取食，性别，多样性

Effect of different foods on the fungal community cultured from adult female and male *Nasonia vitripennis* (Walker)

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Abstract **[Objectives]** To clarify the effect of different foods on the fungal community of adult male and female *Nasonia vitripennis* (Walker). **[Methods]** We cultured fungi from *N. vitripennis* after feeding them with different concentration and dilution plate techniques, and different media, and quantified the results using both morphological and molecular (ITS gene) methods. **[Results]** Forty-nine different fungal species were isolated from male and female adults of *N. vitripennis* before and after feeding. These were classified into 4 classes (Dothideomycetes, Eurotiomycetes, Leotiomycetes, Ascomycetes) and 10 genera of Ascomycota. The predominant genera were *Alternaria*, *Cladosporium* and *Penicillium*. **[Conclusion]** Males had a greater number of fungal species than females, and more fungi were cultured after feeding than before. These results indicate that food and the gender are important factors affecting fungi that can be cultured from *N. vitripennis*.

Key words *Nasonia vitripennis*, culturable fungi, feeding, gender, diversity

真菌起源于大约 10 亿年前带鞭毛的原生动物，与动物的亲缘关系非常密切（Wainright *et al.*, 1993）。昆虫作为动物界中第一大类群，与真菌在漫长的进化过程中逐渐形成了复杂而密切关系，包括了致病、寄生、互惠共生、偏利共生、携播、噬菌、竞争和捕食等（李增智，1997）。无论在昆虫的体表还是体内，都存在着丰富的真菌类群，与昆虫形成密切的关系，已经受到越来越多的关注。

目前，已经描述的真菌约为 7 万种，但科学

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6 种杀虫剂对侧柏幼苗根系活力的影响

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摘　要：笔者筛选出甲基异柳磷、辛硫磷、毒死蜱、增效喹硫磷、DDV 和乐果 6 种防治地下害虫的杀虫剂，对侧柏幼苗进行灌根处理，24 h 后取样测定 6 种杀虫剂对侧柏幼苗根系活力的影响。结果表明，增效喹硫磷能使侧柏幼苗根系活力增加，而甲基异柳磷、辛硫磷、毒死蜱、DDV 和乐果能显著降低侧柏幼苗根系活力。

关键词：侧柏幼苗；杀虫剂；根系活力

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Effects of Six Insecticides on Root Activity of *Platycladus orientalis*

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Abstract: The arborvitae seedlings root activity treated with six insecticides, isofenphos-methyl, phoxim, chlorpyrifos, quinalphos, dichlorvos and dimethoate were tested after 24h. The results showed that quinalphos could improve arborvitae seedlings root activity, while the others obviously restrained arborvitae seedlings root activity.

Key words: *Platycladus orientalis seedling*; Insecticide; Root activity

铜绿丽金龟（*Anomala corpulenta*）、黑绒金龟（*Maladera orientalis*）、小地老虎（*Agrotis ypsilon*）、大地老虎（*A. tokionis*）、黄地老虎（*Eudoa segetum*）、华北蝼蛄（*Gryllotalpa unispina*）、沟金针虫（*Pleonomus canaliculatus*）和细胸金针虫（*Agriotes fuscicollis*）等是苗圃地常见的地下害虫，一般危害林木幼苗根系。而根的生长情况和活力水平直接影响地上部分的生长和营养状况，其中，根系活力是植物根系的重要生理指标。许多研究表明，多种因素均可影响林木根系活力。如，张永利等 2009 年报道，用“根宝”可提高侧柏根系活力；1998 年，张留庆等证实晾晒可降低油松、侧柏根系活力。而有关杀虫剂对林木幼苗根系活力影响的报道较少。为此，笔者筛选了甲基异柳磷、辛硫磷、毒死蜱、增效喹硫磷、DDV、乐果 6 种防治地下害虫的药剂，以侧柏幼苗为供试植物，研究了杀虫剂对侧柏幼苗根系活力的影响。

1　材料与方法

1.1　材料

试验地选在泰安祝阳镇苗圃，供试植物为侧柏（*Platycladus orientalis*）1 年生实生苗，供试幼苗苗高 14.5 cm～45.5 cm，长势相差不大，土壤、浇水等其它处理因子相同。

本次供试杀虫剂有 40% 甲基异柳磷乳油（青岛双收农药化工有限公司生产）、40% 辛硫磷乳油（广东金农达生物科技有限公司生产）、40% 乐果乳油（河北省沧州志城化工有限公司生产）、50% DDV 乳油（赣州卫农农药有限公司生产）、40% 毒死蜱乳油（江苏蓝丰生物化工股份有限公司生产）、25% 增效喹硫磷乳油（四川省成都宇辰农药有限责任公司生产），以上药剂均由山东农业大学植物保护学院化保实验室免费提供。（下转第 64 页）

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三种农药对苹果幼苗根系活力的影响

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摘要:用微量注射器分别吸取不同浓度的苯醚甲环唑、氧乐果和毒死蜱 40 μL 涂抹于苹果幼苗叶片上,24 h后测定苹果幼苗根系活力。结果表明,苯醚甲环唑、毒死蜱均能使苹果幼苗根系活力上升,氧乐果则导致苹果幼苗根系活力下降。

关键词:苯醚甲环唑;氧乐果;毒死蜱;苹果;根系活力

中图分类号:S661.1;S481+3　　**文献标识码**:A　　**文章编号**:1006-9402(2016)01-0004-01

对于苹果病虫害的防治,化学防治法仍占主导地位,各种化学农药被广泛应用于病虫害的防治[1,2]。农药在使用过程中,更多关注的是杀虫和防病效果,对根系产生的影响,往往被忽视。本试验以目前防治苹果病虫害常见药剂苯醚甲环唑、氧乐果和毒死蜱为试验药剂,测试其叶部用药对苹果幼苗根系活力的影响,以期为合理使用和评价农药生态安全性提供依据。

1　材料与方法

1.1　材料　试验地点在山东农业大学校本部。供试植物为苹果 1 年生幼苗,品种为红富士。采用盆栽法,管理一致,测试时选取株高和地径相近的幼苗。

供试药剂为 40%苯醚甲环唑悬浮剂(天津汉邦植物保护剂有限责任公司生产)、40%毒死蜱乳油(江苏蓝丰生物化工股份有限公司生产)、40%氧乐果乳油(河北军星生物化工有限公司生产)。以上 3 种农药均由山东农业大学植物保护学院农药实验室免费提供。

1.2　测试方法　依据 3 种农药在生产中防治苹果病虫害用药浓度和产品使用说明书,用丙酮(分析纯)将苯醚甲环唑[3,4]、氧乐果[5,6]和毒死蜱[7,8]分别稀释 3 000、1 000、1 000 倍液。用微量进样器分别吸取不同浓度的药液,均匀涂抹于叶片上,每个叶片 40 μL,共处理 3 个叶片。以丙酮为对照。

24 h 后取新鲜根样进行测试,每处理随即抽取 3 株进行测试,重复 3 次。取样时,将苹果幼苗拔出,用自来水冲洗掉泥土,再用蒸馏水冲洗并擦干,取根尖部分测定根系活力。根系活力的测定采用 TTC 法[9],用岛津公司 U-2800 紫外分光光度计进行比色测定,以 TTC 还原量作为根系活力评价指标。

1.3　数据统计分析　用 DPS 进行数据整理分析,文中所有数据均用平均值表示。对所有数据进行方差分析,处理间的差异显著性用 LSD 检验。

2　结果与分析

从表 1 可以看出:药后 24 h,苯醚甲环唑和毒死蜱均使苹果幼苗的根系活力较对照上升,其中苯醚甲环唑较对照上升了 20.87%,与对照相比差异极显著(＞0.01),毒死蜱则较对照增加了 5.91%;而氧乐果与上述两种药剂作用相反,使苹果幼苗根系活力较对照下降了 15.28%。

表 1　三种农药对苹果幼苗根系活力的影响

处理	根系活力/μg・g^{-1}・h^{-1}
对照	62.6197aA
苯醚甲环唑 3000 倍液	75.6858bB
氧乐果 1000 倍液	53.0512bA
毒死蜱 1000 倍液	66.3203aA

注:5%显著水平时用小写字母表示,1%极显著水平用大写字母表示;字母相同时无显著差异,字母不同时表示差异显著。

3　结论与讨论

本次验结果证实,苯醚甲环唑、毒死蜱能促进苹果幼苗根系活力的增加,而氧乐果则导致苹果幼苗根系活力的下降。根系的生长、代谢和活力变化可直接影响到地上部的生长发育[10],一般情况下,根系活力越高,吸收养分的能力越强[11]。从试验结果来看,很显然苯醚甲环唑、毒死蜱对苹果幼苗根系活力能起到促进作用,相反,有机磷类杀虫剂氧乐果能抑制幼苗的根系活力。究其原因,我们认为与上述杀虫剂的理化性质有关,但有待于进一步探讨。

上述试验结果表明,在苹果病虫害综合治理中,从苹果生态安全性角度考虑,苯醚甲环唑、毒死蜱对苹果根系是安全的,氧乐果则会对苹果根系产生不利的影

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三种杀虫剂对桃树叶片叶绿素含量的影响

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摘要：用微量注射器分别吸取不同浓度的杀虫剂氧乐果、溴氰菊酯和吡虫啉 50 μL 涂抹于桃树叶片上，24 h 后测定桃树叶片叶绿素和类胡萝卜素的含量。试验结果表明：氧乐果和溴氰菊酯导致桃树叶片叶绿素 a、叶绿素 b、叶绿素总量下降，类胡萝卜素含量上升；吡虫啉则使叶绿素含量上升。

关键词：桃树；吡虫啉；溴氰菊酯；氧乐果；叶绿素；类胡萝卜素

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所报道的一些杀虫剂对多种农作物叶绿素含量的影响主要有以下几种：有些杀虫剂能够促进作物叶绿素含量的增加，如阿维菌素能导致玉米幼苗叶绿素和类胡萝卜素含量增加[1]；有些杀虫剂能够使作物叶绿素含量下降，如甲维盐能导致三种供试作物白菜、棉花、萝卜叶片中叶绿素 a、叶绿素 b、叶绿素总量和类胡萝卜素含量下降[2]；虫酰肼、丁醚脲和多杀霉素导致烟草幼苗叶绿素 a、叶绿素 b、叶绿素总量和类胡萝卜素含量下降[3]。但未见杀虫剂对果树叶绿素含量影响的报道。桃树在生长过程中，易受到桃蚜、桃瘤蚜和桃粉蚜等害虫的危害[4-5]，对于这些害虫的防治，化学防治法仍起着重要作用。

本试验以桃树为测试植物，探讨防治桃树害虫常用杀虫剂对桃树叶片叶绿素含量的影响，以期为合理使用和评价农药提供依据。

1　材料与方法

1.1　材料　试验地点在山东农业大学校本部。供试植物为桃树，树龄 10 年，地径约 26 cm，树高 2.8 m。测试时选取无明显叶部害虫、无蛀干害虫的植株为样本，选取叶片大小相近、无害虫、近于同一树冠层的叶片为测试叶片。

供试药剂为 10%吡虫啉可湿性粉剂（拜耳作物科学有限公司生产）、25 g/L 溴氰菊酯乳油（拜耳作物科学有限公司生产）、40%氧乐果乳油（杭州庆丰农化有限公司生产）。以上 3 种杀虫剂由山东农业大学植物保护学院慕卫和薛超彬教授免费提供。

用丙酮（分析纯）将氧乐果、溴氰菊酯和吡虫啉分别稀释至 500、1 000、1 500 倍液。用微量进样器分别吸取不同浓度的药液，均匀涂抹于叶片上，每个叶片 50 μL。以丙酮为对照。

1.2　叶绿素含量的测定　叶绿素含量的测定参照张悦等人的试验方法[3]。取新鲜植物叶片，擦净叶片表面污物，剪碎（去叶脉），混匀。秤取剪碎的新鲜样品 0.2 g，放入研钵中，加少量石英砂和 2~3 mL 80%丙酮，研磨成匀浆，再加丙酮 10 mL 研磨至样品组织变白，暗处静置 5 min，过滤到 50 mL 棕色容量瓶中，洗涤研钵和残渣数次，定容后备测。80%丙酮作为参比，在 665、649、470 nm 下，应用日本岛津 UV-2450 紫外分光光度计测吸光度值。

计算公式：

$$C_a=12.21A_{663}-2.81A_{646}$$

$$C_b=20.13A_{646}-5.03A_{663}$$

$$C_T=C_a+C_b$$

$$C_{x\cdot c}=\frac{1000A_{470}-3.27C_a-104C_b}{229}$$

式中：C_a、C_b、C_T和$C_{x\cdot c}$分别为叶绿素 a、b 的浓度，叶绿素总量，类胡萝卜素的含量；A_{663}、A_{646}和A_{470}分别为叶绿体色素提取液在波长为 663 nm、646 nm 和 470 nm 下的光密度值。

1.3　数据统计分析　用 DPS 进行数据整理分析，文中所有数据均用平均值表示。对所有数据进行方差分析，处理间的差异显著性用 LSD 检验。

2　结果与分析

2.1　吡虫啉对桃树叶片叶绿素及类胡萝卜素含量的影响　从表 1 可以看出：药后 24 h，桃树幼苗叶绿素 a 含量较对照均有上升，其中 1 500 倍吡虫啉上升幅度最大，较对照上升了 46.92%，与对照相比差异极显著（>0.01）。叶绿素 b 和叶绿素总量的变化趋势和叶绿素 a 的变化规律基本一致，均为 1 500 倍吡虫啉使叶绿素 b 和总量上升幅度最大，500 倍吡虫啉使叶绿

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Improving the thermostability of a thermostable endoglucanase from *Chaetomium thermophilum* by engineering the conserved noncatalytic residue and N-glycosylation site

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ABSTRACT

Endoglucanases provide an attractive avenue for the bioconversion of lignocellulosic materials into fermentable sugars to supply cellulosic feedstock for biofuels and other value-added chemicals. Thermostable endoglucanases with high catalytic activity are preferred in practical processes. To improve the thermostability and activity of the thermostable β-1,4-endoglucanase CTendo45 isolated from the thermophilic fungus *Chaetomium thermophilum*, structure-based rational design was performed by using site-directed mutagenesis. When inactivated mutation of the unique N-glycosylation sequon (N88-E89-T90) was implemented and the conserved Y173 residue was substituted with phenylalanine, a double mutant T90A/Y173F demonstrated enzymatic activity that dramatically increased 2.12- and 1.82-fold towards CMC-Na and β-D-glucan, respectively. Additionally, T90A/Y173F exhibited extraordinary heat endurance after 300 min of incubation at elevated temperatures. This study provides a valid approach to the improvement of enzyme redesign protocols and the properties of this endoglucanase mutant distinguish it as an excellent candidate enzyme for industrial biomass conversion.

1. Introduction

Lignocellulosic biomass is the earth's most abundant utilizable organic resource with great potential for the sustainable production of chemicals and biofuels [1,2]. Cellulose serves as a major component (approximately 35–50%) of lignocellulosic biomass, followed by hemicellulose (20–30%) and lignin (20–30%) [3]. Endoglucanase (EC 3.2.1.4), which is a critical set of cellulases for lignocellulose deconstruction, randomly cleaves the internal β-1,4-glucosidic bonds in cellulose fibers and triggers the initial catalytic attack on biopolymer chains in amorphous regions [4].

From a practical point of view, the inadequate performance and instability of commercial cellulases under extreme reaction conditions is now considered as the main obstacle to their effective application in industrial and agricultural fields which results in extending the reaction period and raising the production cost [5]. Therefore, for modern cellulase preparation research and development, the improvement of enzyme specific tolerability and catalytic efficiency is essential [6,7]. To date, structure-based rational engineering is an effective approach to enhancing enzymatic properties, which is implemented based on the modification of some crucial residues and domains in terms of the enzyme's structure-function relationship [8,9]. In addition, more attention has been focused on the optimization of conserved residues in the active site architecture, which play important roles in modulating enzyme structure and catalytic properties [10,11].

Elevating the reaction temperature is generally conducive to substantially increasing the hydrolysis rate and preventing microbial contamination in the realistic biorefinery of lignocellulosic saccharification [12]. Thus, thermostability is a valuable property for cellulases in practice. Utilizing potent thermostable cellulases endowed with pronounced activity at high temperatures is beneficial for accelerating the catalytic process, shortening the reaction period and reducing the enzyme dosage [2,13]. N-glycosylation, which represents a ubiquitous post-translational modification in eucaryon, is capable of regulating the thermal stability and hydrolysis activity of cellulases [14–17]. In the case of a glycoprotein, the carbohydrate unit is covalently connected to the specific asparagine residue within the sequon Asn-Xaa-Ser/Thr (where Xaa cannot be Pro) [18].

In our previous work, rational design of a thermostable GH45 β-1,4-endoglucanase CTendo45 from *Chaetomium thermophilum* was performed to further enhance the thermostability by substituting the conserved noncatalytic Y173 residue with F in the active site architecture using site-directed mutagenesis [19]. Additionally, when the unique N-glycosylation sequon in CTendo45 was modified, a T90A variant exhibited superior characteristics of high temperature tolerance [17]. In this study, a double mutant T90A/Y173F was generated and exhibited enhanced thermostability compared with that of its single counterparts, providing a promising biocatalyst for diverse biotechnological

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德州市农作物病虫害统防统治发展现状及对策

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摘 要：随着农村产业结构的调整，农村劳动力大量转移输出，促进了土地流转，随着农作物种植结构的调整、耕作制度以及气候条件的变化等，病虫害发生的种类和危害的程度也发生了变化，突发性、暴发性、流行性病虫害时有发生，防治难度加大，多元化的植保服务组织应运而生，防治方式发生了根本性的改变，以户为单位的零散式粗放型防治，逐渐改变为由植保服务组织为主的规模化机械化专业化的集约型的统防统治，并逐步实现飞机防治.

关键词：农作物；病虫害；统防统治；发展及方向

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1 引言

德州市位于山东省的西北部，是一个以农业生产为主的大市，辖11个县市区，耕地面积$6.3\times10^5 hm^2$，是全国主要的优质商品粮生产基地，以种植小麦、玉米、棉花三大农作物为主，小麦、玉米常年种植面积分别在$5.0\times10^5 hm^2$以上，病虫草鼠害常年发生面积$4.2\times10^6 hm^2$次以上，通过防治，每年挽回粮食1.1×10^9 kg以上，有效地控制了病虫草鼠危害. 近年来，随着种植结构的调整、耕作制度和气候条件的变化，新的病虫害不断出现，次要病虫上升为主要病虫，突发性、暴发性、流行性病虫害时有发生，给病虫害防治带来了很大困难，传统的一家一户单独分散的防治方式，很难达到预期的防治效果[1].

农作物病虫害统防统治始于上世纪九十年代，在防治方式上，主要推广了“五统一分”（即统一病虫预报、统一防治方案、统一印发明白纸、统一防治时间、统一施药标准、统一组织实施等）、技术指导带药、单项病虫承包防治等形式，随着时间推移，逐渐增加了新的元素，赋予了新的内涵. 近几年，植保专业合作社、种粮大户、家庭农场等新型农业经营主体参与进来，使植保体制转变为植保部门、新型农业经营主体和农民三位一体[2]，通过扶持发展植保服务组织，引进示范先进的大中型植保机械，由人背肩扛的人工防治，逐渐向大型植保机械防治转变，传统的以户为单位的单独分散式的防治方式，逐渐向规模化、机械化、专业化的统防统治转变，并逐步实现飞机防治.

2 植保服务组织发展情况

2.1 植保服务组织历程

2008年植保专业合作社在陵城、夏津等县市开始成立，主要承担农作物病虫害的防治任务，2009年山东省开始实施农作物病虫害专业化防控体系建设项目，2013年本项目更名为山东省农业病虫害专业化统防统治能力建设示范项目，其主要内容就是由省财政投资，招标植保机械，用于扶持示范县的植保服务组织，开展专业化的统防统治作业. 目前德州市共投资1000余万元，项目实施最初几年，主要是购买机动喷雾器和防护服等，以后逐渐购买大中型植保机械和无人机，机械全部由省农业厅统一招标采购，示范县在中标产品中选购，县级农业局或植保站与扶持的服务组织签订协议，机械由所扶持的组织使用、存放和维护，所有权归县级农业局，在病虫害大发生或暴发流行时，由县级农业局统一调配使用. 德州市的陵城、夏津、齐河、武城、临邑、乐陵等县市区均被列为示范县，扶持了一批植保专业化服务组织，大力开展了农作物病虫害专业化统防统治和飞机防治，齐河齐力新、夏津农家丰、临邑富民合作社等先后被评为山东省优秀植

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Lauric Acid Is a Potent Biological Control Agent That Damages the Cell Membrane of *Phytophthora sojae*

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Sustainable management of plant pathogens is becoming more challenging, and novel solutions are needed. Plant biologically active secondary metabolites are important sources of novel crop protection chemistry. Effective individual compounds of these natural products have the potential to be successful new agrochemicals. In this study, we identified lauric acid (LA) from soybean defense leaf volatiles. LA inhibited the growth of *Phytophthora sojae*, the causal agent of soybean root rot. It influenced mycelial development, sporangium formation, and zoospore generation and germination by damaging the *P. sojae* cell membrane. Additionally, we showed that LA and several of its derivatives, such as glycerol monolaurate (GML), had similar biological activities. Both LA and GML were safe to soybean plants when used at less than 0.3 g a.i./plant and could promote soybean growth, implying their potential as eco-friendly biological control agents.

Keywords: lauric acid, leaf volatile compounds, *Phytophthora sojae*, cell membrane damage, biological control

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INTRODUCTION

Oomycetes are fungus-like eukaryotic organisms that belong to the class Saprolegniomycetidae of the kingdom Stramenopila (Yutin et al., 2008). They encompass notorious plant disease agents, including *Phytophthora*, *Pythium*, and *Albugo*, and a group of downy mildews (Tyler, 2001). Oomycetes have a negative impact on natural and farm ecosystems due to their strong pathogenicity and infectivity (Kamoun et al., 2015). In addition to the well-known potato late blight caused by *Phytophthora infestans*, which led to the 19th-century Irish famine, the persistence of sudden oak death caused by *Phytophthora ramorum* and grape downy mildew caused by *Plasmopara viticola* demonstrate that oomycete phytopathogens are a persistent threat to subsistence and commercial farming and destructive to native plants (Erwin and Ribeiro, 1996). Soybean (*Glycine max* L.) root rot caused by *Phytophthora sojae* is the leading cause of global soybean production loss (Tyler, 2007). Agrochemicals are largely used to control oomycete diseases, resulting in the emergence of resistant strains and resurgence events (Randall et al., 2014). The effective and sustainable control of oomycete-driven diseases requires the identification of novel pharmaceuticals and pesticides to drive the design of fungicides compatible with integrated pest management (IPM) approaches (Gessler et al., 2011).

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Research

Stored-Product

Biological Activity of *trans*-2-Hexenal Against the Storage Insect Pest *Tribolium castaneum* (Coleoptera: Tenebrionidae) and Mycotoxigenic Storage Fungi

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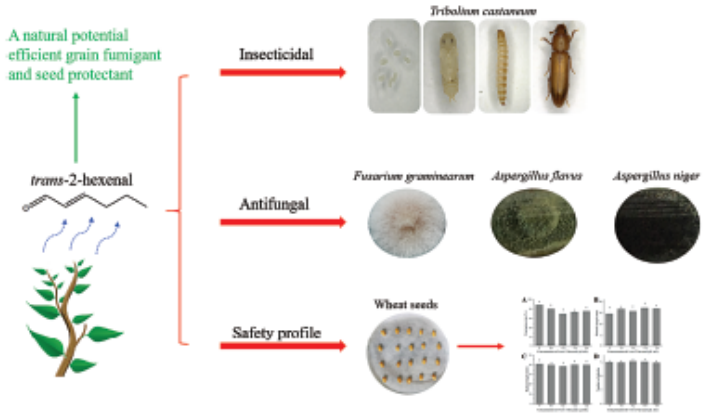
Abstract

Grain commodities in postharvest storage often deteriorate because of fungal and insect attacks. With the green consumption requirements of consumers, ecofriendly and safe pesticides are needed for grain storage. The current study investigated the efficacy of the plant volatile compound *trans*-2-hexenal against the storage insect pest *Tribolium castaneum* (Herbst) and three commonly occurring storage fungi, viz., *Fusarium graminearum*, *Aspergillus flavus*, and *Aspergillus niger*, to recommend its application as a botanical fumigant for grain commodities. *trans*-2-Hexenal weakly repels *T. castaneum* but has favorable insecticidal activity against multiple developmental stages of *T. castaneum*, ranging in sensitivity as follows: eggs (LC_{50} = 14.3 μl/l) > adults (31.6 μl/l) > young larvae (42.1 μl/l) > mature larvae (64.5 μl/l) > pupae (70.5 μl/l). Moreover, *trans*-2-hexenal caused a high malformation rate and high mortality in adults developed from fumigated pupae. In a 7-d grain, *trans*-2-hexenal at 0.8 μl/ml provided an appreciable efficacy (81.3%), and concentrations ≥ 0.1 μl/ml completely inhibited the offspring of *T. castaneum*. *trans*-2-Hexenal was nonphytotoxic to the seed germination and seedling growth of wheat seeds. Furthermore, *trans*-2-hexenal completely inhibited the growth of *A. flavus*, *F. graminearum*, and *A. niger* at 5, 10, and 10 μl/l, respectively. The favorable biological activity of *trans*-2-hexenal against *T. castaneum* and three frequently occurring mycotoxigenic storage fungi indicated the potential of *trans*-2-hexenal for simultaneously controlling pests and pathogens, which could reduce its application frequency in grains and decrease pesticide resistance risks.

Graphical Abstract



综述专论

植物免疫诱抗剂的发现、作用及其在农业中的应用

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摘要：植物免疫诱抗剂利用植物天然免疫系统，提高植物抗病性，激发植物代谢调控，促进植物生长发育，达到增产和提质效果。主要介绍了植物免疫诱抗剂的发展历程、种类和作用机理，阐述了植物免疫诱抗剂在农业生产中的应用及其优势，展望了应用前景。

关键词：植物免疫诱抗剂；抗病；增产；农业生产

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Discovery, action and application of plant immune inducers in agriculture

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Abstract: based on the plant's natural immune system, plant immune inducers can improve plant resistance to diseases, stimulate metabolic regulation and promote growth and development to increase output and improve quality of plant. The paper introduced the development, types and mechanisms of action of plant immunity inducers, described the application situation and advantages of plant immune inducers in agriculture production, and look forward to the prospects of plant immunity inducers.

Keywords: plant immune inducers; resistance to disease; increase production; agricultural production

植物病虫害的化学防治是我国粮食产量和品质的保障，但是化学农药的过度使用给环境和人类健康造成严重损害。2015年农业部提出化肥农药零增长计划，因此开发生物农药是解决农药污染问题，实现农药减量的有效手段之一。植物免疫诱抗剂作为新型生物农药，并不能直接对病原菌进行强烈的毒杀，而是以植物为目标，通过调节植物本身的免疫、代谢系统来增强植物对病原菌的抗性，提高植

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草地贪夜蛾对山东玉米的为害情况研究进展

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摘 要：草地贪夜蛾侵入我国后，已经造成大规模迁飞为害。为保护我国粮食作物的产量和品质，总结了草地贪夜蛾在玉米上的发生规律、为害特点以及基础防治措施等方面的研究进展，为有效防治玉米上草地贪夜蛾的为害提供依据。

关键词：草地贪夜蛾；山东；玉米

草地贪夜蛾（*Spodoptera frugiperda*）属于鳞翅目夜蛾科灰翅夜蛾属，原产于美洲热带和亚热带区域，并且分布十分广泛，2018 年侵入我国，2019 年在我国大规模迁飞为害。草地贪夜蛾寄主广泛，喜食禾本科作物与杂草，在我国，玉米是其为害最重的主要农作物之一，但由于草地贪夜蛾为近年入侵的物种，有很多农业工作者对其在玉米上的发生规律与为害特征了解不多，给有效防治带来困难。故此，总结了学术界对草地贪夜蛾在玉米上的生活习性、为害特点以及基础防治措施等方面的研究进展，给有效防治玉米上草地贪夜蛾的为害提供依据。

1 发生规律

为摸清草地贪夜蛾在玉米上的为害特点与发生规律，林丹敏等[1]选用玉米来调查该虫在不同生育期（苗期、拔节期、大喇叭口期、抽雄期、开花抽丝期以及成熟期）的虫口数量和密度。调查发现，部分不同地区、同一地区不同田块草地贪夜蛾有发生不均衡的现象，这是由于草地贪夜蛾迁飞具有偶发性，而且不同地区和田块玉米种植类型、数量和比例并不会完全相同，其中可能存在草地贪夜蛾转移到花生、高粱等寄主上为害，也容易对调查结果产生较大影响。但总体来看，玉米不同生育期受害程度为：苗期受害最严重，其余依次为大喇叭口期、成熟期、拔节期、开花抽丝期，抽雄期受害最轻。可能是由于苗期、大喇叭口期的玉米叶片幼嫩易取食的缘故。

刘元兵等[2]调查发现，老龄幼虫取食玉米叶片常会造成不规则的长形孔洞，产生点片破损，严重时可吃光整株叶片，并排泄大量虫粪，风干后似锯末。

2 取食习性

在我国，除玉米受草地贪夜蛾为害严重之外，小麦、花生、高粱等也存在较高的被为害风险。张云慧等[3]发现，成虫在玉米上的日平均落卵量显著大于在其相应田间杂草上的产卵数量，且玉米叶片背面是成虫最喜欢的产卵部位。在不同的龄期阶段幼虫表现出的对玉米的需求和趋向性上，姚领等[4]认为草地贪夜蛾幼虫对玉米的取食选择性随龄期增长而逐渐减弱，趋于稳定。在对幼虫在玉米、小麦与其他杂草上存活率的研究中，张云慧等[3]发现在玉米和小麦上存活率最高，但禾本科雀麦却是两类农田杂草（指玉米田杂草和小麦田杂草）中存活率最低的，分析其原因可能由于雀麦茎叶外表均被绒毛，不利于幼虫附着与取食，因而草地贪夜蛾幼虫在其上存活率最低。

吕亮等[5]比较玉米和小麦哪一种是草地贪夜蛾在二者都存在的情况下优先选择的作物试验中，在玉米和小麦都可供草地贪夜蛾幼虫利用时，会优先选择玉米为害；但若无玉米存在时，则会选择小麦且可以转移为害。

3 来源研究

针对入侵山东省的草地贪夜蛾来源研究中，王鹏等[6]以 mtCOI 作为分子标记，对首次入侵山东各地的草地贪夜蛾样本进行了分子鉴定。结果发现，山东省草地贪夜蛾的单倍型主要为玉米型和水稻型，其中水稻型占绝大多数。但若综合核基因与线粒体基因分析来看，占入侵我国草地贪夜蛾种群数量主导地位的是核基因为“玉米型”，而线粒体基因为“水稻型”的杂合体玉米型，基因纯合的玉米型只有不到 10%。

4 为害严重的原因

影响草地贪夜蛾发生为害的因素有很多，主要包括环境因素（主要是温度）、寄主、天敌等。

4.1 温度

何莉梅等[7]研究发现，在草地贪夜蛾适生的温度范围内，随着温度的降低，7 龄幼虫出现的概率会逐渐增加。而在高温和低温条件下，草地贪夜蛾的蛹都比其他虫态存活率高。罗举等[8]通过测定草地贪夜蛾各虫态和幼虫各龄期过冷却点的高低得出，其结冰点最高为卵和 1 龄幼虫，最低为蛹。因此，可以断定草地贪夜蛾以抗热抗寒能力较强的蛹进行越冬。

温度还影响草地贪夜蛾的迁飞能力。谢瑕杰等[9]通过观测与试验得出，草地贪夜蛾成虫在田间 24℃的条件下飞行速度最快、飞行距离最长且持续时间久；而在实验室饲养的环境条件下，32℃时成虫的迁飞能力最强，甚至比其适生温度范围内能力还强，分析原因应该是过高的温度不适宜草地贪夜蛾的生存，从而刺激该虫进行迁飞逃离不利环境。

虫螨腈及其代谢物在韭菜和姜上的残留检测

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Determination of Residues of Tetranylonitrile and Its Metabolites in Leek and Ginger

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Abstract: The study established a method for the determination of chlorfenapyr and its metabolite tralopyril in leek and ginger. The samples were extracted with acetonitrile and purified with 40mg PSA, 10mg C_{18} and 5mg MWCNT. Chlorfenapyr was analyzed by GC-MS/MS and tralopyril by UPLC-MS/MS. The results showed that there was a good linear relationship for chlorfenapyr and tralopyril in the concentration range of 0.001 ~ 0.5mg/L, with the correlation coefficient of more than 0.99. When the matrix concentration were 0.01mg/kg, 1mg/kg and 10mg/kg, the average recoveries of chlorfenapyr in leek were 88.4% ~ 104% with relative standard deviations of 1.5% ~ 3.9%, and the average recoveries 93.0% ~ 99.9% with the relative standard deviations 2.8% ~ 4.9% in ginger. The average recoveries of tralopyril in leek were 87.8% ~ 98.9%, with relative standard deviations of 4.0% ~ 4.6%, and the average recoveries 87.2% ~ 94.7% with the relative standard deviations 2.7% ~ 4.6% in ginger. The method is accurate, sensitive and suitable enough for the determination of chlorfenapyr and its metabolites in leek and ginger.

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新型介孔材料_QuEChERS_超高效液相色谱_串联质谱法检测茶叶中 10 种农药残留

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摘 要 基于 QuEChERS 法,以自组装的新型介孔材料($SBA-15-C_{18}$)为净化剂,结合超高效液相色谱_串联质谱法(UPLC-MS/MS),建立了茶叶基质中 10 种农药残留的分析方法。在模板剂作用下,采用溶液化学反应法制备了 SBA-15;通过十八烷基硅烷偶联剂在 SBA-15 表面以原位共缩合法制备了 $SBA-15-C_{18}$,采用扫描电镜和红外光谱进行表征。样品以乙腈提取,经 $SBA-15-C_{18}$、纳米氧化锆和多壁碳纳米管组合净化,利用 UPLC-MS/MS 外标法定量分析。结果表明,$SBA-15-C_{18}$ 具有二维通孔结构,粒径大小均一,呈圆柱形弯曲状,平均粒径在 240~340 nm 之间。茶叶基质中 10 种农药在 0.0025~0.5 mg/L 浓度范围内线性关系良好,相关系数(R^2)在 0.9978~0.9999 之间;在 0.01、0.1 和 20 mg/kg 添加水平下,10 种农药的平均回收率在 72%~111%之间,相对标准偏差在 1.0%~6.9%之间。茶叶基质中 10 种农药的检出限为 0.1~0.3 μg/kg,定量限为 0.2~0.9 μg/kg。将本方法应用于实际茶叶样品的检测,结果显示,所有样品均符合农药最大残留限量标准。本方法具有灵敏度高、通用性强、准确度高、稳定性好、操作简便等优点,适用于茶叶中农药多残留的检测。

关键词 茶叶; 介孔材料 $SBA-15-C_{18}$; 超高效液相色谱_串联质谱; 农药残留

茶叶是我国主要的出口经济作物之一[1]。茶树喜欢生长在湿热的环境,因此,茶树病虫害频繁发生,使用农药是防治茶树病虫害最有效的方法。用于茶树病虫害防治的农药多达数十种,茶叶中的农药残留问题越来越受到关注[2]。目前,我国制定了茶叶中 65 项农药最大残留限量标准(MRLs),作为主要茶叶消费市场的欧盟和日本分别规定了茶叶中 502 种和 230 种农药的 MRLs[3],相比之下,我国仍存在巨大差距。欧盟和日本规定茶叶中化学农药的 MRLs 的最小值均为 0.01 mg/kg,比我国的标准更加严格,这严重影响了我国茶叶的对外贸易。

目前,有关茶叶中农药残留的分析方法已有很多报道,主要以质谱技术和色谱技术为主[4]。诸力等[5]利用超高效液相色谱_串联质谱(UPLC-MS/MS)法同时测定茶叶中 11 种植物生长调节剂及吡虫啉、啶虫脒的残留,样品用乙腈_甲酸溶液提取,采用 C_{18}、强阴离子交换剂和 N-丙基乙二胺(PSA)分散固相萃取,13 种农药的定量限(LOQ)为 0.6~32 μg/kg。张新忠等[6]利用分散固相萃取净化气相色谱_质谱法测定茶叶、西葫芦和芒果中噻螨酮和噻嗪酮残留量,样品以乙腈提取,经 PSA 和石墨化碳(GCB)净化,2 种农药的 LOQ 均为 10 μg/kg。与蔬菜、水果基质相比,茶叶成分复杂,其农药残留分析难度大,前处理作为关键技术会直接影响分析方法的灵敏度、准确度和精密度。目前,茶叶基质的前处理方法主要有 QuEChERS 法[7,8]和固相萃取法[10-12]等。QuEChERS 法由于具有简便、廉价和通用性强等优点,被广泛应用于茶叶的农药残留前处理过程中[13]。QuEChERS 法常用的净化剂有 C_{18}、PSA 和 GCB 等[14],由于茶叶基质效应较为严重,常规净化剂净化效果一般,并且用量较大、成本较高。新型介孔材料由于具有规则的孔道结构、高的比表面积等优点,被广泛用于吸附领域[15]。

UPLC-MS/MS 技术因具有检测灵敏度高、适用范围广、分析速度快、高通量和有效排除复杂基质干扰等优点[16-17],已成为茶叶中农药残留检测的首选方法。介孔材料 SBA-15 及其衍生物具有吸附通用性高、吸附稳定性强和吸附容量大等特点。本研究在 SBA-15 的活性位点引入 C_{18} 基团,使 $SBA-15-C_{18}$ 在具备 SBA-15 结构特点的基础上,又兼具 C_{18} 对非极性和弱极性化合物良好的吸附特性,并将其作为常规净化剂的替代净化剂,用于茶叶基质中农药多残留检测。本研究基于 QuEChERS 方法,以

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Janthinoid A, an unprecedented tri-*nor*-meroterpenoid with highly modified bridged 4a,1-(epoxymethano)phenanthrene scaffold, produced by the endophyte of *Penicillium janthinellum* TE-43†

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Janthinoid A (1), an unprecedented C_{22} meroterpenoid featuring a highly modified bridged 4a,1-(epoxymethano)phenanthrene scaffold incorporating eight continuous quaternary carbons, along with its biosynthetic-related C_{25} analogue andrastone I (2), were isolated from the endophytic fungus *Penicillium janthinellum* TE-43. Their structures were unambiguously established by comprehensive analyses of spectroscopic data, quantum chemical calculations, and X-ray diffraction. The tri-*nor*-meroterpenoid skeleton of **1** was produced by a unique biosynthetic pathway, involving the ring cleavage and continuous carbon degradations of the aromatic polyketide precursor, which is distinct from the commonly-observed meroterpenoids both structurally and biogenetically, representing a new type of meroterpenoid. Both compounds showed significant *in vivo* anti-tumor activities against NSCLC cells A549.

Introduction

Fungi have played a key role in the search for new drug leads in the course of human history, as they possess a tremendous capacity to synthesize diverse secondary metabolites with significant medicinal potential.[1] The discovery of penicillin from *Penicillium* species was a monumental discovery in medical research, which drives continuous effort to find more novel metabolites from fungi.[2] The increase of chemical investigations on fungal metabolites has unabated, leading to the isolation of a plethora of intriguing molecules.[1–3] As one of the largest groups of fungal metabolites, meroterpenoids derived from mixed polyketide/nonpolyketide-terpenoid biosynthetic pathways have received particular attention.[4] Among them, andrastone-type meroterpenoids represent a small but unique group of bioactive meroterpenoids with approximately forty members. Biosynthetically, the C_{25} skeleton of andrastones is composed of a C_{15} farnesyl pyrophosphate (FPP) precursor incorporating a C_{10} DMOA (3,5-dimethylorsellinic acid, Fig. 1) unit. The subsequent cyclization of the FPP fragment followed by the ring-contraction of DMOA provides the all-carbon, fused 6/6/6/5 tetracyclic core.[5]

As a part of our ongoing research on bioactive fungal metabolites,[6–8] especially on miscellaneous meroterpenoids,

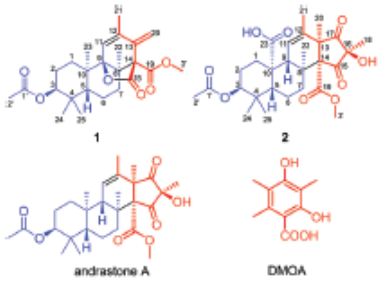

Fig. 1 Chemical structures of **1** and **2**, andrastone A, and the key precursor DMOA.

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添加适宜氮磷对连作平邑甜茶幼苗生长及土壤环境的影响

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摘要: 以盆栽平邑甜茶幼苗为试验材料, 设置连作土对照(CK)、单施氮肥(N)、施氮磷肥(NP)和单施磷肥(P) 4个处理, 探讨氮磷添加对连作平邑甜茶幼苗及土壤环境的影响。结果表明, 与对照相比, 施NP显著促进了平邑甜茶幼苗株高、地径、干重和鲜重的增加, 分别为对照的1.58倍、1.38倍、2.48倍和2.6倍; 显著提高了根系长度, 为对照的2.51倍; 提高了平邑甜茶幼苗的根系呼吸速率和根系超氧化物歧化酶(SOD)、过氧化物酶(POD)和过氧化氢酶(CAT)的活性, 分别为对照的1.92倍、1.26倍、2.69倍、3.23倍; 同时提高了土壤细菌/真菌比值和土壤主要酶活性。综上所述, 连作土中添加适宜氮磷可显著改善连作土壤环境, 促进根系生长, 最终减轻苹果连作障碍现象。

关键词: 平邑甜茶; 苹果连作障碍; 氮肥; 磷肥; 土壤微生物

苹果产业是我国的优势产业, 到2014年我国苹果种植面积已达222.15万hm^2, 目前近80%的苹果栽培面积都集中在环渤海湾和西北黄土高原地区(里程辉等2016)。由于山东省大部分果树分布于山丘地, 土壤相对贫瘠, 土地资源有限, 各种农作物争地以及栽培条件等各方面因素的限制, 使苹果主产区面临严重的连作障碍问题(Tewoldemedhin等2011; 王闯等2009)。连作障碍通常表现为植株矮小、幼苗存活率低、低产量和品质下降、病虫害加重等问题, 造成巨大的经济损失(Liu等2014; Mazzola和Manici 2012)。苹果连作障碍的致病因素非常复杂, 主要包括生物和非生物因素两大类, 有研究表明土壤有害真菌的增加是导致连作障碍发生的重要原因之一(Cardinale等2006; 李家家等2016), 有害真菌的增加导致土壤微生物种群结构发生改变(Yim等2013)。非生物因素包括土壤有毒物质积累, 土壤理化性质恶化, 土壤养分失衡等(Zhang等2012)。

氮磷肥是苹果幼树管理过程中必须施用的肥料, 会对苹果幼树的根系发育及正常生长产生影响。在果园实际生产中, 施肥对苹果生长发育具有重要意义。植物与土壤养分具有相互协调的作用, 改变土壤中营养元素的水平可以影响植物根系的生长发育和分泌特性, 进而导致植物根际微生物种群结构与功能、土壤酶活性以及土壤pH值等发生变化(王富林等2013; 罗燕和樊卫国2014; 祖艳群等2015)。不同的施肥制度对土壤微生物数量、群落结构影响差异显著(时鹏等2010), 早有国外研究表明果园施用磷酸盐肥料可有效地控制苹果再植病的发生或减轻其损害程度(Utkhede 1995; Gur等1998)。尹承苗等(2013)研究发现有机物料能够有效地减轻苹果连作障碍对幼树生长发育的影响。施用适量的肥料可增加重茬苹果根系生物量, 提高土壤有效养分含量, 促进微生物生长繁殖(陈伟2007; 樊红科2008)。

因此, 通过施肥调控果树生长发育及果树根系生长的土壤环境是不可避免的, 但氮和磷如何影响连作苹果植株的生长、连作土壤微生物结构以及对连作障碍发生程度的影响有何不同鲜见报道。本研究以平邑甜茶幼苗为试材, 探讨添加氮磷对平邑甜茶幼苗生长及连作土壤环境的影响, 以期为减轻苹果连作障碍提供新的措施。

材料与方法

1 试验材料与处理

试验于2015年在山东农业大学南校区国家苹果工程技术研究中心试验基地进行。供试土壤取自山东省泰安市满庄镇25年生红富士苹果园, 砧木为八棱海棠(*Malus micromalus* Makino), 土壤类型为棕壤土, 速效钾含量为90.6 mg·kg^{-1}, 速效磷含

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基于 Web of Science 的土壤微生物研究文献国际发展态势分析

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摘　要：在文献调研、专家咨询和文献计量分析的基础上，对国际土壤微生物研究的发展历程进行了回顾，利用文献计量学方法分析了土壤微生物学研究的主要国家和机构状况，分析了近年来相关研究的学科分布以及热点主题内容，分析了国际土壤微生物研究的发展态势和挑战。结果表明：国际土壤微生物研究的学科分布主要集中在土壤科学、环境科学，而且土壤科学、环境科学、生态学、微生物学、农学生物技术应用微生物学的文献数量达到文献总量 75.04%，表明这几个学科领域是土壤微生物研究的前沿热点；热点主题主要集中在土壤微生物多样性、生物量、有机质、土壤碳等方面，总结了这些领域的发展趋势和重点研究方向，并根据分析结果，对我国土壤微生物学领域采取的措施、今后发展的方向提出了建议。

关键词：土壤微生物；文献计量学；研究进展

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土壤是人类赖以生存与发展的基础，是地球系统大气圈、水圈、岩石圈及生物圈相互作用最复杂、最活跃的交界面。土壤中的微生物数量巨大、种类繁多，是联系不同圈层物质与能量交换的重要纽带，被称为地球关键元素生物地球化学循环过程的引擎。

土壤微生物在土壤中度过其全部或部分的生命历程，并在土壤内部各种过程中发挥着重要作用。土壤微生物参与次生矿物的形成，以及 Fe、Mn、Cu、S 等元素的生物地球化学转化过程[1]，土壤中各种来源和形态的有机质也都必须经过微生物的分解矿化过程才能重新进入土壤生物地球化学循环。大气温室气体的动态变化与土壤生物紧密相关。据估计，仅湿地和水稻田产甲烷菌引起的 CH_4 排放约占全球总排放量的 1/3[2]，而农田施肥相关过程所排放的 N_2O 约占全球年排放总量的 75%[3]。微生物在污染物的迁移转化过程中起着关键作用。土壤中有些微生物携带一些功能基因(如双加氧酶基因等)，其表达的蛋白是降解有机污染物的关键酶。有些微生物在长期进化过程中形成了以有机污染物为唯一碳源的生理代谢特点，通过降解污染物获得能量进行生长繁殖[4-5]。还有一些微生物通过共代谢(或共氧化)的方式降解有机污染物[6]。微生物也可以控制重金属的氧化还原及其相应的形态转化，如 Hg 和 As 的甲基化[7]。

在科技发展日新月异的新形势下，系统梳理土壤微生物学研究，有利于强化土壤微生物的知识积累与理论创新能力，充分理解其在土壤肥力形成和培育、污染土壤修复和全球环境变化中扮演着重要角色，从而为农业生产实践、全球环境变化和生态环境安全等国家战略需求提供新思路。

1　土壤微生物领域研究动态分析

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纤维素降解菌剂对桃园土壤养分及果实品质的影响

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摘要: 试验研究了桃园土壤在施用有机肥(牛粪)的同时施加纤维素降解菌剂,对土壤养分含量及桃果实品质的影响。结果表明,桃园基施有机肥及菌剂处理比单施有机肥处理,土壤有机质含量增加6.5%~29.9%,比空白对照高出1.3~1.8倍;施加菌剂使土壤速效氮、磷、钾均有不同程度的增加,其中土壤碱解氮增幅最大,达24.7%~27.0%;土壤速效磷含量增加12.1%~15.0%,土壤速效钾含量增加11.0%~15.0%,尤其是施加混合菌剂的处理效果最为明显。纤维素菌剂与有机肥混合处理,对桃单果重影响不显著,可显著提高桃果实品质。

关键词: 纤维素降解菌剂;果园;土壤养分;果实品质

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Influence of Cellulose Degrading Bacteria on Peach Orchard Soil Nutrients and Fruit Quality

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Abstract In this research, the effects of applying cellulose degrading bacterium agent in accompany with organic manure (cow dung) on peach orchard soil nutrient content and fruit quality were studied. The results showed that the soil organic matter content of basal application of organic manure and microbial agents increased by 6.5% ~29.9% than those of application of organic manure, and 1.3~1.8 times higher than those of CK. Application of cellulose degrading bacterium agent increased the soil available nitrogen, phosphorus and potassium to some extent. Among which, the available nitrogen had the largest increase amplitude of 24.7% ~27.0%, and that of soil available phosphorus and potassium was 12.1% ~15.0% and 11.0% ~15.0% respectively. The treatments with mixed bacterium agent had the most obvious effects. The single fruit weight of peach had no obvious change, but the fruit quality was significantly improved by mixed application of cellulose degrading bacterium agent and organic manure.

Keywords Cellulose degrading bacterium agent; Orchard; Soil nutrients; Fruit quality

自然界中有许多细菌、放线菌和真菌,大多具有分解纤维素和降解纤维素的功效[1]。近年来研究者在筛选纤维素降解菌方面取得显著成果,孟会生等[2]筛选出了哈茨木霉,具有较强的降解能力;郝月等[3]获得了降解纤维素较好的的青霉菌株;蔡兴旺等[4]从肥料中筛选出降解纤维素的单孢菌,这些菌株对研究纤维素降解及田间肥效起到非常重要的作用[5]。

随着农业的发展,作物赖以生存的土壤尤为重要,改善土壤结构、增加土壤有机质含量,合理高效施肥等研究成为主题。本试验在果园土壤施用有机肥(牛粪)的同时增加纤维素降解菌剂的

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食用向日葵盆栽种植土壤优化的初步研究

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摘 要 本研究以盆栽种植为桥梁,将向日葵的食用与观赏两大功能有机结合,从而更好地挖掘出向日葵的潜在价值。向日葵盆栽种植主要是克服盆栽土壤的结构和养分 2 个问题,试验结果表明,以“园土 + 蛭石 + 牛粪 + 草木灰”进行土壤配置,可为向日葵盆栽种植提供良好的生长环境。

关键词 向日葵;食用;观赏;盆栽;土壤配置

向日葵是一种生长发育迅速、物质积累快、抗逆性强并且有着食用和观赏两大功能的作物,基于它的诸多优点,广受欢迎[1]。但就目前市场调查来看,食用向日葵和观赏向日葵 2 个领域却相对独立,缺少将其两大功能相结合的应用,因此不管是食用向日葵,还是观赏向日葵,其市场价格都有一定的局限性。若能将向日葵的食用与观赏两大功能相结合,势必会扩大其市场需求。而食用向日葵盆栽种植研究正是基于这个思想,力求研究出食用向日葵盆栽的种植与管理方法,从而使得食用向日葵具备更高的观赏价值。

食用向日葵盆栽种植管理方便、节省种植空间,为近年来兴起的城市阳台和屋顶农业提供了很好的种植样板。食用向日葵盆栽种植与田间种植的主要区别在于土壤的养分和结构,盆栽中的土壤养分相对固定,且容易造成板结,可能导致向日葵发育不良、果实质量不好等问题。而盆栽中的水分、养分和光照等生长条件,便于根据向日葵的生理需求进行人工调控。因此,探究食用向日葵盆栽种植的优化问题,可以转化为探究土壤的结构优化和养分优化 2 个方面。我们选取生活中易于获得的 5 种优质廉价的材料作为盆栽用土的选择,它们分别为园土、蛭石、牛粪、草木灰及松针。根据这 5 种土质的性质[2-5],试验中,用园土作为盆栽土中的基本土壤,松针和蛭石作为盆栽种植中的土壤构性优化的土质,牛粪和草木灰作为土壤养分优化的土质。本试验通过探究 5 种土质间的不同搭配,观察向日葵的生长状况,从而得出最适合作为食用向日葵盆栽种植的土壤配置。

1 材料与方法

1.1 试验材料

本研究场地在山东农业大学试验站,所用的向日葵为美国黑贝食葵。美国黑贝食葵具有花盘大、果实饱满、观赏性较强、半矮化型等优良性状,并且经过市场及实地调查,它广受当地种植者的欢迎,因此,不管从其本身的性状,还是从实效性来看,都可作为本研究的良好实验材料。

盆栽种植的土壤选择,定位于生活中常见的优质廉价土,本试验采用了园土、蛭石、牛粪、草木灰及松针土 5 种土质。盆栽盆使用规格为:直径 35cm,高 28cm 的花盆。

1.2 试验方法

本试验以盆栽种植土的种类为变量,共设计了 14 组试验(见表 1),每组设 3 个平行试验。

表 1 试验对照组设置

处理	园土	松针土	蛭石	牛粪	草木灰
1	√				
2	√	√			
3	√	√		√	
4	√	√			√
5	√		√		
6	√		√	√	
7	√		√		√
8	√	√	√		
9	√	√	√	√	
10	√	√	√		√
11	√			√	√
12	√	√		√	√
13	√		√	√	√
14	√	√	√	√	√

1.2.1 用土量的确定。根据各种土质的性质及向日葵生长周期的营养特性需求,将盆栽盆的容量分为 7 等份,园土、蛭石、牛粪、草木灰及松针土的添加量分别占盆的 3 份、1 份、1 份、1 份及 1 份。

1.2.2 设计原则。(1) 以 1、2、3、4 组为一大组,探究在松针土作为土壤结构优化的情况下,牛粪与草木灰的作用情况。(2) 以 1、5、6、7 组为一大组,探究在蛭石作为土壤结构优化的情况下,牛粪与草木灰的作用情况。(3) 以 8、9、10 组为一大组,在松针土与蛭石协同作为土壤结构优化的情况下,比较牛粪与草木灰的作用。(4) 以 11、12、13 组为一大组,在草木灰与牛粪协同作为土壤养分优化的情况下,比较蛭石与松针土的作用。(5) 2、3、4 与 5、6、7 对照,分别在牛粪、草木灰作为土壤养分优化的情况下,比较松针土与蛭石对土壤结构的优化功效。(6) 14 组牛粪与草木灰协同优化土壤养分,松针土与蛭石协同优化土壤结构,以此来与各组进行对照。

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Identification and characterization of cherry (*Cerasus pseudocerasus* G. Don) genes responding to parthenocarpy induced by GA3 through transcriptome analysis

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Abstract

Background: Fruit set after successful pollination is key for the production of sweet cherries, and a low fruit-setting rate is the main problem in production of this crop. As gibberellin treatment can directly induce parthenogenesis and satisfy the hormone requirement during fruit growth and development, such treatment is an important strategy for improving the fruit-setting rate of sweet cherries. Previous studies have mainly focused on physiological aspects, such as fruit quality, fruit size, and anatomical structure, whereas the molecular mechanism remains clear.

Results: In this study, we analyzed the transcriptome of 'Meizao' sweet cherry fruit treated with gibberellin during the anthesis and hard-core periods to identify genes associated with parthenocarpic fruit set. A total of 25,341 genes were identified at the anthesis and hard-core stages, 765 (681 upregulated, 84 downregulated) and 186 (141 upregulated, 45 downregulated) of which were significant differentially expressed genes (DEGs) at the anthesis and the hard-core stages after gibberellin 3 (GA3) treatment, respectively. Based on DEGs between the control and GA3 treatments, the GA3 response mainly involves parthenocarpic fruit set and cell division. Exogenous gibberellin stimulated sweet cherry fruit parthenocarpy and enlargement, as verified by qRT-PCR results of related genes as well as the parthenocarpic fruit set and fruit size. Based on our research and previous studies in *Arabidopsis thaliana*, we identified key genes associated with parthenocarpic fruit set and cell division. Interestingly, we observed patterns among sweet cherry fruit setting-related DEGs, especially those associated with hormone balance, cytoskeleton formation and cell wall modification.

Conclusions: Overall, the result provides a possible molecular mechanism regulating parthenocarpic fruit set that will be important for basic research and industrial development of sweet cherries.

Keywords: Sweet cherry, GA3, Transcriptome, Parthenocarpy, Fruit set and cell division

Highlight

Cherry genes respond to parthenocarpy and promote fruit setting induced by GA3

Background

Fruit set is an important step in fruit growth and development. In this process, the ovary becomes enlarged with development of the embryo after fertilization, inducing fruit formation [1]. A variety of hormonal synergies play a major role in controlling fruit set; GA, auxin, and cytokinin alone cause the fruit to grow and develop to a certain stage, but the fruit develops normally due to their synergistic effect [2, 3].

Exogenous spraying of 5 mg / L GA3 maintains the activity of citrus parietal cells and promote their division, thereby increasing the rate of fruit setting [4], and it was reported that treatment of grape flower spikes with 30 mg/L GA3 before full bloom significantly increased the

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硫代腺苷甲硫氨酸促进番茄百菌清降解的生理机制

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摘要：【目的】探讨硫代腺苷甲硫氨酸（SAM）对番茄农药代谢调控的生理机制，为蔬菜安全生产奠定理论基础。【方法】本研究选取鲜食樱桃番茄‘千禧’为供试植物，以生产上广泛应用的广谱性杀菌剂‘百菌清’为供试农药，研究外源 SAM 对番茄果实喷施百菌清后的农药残留量、活性氧含量和谷胱甘肽代谢系统的影响。【结果】外源喷施 0.5—2 μmol·L^{-1}的 SAM 均能显著降低番茄果实的百菌清残留量；但当外源 SAM 处理浓度达到 0.5 μmol·L^{-1}后，番茄果实中的百菌清残留量并未随着处理浓度的增加而下降。与对照相比，喷施百菌清后番茄果实的谷胱甘肽 S-转移酶（GST）、谷胱甘肽还原酶（GR）和脱氢抗坏血酸还原酶（DHAR）活性显著上升；还原型谷胱甘肽（GSH）、氧化型谷胱甘肽（GSSG）和总谷胱甘肽含量（GSH+GSSG）变化也呈现上升趋势，谷胱甘肽的还原氧化比（GSH/GSSG）呈先上升后下降趋势；超氧阴离子（$O_2^{\cdot-}$）和过氧化氢（H_2O_2）含量显著上升，而丙二醛（MDA）含量与对照相比差异不显著。外源喷施 0.5 μmol·L^{-1}的 SAM 可显著提高百菌清处理下 GST、GR 和 DHAR 的活性，增加 GSH 和 GSH+GSSG 含量，降低处理 10 d 后的植株 GSSG 含量，提高 GSH/GSSG；提高处理 2—5 d 的植株$O_2^{\cdot-}$和 H_2O_2含量，但对 MDA 含量无显著影响。【结论】外源添加 0.5 μmol·L^{-1}的 SAM 可激活谷胱甘肽循环系统关键酶 GR 和 DHAR 活性，促进 GSH 再生，依赖 GST 并协同$O_2^{\cdot-}$和 H_2O_2的氧化作用，促进番茄果实中的百菌清降解。

关键词：番茄；农药代谢；硫代腺苷甲硫氨酸；活性氧；谷胱甘肽代谢

Physiological Mechanism of *S*-adenosylmethionine on Alleviating Chlorothalonil Residue in Tomato

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Abstract: 【Objective】 In order to provide basic mechanism for vegetable safe production, this study aimed at revealing the physiological mechanism of S-adenosylmethionine (SAM)-induced pesticide metabolism. 【Method】 The fresh-eating tomato 'QianXi' and broad-spectrum pesticide 'chlorothalonil, CHT' were selected as the experimental materials. Then, we studied the effects of exogenous SAM on the metabolism of pesticide, reactive oxygen species and glutathione. 【Result】 The results showed that the CHT residues were significantly reduced by exogenous spraying 0.5-2 μmol·L^{-1} SAM. However, no significantly changes could be observed when the concentrations of SAM arrived at 0.5 μmol·L^{-1}. Compared with control, the activities of glutathione S-transferase (GST), glutathione reductase (GR) and dehydroascorbate reductase (DHAR) were significantly increased under CHT treatment. And similar results could be obtained with the contents of glutathione (GSH), oxidized glutathione (GSSG) and total glutathione (GSH+GSSG). The GSH/GSSG were induced by CHT firstly, and then decreased. At the same time, superoxide anion ($O_2^{\cdot-}$) and hydrogen peroxide (H_2O_2) were significantly induced by CHT. However, no significant changes were observed in

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试验研究

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局部供磷条件下苹果幼苗根系形态的适应性变化及其对磷素的吸收*

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摘　要　以苹果砧木品种‘M26’的幼苗为材料，设置局部和均匀供磷处理，研究不同供磷方式下根系形态的适应性变化及其对磷的吸收。结果表明：相对于不施用磷肥处理，施用磷肥处理促进了根系和地上部的生长，以局部施磷处理差异显著；根系对供磷方式产生适应性变化，局部施磷显著提高了中层土壤的根系密度；局部施磷还提高了根际土壤磷酸酶活性和植株磷吸收量，其磷肥利用效率是均匀施磷处理的 1.53 倍。局部施磷条件下苹果幼苗根系形态表现出积极的适应性，优化了根系分布和根系参数，提高了土壤磷酸酶活性，改善了磷肥的生物有效性，最终提高了植株吸收磷养分的能力，提高了磷肥的利用效率。

关键词　苹果幼苗；局部供磷；均匀供磷；根系形态；磷素吸收

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Root configuration and phosphorus utilization response of apple seedlings to local phosphorus supply

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Abstract　Apple root stock cultivar ‘M26’ seedlings was used to understand the root configuration and phosphorus utilization response of apple seedlings to local phosphorus supply. The results showed that compared with no phosphorus application, the application of phosphate fertilizer significantly promoted the growth of root system, and the increase of local phosphorus supply was the largest. The root system configuration changed adaptively to different phosphorus supply modes, and the local phosphorus supply significantly increased the root density in the middle soil. Local phosphorus supply increased the activity of phosphatase in rhizosphere soil and phosphorus uptake in plants. Therefore, local source for phosphorus under apple seedlings root morphology table positive adaptation, root system distribution and the root parameters was optimized and improved soil phosphatase activity and improved the biological effectiveness of phosphate fertilizer, eventually improve the ability of plants to absorb phosphorus nutrient, and raise efficiency of utilization of the phosphate fertilizer.

Key words　apple seedlings; local phosphorus supply; uniform phosphorus supply; root morphology; phosphorus absorption

磷是果树生长发育必需的矿质营养元素，对促进花芽分化、提高果实品质和提高抗逆性具有重要作用[1]。目前，磷肥在我国苹果生产中的施用量较高，大部分产区果园土壤磷累积严重，土壤磷含量已超过树体需求量，环境风险较高[2]。由于磷非常容易被土壤固定，导致可被植株吸收利用的磷非常有限；再加上不合理的施磷方式，造成了当前磷肥利用率低的现状。为了满足苹果生长发育对磷的需

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Botany Characteristic of Different Apple Variety on M9T337 Rootstock
M9T337 自根砧嫁接不同苹果品种的植物学特性调查

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摘 要：该文对 M9T337 矮化自根砧嫁接 5 个富士短枝品种和金帅进行了调查，用 M9T337 自根砧嫁接短枝品种，栽后第 2 年结果，金帅（五莲）、龙富短枝（烟台）结果数分别是 16.8 个、14.9 个，龙富短枝结果较多。在不同试验地宫崎短枝结果数量不同，且双矮栽培在肥沃土壤上采用支架可实现矮化早丰。

关键词：龙富短枝；烟富 6；宫崎短枝；矮化；M9T337；自根砧
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Abstract　This paper explained the result of five spur apple varieties grafted on M9T337. Fruit number of Gold delicious （Wulian）and Longfu spur Fuji （Yantai ） were 16.8 and 14.9 respectively at second season after planting. Fruit number of Miyazaki spur Fuji were different in two regions. Spur Fuji grafted on M9t337 rootstock could bear fruit early on fertile soil with support system.

Key words　Longfu spur Fuji；Yanfu 6；Miyazaki spur Fuji；Dwarf；M9T337；Self－rooted rootstock

M9T337是荷兰木本植物苗圃检测服务中心从M9中选出来的矮化砧木优系，目前是世界各国应用最成功、最广泛的矮化砧木。苹果矮化集约栽培是倡导的一种生产模式，现代化矮砧密植苹果园多采用M9T337自根砧繁育的大苗来建园。矮化果树管理简单、用工少、树体结果早、果实着色好、含糖量高，是果树生产栽培者较为常用的栽培方法。M9T337苗木压条繁育比较容易，克服了 M9 压条不易生根的难题，也是M9T337砧木广泛应用的一个重要因素[1]。其砧木繁育为无性繁殖，保持母本的遗传性状，整齐一致，根系为茎源根系，没有主根，分布较浅。M9T337 作为砧木嫁接苹果树易成花，结果早，果个大小均匀，丰产性好，嫁接亲合性好[2]。嫁接风味偏酸的苹果 品种和嘎拉系列品种结果早，树势中庸，易管理[3]。有研究对比嘎拉、富士嫁接 M9T337 自根砧与乔化实生砧的产量及品质，发现嫁接 M9T337 矮化自根砧的单株产量极高，品质方面也优于乔化实生砧。意大利、法国等推广并已获成功的高纺锤形果园多采用M9T337砧木，荷兰、法国等国利用 M9T337 砧木将2年生自根砧成品苗于70～80 cm处短截，培育成分枝大苗，建园成形快、结果早，一般第 2～3 年即可获得高产[2]。山东省烟台地区和五莲县是苹果主产区，M9T337砧木是否适合山东的气候条件和土壤环境，笔者对两地的 2 年生 M9T337 自根砧苹果幼树的生长量、结果数量、根系分布进行实地调查。

1　材料与方法

1.1　材料

1.1.1　调查地点。选择位于山东省烟台市福山区的

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丽江市青贮饲用玉米高产栽培技术

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摘要:通过对丽江市青贮饲用玉米的高产栽培技术的研究,加大实施高产优质青贮饲料玉米品种及配套栽培技术的应用,最终使本地的青贮玉米进入规模化、标准化种植,对提高本区域内玉米种植结构的调整,带动全市青贮玉米生产及草食畜牧业的发展,促进全市粮、经、饲三元种植结构的构建发挥着重要作用。

关键词:丽江;青贮玉米;栽培种植

玉米是丽江市分布最广、种植面积最大的主要粮食作物,常年播种面积 58 万亩左右,玉米生产质量的好坏将直接影响全市主要粮食产量的稳定和提高,同时作为饲料用粮也将很大程度影响全市畜牧业生产的发展。当前种植业结构矛盾中,玉米生产表现出的供过于求、库存压力过大已成为突出问题。巩固提升优势产区、适当调减非优势区,调减籽粒玉米、扩大青贮玉米、适当发展鲜食玉米是当前调整的重点和方向。而加大实施高产优质青贮饲料玉米新品种及配套栽培技术的应用,可以有效减少牛羊等草食家畜的优质饲草料供需缺口、大幅降低生产成本,同时将单纯的粮仓变为“粮仓+奶罐+肉库”,同时也减轻了玉米收储压力。最终转变发展方式,巩固提升粮食产能,推进种植业结构调整,优化品种结构和区域布局,构建粮经饲统筹、农牧结合、种养加一体、一二三产业融合发展的格局。

1 选地整地

选择地势平整,土层深厚,肥力中等,排灌方便的地块。播前深犁细耙(耕深 25~30 cm),使之达到平整土碎、上虚下实、沟直墒平,内无残根、残膜。

2 品种选择

可选择蛋白质含量高,活秆成熟,抗病性好,抗倒伏的杂交玉米品种作为青贮种植,如:西抗 18,足玉 7 号等。

3 播种

3.1 种子质量

种子纯度≥96.0%,净度≥99.0%,发芽率≥85%,水分≤13.0%。种子质量应符合 GB 4404.1 规定指标。

3.2 播种量

每亩用包衣种 2~3 kg。包衣种使用应符合 GB 15671 规定指标。

3.3 播种期

3.3.1 春播 海拔 2 000~2 500 m 段的地区适宜春播,播期为 3 月 20 日至 4 月 20 日。

3.3.2 夏播 海拔 1 050~2 000 m 段的地区适宜夏播,播期为 4 月 20 日至 5 月 20 日。

3.4 播种密度

春播期亩播种密度为 6 000~6 600 株;夏播期亩播种密度为 5 500~6 000 株。

3.5 打塘播种

采用地膜覆盖宽窄行垄作法,大行距 65 cm、小行距 35 cm,打塘直播,每塘播 2~3 粒籽。春播期株距 40~44 cm;夏播期株距 44~48 cm。

4 施肥

以有机肥为主,无机肥为辅。重施基肥,氮磷钾合理配比,补施锌肥,适时追肥。肥料的使用应符合

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不同品种及栽培措施对青贮饲用玉米产量的影响

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摘要:本试验采用随机区组设计,研究不同青贮品种、不同种植密度、不同收获期对产量和品质的影响。研究结果表明:不同青贮品种、不同种植密度、不同收获期均能极显著影响青贮玉米的产量和品质。

关键词:青贮饲用玉米;品种;密度;产量;价值

青贮玉米较普通籽粒玉米具有生物产量高、纤维品质好、持绿性好、干物质和水分含量适宜用厌氧发酵的方法进行封闭青贮的特点。加大实施高产优质青贮饲料玉米新品种及配套栽培技术,可以有效减少奶、肉等家畜的优质饲草料供需缺口、大幅降低生产成本,将单纯的粮仓变为“粮仓+奶罐+肉库”,同时也减轻了玉米收储压力,有效推进种植业结构调整,优化品种结构和区域布局,构建粮经饲统筹、农牧结合、种养加一体、一二三产业融合发展的格局[1]。研究表明优质品种的选择、密度、收获期是影响青贮玉米产量和品质的三个重要因素[2]。为了探明青贮玉米在丽江复杂生态条件下种植的高产优质栽培技术,从2016年起,丽江市种子管理站率先在本区域开展了青贮玉米不同的品种、不同栽培种植密度、不同收获期对产量和品质的影响研究,旨在通过合理的栽培措施,提高青贮玉米的产量和品质,为丽江青贮玉米品种、优质栽培技术和合理利用提供理论依据与技术支持。

1 材料与方法

1.1 试验概况

试验地位于丽江市七河镇勒马村丽江市种子管理站实施的粮改饲标准化百亩示范区内。试验地地势平整,土层深厚,肥力中,排灌方便,前茬为冬闲田。

供试品种:足玉7号、西抗18、靖青1号、鑫白单7号、祥单3号、隆白1号、惠农三9号、惠农单5号、HN1701(黄粒)、陵玉987(黄粒)、金亿219(黄粒)、盛谷8号(黄粒),共12个。

试验在2018年4月8日播种,采用地膜覆盖宽窄行垄作法,行距1.2 m,人工拉线打塘精准直播,每塘双苗,播种深度4~5 cm。

1.2 试验设计

1.2.1 品种对比试验 试验采用随机排列,无需重复,每个小区面积96 m^2,10行区(8 m×12 m),小区间走道宽0.8 m,12个参试品种。采用地膜覆盖宽窄行垄作法,行距1.2 m,株距40 cm,打塘直播,每塘留双苗。

1.2.2 密度试验 ①参试品种:足玉7号。②小区设计:试验采用随机排列,设3次重复。每个小区面积72 m^2,3个密度处理,采用地膜覆盖宽窄行垄作法,行距1.2 m,株距根据密度而定,人工拉线打塘精准直播,每塘留双苗。③密度设计:3 500株(株距63 cm)、

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RESEARCH ARTICLE

Study on the microbial community in earthworm and soil under cadmium stress based on contour line analysis

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Abstract

Cadmium (Cd) contamination in soil has become the focus of widespread concern in society today. In this paper, with *Eisenia fetida* as research subjects, an indoor simulation experiment was conducted. A BIOLOG microplate technique was used to determine the carbon source (single-carbon) utilization of the microbial communities in the contaminated soil and earthworms under Cd stress. Contour line analysis was used for the first time to study the difference of carbon source metabolism in microbial communities. And the effects of Cd stress on the functional diversity of the microbial communities and the detoxification mechanism in earthworms were researched. With two test groups, a short-term test and the long-term test were performed. The former test lasted for 10 days, with the removal of an earthworm every day for analysis; the latter test lasted for 30 days, with the removal of an earthworm every 10 days. The Cd^{2+} concentration was set at 0, 50, 100, 125, 250, or 500 mg kg^{-1} dry weight, and 10 earthworms were inoculated in each concentration treatment. The earthworm homogenate and soil extracts were used to determine the carbon source utilization of the microbial communities. The results show that Cd stress changed the functional diversity of the microbial communities in the soil and earthworms. With the extension of stress time and the increase of stress concentration, earthworms will adjust their own physiological functions (including the microbial community structure and stress mechanism in the body) and regulate the microbial community structure in the external environment to obtain the necessary substances for growth. In addition, 2-hydroxybenzoic acid, γ-hydroxybutyric acid, glutamyl-L-glutamic acid, α-butyric acid, threonine, and α-cyclodextrin were important carbon sources for the earthworms to maintain their normal physiological metabolism under Cd stress. This study confirms that changes in microbial communities can be used to reveal the detoxification mechanisms of earthworm under heavy metal stress.

Keywords Cd stress · *Eisenia fetida* · Microbial community · Single carbon resource · Contour line analysis

Introduction

The research on the biological toxicity and detoxification mechanism of cadmium (Cd) pollution on soil animals is the focus and hotspot in ecotoxicology and agricultural ecology (Liu et al. 2013; Wei et al. 2015; Shen et al. 2017). The toxic effects of Cd stress on earthworms and its detoxification mechanism are mainly concentrated on individual body weight (Chen et al. 2017; Das et al. 2016; Zaltauskaite and Sodiene 2014), antioxidant enzyme activity (Li et al. 2014), cell membrane stability (Samal et al. 2017), and gene damage (Liu et al. 2011). For microbial community, current reports are only confined to the speculation and explanations of earthworm physiological changes after Cd stress. In the study of toxic effects of antibiotic stress on earthworms by Ji et al. (2014), they suggested that the reason of the part of gene expression disorder may be that the antibiotic has changed the microbial community in soil and earthworm, making the worms lose the function of obtaining the essential material or degrading harmful substances; the worms have to force itself to change so that genes can be expressed differently in order to acquire these functions. Kreps et al. (2002) and Kant et al. (2008) have similar inference in the study of salt, osmosis, and low temperature stress on the genetic damage of

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白菜 *CesA* 基因家族鉴定及表达模式分析

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摘要：为探究 *CesA* 基因家族在白菜生长发育及纤维素合成过程中的作用机制，该研究通过生物信息学的方法，以白菜的全基因组序列为研究区域，进行了理化特征、基因结构、进化特征、保守基序及结构域、顺式作用元件和组织表达等鉴定分析。结果表明:（1）白菜基因组中鉴定出 16 个编码纤维素合成酶亚基的 *CesA* 基因，该家族成员所编码蛋白的理论等电点介于 4.76~9.12，相对分子量 17.76~122.67 kD，长度 153~1 089 aa；（2）其中 15 个基因不均匀地分布于白菜的 7 条染色体上，*Bra*036008 定位于 scaffold 上；（3）大部分成员包含 4-14 个外显子，1-11 个保守基序；（4）序列比对显示该家族具有保守的 DDD-QXXRW 保守功能域；（5）该家族编码蛋白主要分布在质膜上，二级结构以无规则卷曲与α-螺旋为主，多数成员都含有 CesA 蛋白典型的 N 端、C 端和跨膜区；（6）*CesA* 基因在茎中表达量相对较高，其中 *Bra*011865、*Bra*023952 和 *Bra*029874 在茎、叶、花中显著表达。该研究利用生物信息学方法对白菜 *CesA* 基因家族进行了全基因组鉴定，为后续深入研究 *CesA* 基因功能奠定了基础，也为白菜生长发育研究奠定基础。

关键词：纤维素合成酶，白菜，*CesA*，基因家族，基因表达

Identification and expression analysis of *CesA* gene family in *Brassica rapa*

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Abstract Cellulose is the main component of plant cell walls. It is involved not only in cell morphology and development, but also in various cellular signal transduction pathways, thus

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生物促生剂在水和土壤治理中的应用分析

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摘　要：摘　要：结合生物促生剂的成分、机理及其特点，分析了其在造纸污水处理、天然水体治理、油脂含量较高的水体或土壤改善等方面的应用。

关键词：生物促生剂；实际生产；未来发展
中图分类号：X 52；X 53　　文献标识码：B
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引言

随着我国经济的快速发展和城市化建设的不断推进，城市生活污水排放量也在逐年增长。同时，在我国农村或者很多偏远地区，由于经济的落后或是高新技术手段的缺乏，导致很多地区的污水处理(如生活污水和工业废水等)存在较大问题，很多污水未能经过合理地处理便被排放到河流湖泊当中，这对水域乃至其周围的居民身心健康和土壤环境造成严重的影响和破坏，如何利用目前现有的技术手段能够低成本、快速、高效地解决此类问题，已成为当前业界研究和探讨的重点和热点。

1　生物促生剂

生物促生是修复生态的重要手段之一，其以促生技术(如利用有机酸携带的营养物质，通过生物和非生物的过程除去受污染环境的毒性并降解污染物的一类技术，20 世纪 70 年代首次应用于农业领域)为基础，再结合微碳技术(承担营养物质载体与高效输送任务，主要是运用小分子有机酸片段作为载体，使得营养物质吸收转化效率达到常规产品的 7～11 倍)将微生物生长所需要的各种营养成分与高效载体相结合，在污水环境中让微生物对污染源进行快速吸收和利用，从而提高系统对污染物的降解能力和抗冲击能力。

生物促生剂由有机酸、氮、缓冲液、酶、营养物质和能量等组合而成，能刺激污水处理系统中的好氧菌对废水中有机物进行分解，通过促进微生物的繁殖和能量代谢，提高微生物对污染物的氧化分解能力，并且能够屏蔽化学残留物对微生物的毒性，从而修复受污染的河道，进而提高污水处理系统的抗冲击性能和运行稳定性。同时，生物促生剂的使用还能增加物种的多样性，丰富群落结构，使多种微生物协同作用，更有效地降解污染物[1]。

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Analysis of Problems and Countermeasures in Supervision Mechanism of Prefabricated Buildings

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Abstract: This paper analyzes the problems existing in the supervision mechanism of prefabricated buildings from the perspective of supervision subject，scope and development direction，and puts forward the corresponding countermeasures.

Key words: prefabricated building；supervision subject；supervision mechanism；research on the problems；countermeasure analysis

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白菜 *DBB* 基因家族的鉴定与表达分析

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摘 要: *DBB* 基因在开花调控、根系生长、光形态建成、种子萌发、果实发育、逆境胁迫等生物学过程中发挥重要作用，但在白菜中还未有关于 *DBB* 基因的报道。本研究通过生物信息学方法鉴定到 18 个白菜 *DBB* 基因，其蛋白分子量为 11285.63~35796.93D，理论等电点介于 4.66~9.35 之间，外显子数量为 2~5 个；大多数成员 N 端都含有保守结构域 B-box1 和 B-box2，精氨酸和亮氨酸是在所有成员中都保守的氨基酸残基；该家族基因不均匀分布于白菜 10 条染色体上，多数基因出现基因复制，产生 2 个以上同源基因；多数基因与拟南芥 *DBB* 基因具有线性对应关系。顺式作用元件分析表明白菜 *DBB* 基因启动子上含有大量的光、厌氧、激素、低温等响应元件。组织特异性表达分析发现多数基因在不同组织中的表达水平与根类似；*Bra032441* 在花中的表达水平显著高于其它成员。此外，绝大多数白菜 *DBB* 基因对盐胁迫有不同程度的响应。这些结果为后续解析 *DBB* 基因在白菜中的生物学功能奠定了基础。

关键词: 白菜; DBB; 生物信息学分析
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Identification and Expression Analysis of *DBB* Family Genes in *Brassica rapa*

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Abstract: DBBs play an important role in plant flowering regulation, root growth, photomorphogenesis, seed germination, fruit development and abiotic stress response. However, *DBB* gene family has not been identified in *Brassica rapa*. In this study, 18 *DBB* genes were identified by bioinformatics method. The molecular weight was 11285.63 ~ 35796.93 D, the theoretical isoelectric point ranged from 4.66 to 9.35, and the number of exons ranged from 2 to 5; The N-terminus of most members contained the conserved domains B-box1 and B-box2, and the arginine and leucine were found as the most conserved amino acid in all DBBs. The DBBs were distributed unevenly on 10 chromosomes, and most of the genes were duplicated, producing more than 2 homologous genes. Most of the genes had linear correspondence with *Arabidopsis DBB* genes. The cis-element analysis showed that the *DBB* gene promoter contained a large number of light, anaerobic, hormonal and low temperature response elements. Tissue expression analysis showed that the expression level of most genes in different tissues was similar to that in the roots, interestingly, the level of *Bra032441* in flowers was remarkably higher than that in other members. In addition, most of the *DBBs* in *Brassica rapa* respond to salt stress. These results laid a foundation for further analysis of the biological functions of *DBB* genes in *B. rapa*.

Keywords: *Brassica rapa*; *DBB*; bioinformatic analysis

锌指蛋白是真核生物中的一类具有锌指结构域的转录因子家族[1]，其中含有 B-box 基序的蛋白称为 BBX，在拟南芥中有 32 个成员，其 C 端通常含有 CCT 结构域[2]。含有 CCT 结构域的蛋白通常与光周期有关，在植物中与成花相关基因一起调控开花时间[3]。但 BBX 中有一类缺乏 CCT 结构域的蛋白，称为 DBB（double B-BOX），其突出特点是 N 端具有两个或以上的 B-Box 结构域且 C 端缺乏 CCT 结构域[4]。

植物 DBB 家族在逆境胁迫（盐胁迫、温度胁迫、干旱胁迫等）、开花调控、光形态建成、根系

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Deciduous Fruits

·综合评议·

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桃砧木耐涝性研究进展

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摘 要:综述了桃砧木对涝害胁迫的生理响应指标及已有研究报道中涉及到的30多种砧木对涝害胁迫的响应特性;分析了桃砧木耐涝的内在机理;总结了多种国内外耐涝性较强的桃砧木种类。

关键词:桃;砧木;耐涝性

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A review of waterlogging tolerance of different peach rootstocks

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Abstract: This paper summarized the physiological response indexes of peach rootstocks to waterlogging stress, and the response characteristics of more than 30 kinds of rootstocks involved in existing research reports to waterlogging stress, analyzed the internal mechanism of waterlogging tolerance of peach rootstocks, and summarized a variety of peach rootstocks with strong waterlogging tolerance at home and abroad.

Key words: peach; rootstock; waterlogging tolerance

桃(*Prunus persica*)为蔷薇科落叶乔木,浅根性植物,根系呼吸旺盛、耗氧量大,不耐涝。中国南方亚热带和热带季风气候区年降雨量大,地下水位高、雨水过多或排水不良的地区,桃树常发生落叶、落果,甚至涝死树现象[1]。研究表明,砧木对改变接穗长势、延长树龄和抵抗病虫害有重要影响[2],生产上通常将良种嫁接在抗性砧木上,以提高良种对逆境胁迫的忍耐性[3]。果树砧木对改善果实品质和扩大良种适栽区有较大的影响。所以研究桃砧木的耐涝性及其生理基础,筛选优质耐涝砧木种质资源进行区域化推广,对提高桃生产的水平和效益[4-6]意义重大。

1 桃砧木对涝害胁迫的生理响应指标

植物受涝害的最直接器官是根系[7],表现主根伸长速度减慢,颜色逐渐发黑,根毛减少。淹水造成土壤低氧,根际缺氧会抑制根系的有氧呼吸,使根系缺乏能量,抑制根系对水分和矿质营养的吸收,影响根系活力。在缺氧条件下,根系无氧呼吸产生的乙醇、乙醛等中间产物,以及厌氧微生物活动生成的有机酸、还原性物质均会对树体造成毒害[8,9],严重时会导致根系的凋亡[10]。

果树受涝害的直接可见器官反应是叶片黄化、脱

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外源硅处理对草莓果实果胶物质降解的影响

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摘要: 果胶是细胞壁初生壁和中胶层的主要成分, 影响果实的品质。为探究外源硅处理对草莓果实细胞壁降解影响, 试验以‘章姬’草莓(*Fragaria* × *ananassa* ‘Akihime’)为试材, 研究叶面喷硅对草莓绿果期、白果期、转红期及全红期果实细胞壁组分(果胶、原果胶)、细胞壁降解酶[多聚半乳糖醛酸酶(PG)、果胶裂解酶(PL)、纤维素酶(CX)]活性以及PG、PL、果胶甲酯酶(PME)、β-半乳糖苷酶(β-Gal)和内切葡聚糖酶(EG)编码基因表达的影响。结果表明, 外源1 g·L^{-1}流体硅处理, 可溶性果胶含量, PG、PL、CX活性以及*FaPG*、*FaPL*、*Faβ-Gal*、*FaEG*基因的表达量在绿果期、白果期、转红期均高于对照; 外源5 g·L^{-1}流体硅处理后, 可溶性果胶含量, PG、PL、CX活性以及*FaPG*、*FaPL*、*FaEG*基因的表达量在转红期、全红期均低于对照。综上所述, 外源5 g·L^{-1}流体硅处理对延缓细胞壁组分代谢的效果最佳, 具有较高的应用价值。

关键词: 草莓; 硅; 细胞壁降解; 果胶

Effect of exogenous silicon treatment on pectin degradation of strawberry

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Abstract: Pectin is the main component of the primary wall and middle layer of cell wall, which affects the quality of fruit. To explore the effect of exogenous silicon treatment on cell wall degradation of strawberry

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干旱过程中‘M9T337’苹果砧木苗光合特性及MdCP2与MdGLK1互作分析

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摘要: 苹果半胱氨酸蛋白酶基因*MdCP2*在植株干旱过程中发挥着重要的作用。本研究以一年生‘M9T337’苹果砧木苗为试材, 从生理和分子生物学水平分析了苹果砧木苗在自然干旱及复水过程中叶片光合特性、植株含水量、半胱氨酸蛋白酶基因*MdCP2*及相关基因的表达分析以及*MdCP2*基因的生物学功能。结果表明, 水分胁迫使植物的光合速率、气孔导度和蒸腾速率下降。在自然干旱后期, 植株光合作用的主要限制因素由气孔限制转变为非气孔限制。实时荧光定量PCR发现, 在正常培养的苹果树中, *MdCP2*基因主要在根中表达; 且在干旱过程中, *MdCP2*的表达水平呈上升趋势, 而在复水后降低。*MdRCA*、*MdCAB*和*MdRBCS*表达在干旱时降低, 复水后上调。通过酵母双杂交和BiFC试验证明, MdCP2与MdGLK1存在相互作用, 为进一步研究半胱氨酸蛋白酶基因*MdCP2*提供了理论基础。

关键词: 苹果砧木; 干旱胁迫; 半胱氨酸蛋白酶; MdCP2; MdGLK1

Photosynthetic characteristics of ‘M9T337’ apple rootstock seedlings and interaction between MdCP2 and MdGLK1 during drought stress

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果树耐盐机制研究进展

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摘 要:盐胁迫是影响果树栽培的重要环境因子,严重制约我国果树产业的发展。主要对果树耐盐性评价方法和耐盐种质资源筛选、果树耐盐性形成的生理机制、果树耐盐性状的遗传及相关基因等研究现状进行了概述,为后续的果树耐盐碱机制解析及耐盐碱果树新品种培育提供参考。

关键词:果树;耐盐机制;研究进展
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盐胁迫是影响植物生长和发育的重要环境因子之一。近年来,随着工业化进程的加速,耕地面积急剧减少,盐渍化程度显著增加。生产过程中过度施肥及不恰当的灌溉管理,使得土壤盐渍化问题变得日益严峻[1]。

土壤盐渍化已严重影响到我国主要经济树种——果树的发展,影响果品生产的产量和品质。本文对目前果树耐盐机制研究进行了综述,以期为后期抗盐性研究与耐盐育种提供参考。

1 果树耐盐性评价方法和耐盐种质资源筛选

1.1 评价指标

果树耐盐性可以通过测量和计算盐胁迫后各生长指标变化进行评价。常用的生长指标包括植物的植株生长量、株高、叶片数、根系活力及根干鲜质量等[2-5]。

在盐胁迫下变化幅度较为明显的生理生化指标也可作为果树耐盐性评价的指标。常用的生理生化指标包括叶绿素、脯氨酸、丙二醛等物质的含量,钠离子、钾离子质量分数及其比值,过氧化物酶等氧化酶的活性及相对电导率、净光合速率等[6-9]。

果树耐盐机制复杂,通过单一指标或几个指标评价果树的耐盐性,往往具有较大的片面性,无法科学地进行评价,通常对果树各项指标进行分析,从而对其耐盐性进行综合评价[10]。采用综合评价的方法,能够较为全面地评价不同种质间的耐盐性,提高种质表型鉴定的准确性。如在形态指标的选择上,通常采用“盐害指数”这一综合指标,也经常使用平均抗逆系数、耐盐系数等对试验材料进行耐盐性评价。曾丽蓉为评价 5 种不同苹果砧木耐盐性,测定了 14 个与耐盐性相关的指标,与耐盐性呈正相关的指标,计算隶属函数值;与耐盐性呈负相关的指标,计算其反隶属函数值,最后根据隶属函数平均值的大小排序。郁万文等以叶片细胞膜相对透性、可溶性糖和游离脯氨酸含量、SOD 和 POD 活性、叶中 Na^+ 和 K^+ 含量为依据,计算各指标的隶属函数值来综合评价供试桃砧种质的耐盐性。综合各项指标计算隶属函数值的方法比选用某一具体指标更科学合理,此种方法在葡萄、柑橘、梨等大宗水果上也经常应用。研究者进行了积极探索,发现综合各项指标计算隶属函数值的方法比选用某一具体指标更科学合理。

1.2 培养方法及时期

筛选果树耐盐植株的培养方法主要包括土培法、水培法和组织培养 3 种。对耐盐性材料的处理时间,则根据果树种类和试验目的不同而异。如朱世平等为评价 15 种柑橘砧木出苗期耐盐碱性,于播种期对柑橘种子进行盐处理。刘育梅等选用苗龄两年的神秘果为试验材料,来探究 NaCl 胁迫下其叶片的生理响应。

1.3 离子种类及浓度

离子的种类及浓度不同,对果树的伤害性也不同。一般碱性盐比中性盐、高浓度比低浓度对果树的危害往往更大。离子处理种类主要分单盐处理和混合盐碱处理两类。单盐处理一般设置不同浓度梯度的 Na^+ 进行试验,混合盐碱处理则大多根据当地盐碱地主要盐分组成特点进行处理,以期为解决当地盐胁迫问题而服务[11]。

1.4 种质资源筛选

对盐胁迫的适应性在植物不同物种之间、同一物种的不同栽培品种之间,甚至在同一栽培品种的不同个体之间是可变的。通过筛选,明确不同品种间的耐盐性差异,可为果树育种提供更为广泛、优质的遗传资源。Latifa A K 等[12]筛选出耐盐品种 Manoma 和 Umsila,并将其作为遗传资源,提高了椰枣对盐的耐受性。夏思哲等[13]通过对野生葡萄“燕山 -1”×“河岸 -3”种间杂交 F1 代植株的初步筛选,鉴定出 11 个高耐盐的杂种 F1 代株系,作为优良的砧木材料组培苗。

2 果树耐盐性形成的生理机制

2.1 离子区室化

离子区室化是植物抵御盐害的一个重要策略。研究表明,在盐胁迫过程中,积累的高浓度 Na^+ 对植物细胞产生显著伤害,使植物的生长发育受到严重抑制。Agrawal 等[14]研究表明,通过用不同浓度的 NaCl 处理枣品种,发现植物叶片 K^+ 的积累和抑制 Na^+ 从根部向叶片运输,可提高枣树耐盐性。

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Influence of drought and dry-wet alternation on nitrogen transformation and low abundance microorganisms in tea garden soil

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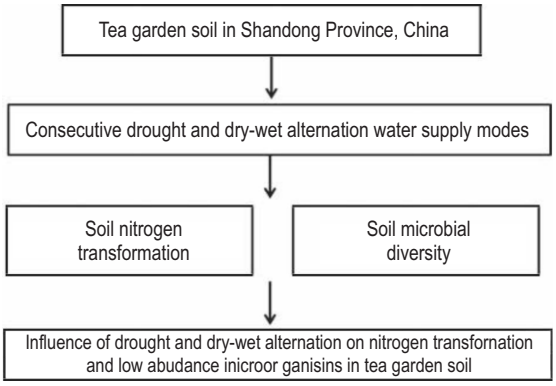
Abstract

Aim: The soil water availability seriously limits the growth and development of tea plants, and soil microorganisms are an important medium to regulate soil nutrient cycling. In this study, the effects of water supply mode on soil nitrogen nutrition and soil microbes in tea gardens were investigated.

Methodology: This experiment set up consisted two water supply modes (consecutive drought and dry-wet alternation) by using the soil microcosm incubation experiment, and four treatments were set: 20% water holding capacity for 21day (D21); 20% water holding capacity for 1-7 days and 60% water holding capacity for 8-21 days (D7W14); 20% water holding capacity for 1-14 day and 60% water holding capacity for 15-21 days (D14W7); 20% water holding capacity for 1-7 days, 60% water holding capacity for 8-14 days, 20% water holding capacity for 15-21 days (D7W7D7). Destructive sampling was carried out to determine soil NH_4^+-N, NO_3^--N, soil enzyme activities. 16S rRNA sequencing technique was used to determine the change in soil microbial diversity.



Results: The results showed that the consecutive drought reduced the content of soil NH_4^+-N to 13.97 mg kg^{-1}, and the net nitrogen mineralization was negative (-2.75 mg kg^{-1}) after 21 days of incubation. Dry-wet alternation promoted the increase in of soil net nitrogen mineralization quantity and net nitrification quantity, which rose to 3.48-26.41 mg kg^{-1} and 8.07-23.11 mg kg^{-1}, respectively. Different water supply modes had no significant impact on the structure of dominant soil microbial community, and the effect mainly focused on relative abundance, especially dry-wet alternation mode. Compared with the continuous drought treatment, the relative abundance of *Nitrospirae, Actinobacteria, Chloroflexi, Patescibacteria, Latescibacteria, Rokubacteria, Acidobacteria* were significantly different in different dry-wet alternation treatments, while the relative abundance of *Nitrospirae, Acidobacteria, Latescibacteria, Gemmatimonadetes, Patescibacteria* and *Chloroflexi* also increased or decreased significantly among different dry-wet alternation treatments. Among the physical and chemical factors of tea garden soil, NO_3- had the most significant effect on the structure of microbial community.

Interpretation: Different water supply can significantly affect the transformation of soil nitrogen and the change in soil bacterial community in tea garden, which provided a theoretical basis for tea garden to cope with adverse weather changes and maintain the stability of tea garden soil ecosystem.

Key words: *Camellia sinensis*, Nitrogen transformation, Soil microorganism, Water regime

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ORIGINAL ARTICLE

Identification of potential pathways associated with indole-3-butyric acid in citrus bud germination via transcriptomic analysis

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Abstract

Indole-3-butyric acid (IBA) is widely used to encourage root development in cuttings of general field crops, vegetables, forest trees, fruit trees, and flowers. However, previous studies reported that IBA inhibited the germination of citrus buds via an unknown molecular mechanism. This study aimed to unravel the regulatory mechanisms underlying this inhibition. Citrus apical buds were sprayed with 100 mg · L^{-1} IBA. Subsequently, the plant hormone levels were analyzed, and transcriptomic analysis was performed. The results identified 3325 upregulated genes and 2926 downregulated genes in the citrus apical buds. The gene set enrichment analysis method was used to determine the Gene Ontology related to the treatment. Genes were enriched into 157 sets, including 17 upregulated sets and 140 downregulated sets, after indole butyric acid treatment. The upregulated gene sets were related to glucose import, sugar transmembrane transporter activity, and photosynthesis. The downregulated genes were mainly related to the ribosomal subunit and cell cycle process under butyric acid treatment. Kyoto Encyclopedia of Genes and Genomes pathway enrichment analysis revealed the enrichment of 11 pathways. Of note, genes related to the ribosome and proteasome pathways were significantly downregulated. Only one pathway was significantly upregulated: the autophagy pathway. Overall, these results provided insights into the molecular mechanisms underpinning the IBA-mediated inhibition of citrus bud germination inhibition. Also, the study provided a large transcriptomics dataset that could be used for further research.

Keywords Citrus · Bud · Gene set enrichment analysis · Indole butyric acid · Transcriptome

Introduction

Previous studies showed that indole-3-butyric acid (IBA) was converted into auxin (IAA) during peroxisome β-oxidation. Also, IBA promoted the growth and development of plant roots. Specifically, it significantly increased the rate of rooting in *Arabidopsis thaliana* (Fattorini et al. 2017), citrus (Yang et al. 2020), *Conocarpus erectus* (Abdel-Rahman et al. 2020), avocados (Umair et al. 2018), and *Laurus nobilis* (Saeed and Amin 2020). Interestingly, a previous study revealed that the treatment of citrus with a specific concentration of IBA significantly inhibited bud germination (data not published). Other studies reported that in the auxin transport model, auxin was secreted by the shoot tip and subsequently entered the axillary bud and inhibited the shoot tip growth. The axillary bud subsequently germinated once sufficient auxin flowed out of the bud (Leyser 2011; Brewer et al. 2015; Lv et al. 2017). We hypothesized that exogenous IBA treatment caused an increase in the auxin concentration in the citrus buds, which subsequently caused the inhibition of germination. However, the specific molecular mechanism underlying this inhibition is yet to be elucidated.

In *Arabidopsis*, glucose conjugation maintains auxin metabolic balance due to the chemical properties influenced by glycosylation (Damodaran and Strader 2019). A previous study showed that glucosyltransferase UGT74D1 affected leaf position by regulating auxin homeostasis and the transcription of polyketide synthase as well as transcription factors possessing *TEOSINTE BRANCHED1, CYCLOIDEA*, and *PCF* (TCP) domain (Jin et al. 2021). In addition, UGT84A2 encoded an indole-3-butyrate glucosyltransferase,

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三种成膜材料对‘赤霞珠’枝条的保水效果研究

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摘　要：我国北方地区春季葡萄抽条现象普遍发生，给生产带来严重影响。试验以‘赤霞珠’1年生枝条为试材，研究壳聚糖（CTS）、羧甲基纤维素钠（SCMC）、海藻酸钠（SA）3种成膜材料对枝条的保水效果。结果表明，同其他品种相比，‘赤霞珠’枝条失水严重，经历越冬季后的失水率为13.46%。采用不同成膜剂处理均能提高‘赤霞珠’离体枝条的持水能力，其中0.3% SA效果最好。喷施成膜剂可以减少越冬期‘赤霞珠’枝条含水量的下降率，保水效果由高到低依次为0.2% SA＞0.2% SCMC＞0.2% CTS。试验表明，采用成膜剂处理能够有效降低‘赤霞珠’枝条水分的散失。

关键词：赤霞珠；枝条；失水；成膜材料

Study on Water Retention Effect of Three Film-Forming Materials on 'Cabernet Sauvignon' Annual Branches

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Abstract: The phenomenon of grapevine spring dieback is common in the north of China in spring, which has serious influence on grape production. In this experiment, annual branches of 'Cabernet Sauvignon' were used as test materials to investigate water retention effects of three kinds of film-forming materials: chitosan (CTS), sodium carboxymethyl cellulose (SCMC) and sodium alginate (SA). The results showed that compared with other varieties, 'Cabernet Sauvignon' annual branches had severe water loss, with a water loss rate of 13.46% after the winter. Treatments with different film-forming materials could improve water holding capacity of isolated canes of 'Cabernet Sauvignon', and 0.2% SA has the best effect. Spraying film-forming materials could reduce water content of 'Cabernet Sauvignon' annual branches during overwintering period, and the effect from high to low is 0.2% SA ＞0.2% SCMC＞0.2% CTS. Experiments showed that the treatment with film-forming materials could effectively reduce the water loss of 'Cabernet Sauvignon' annual branches.

Key words: Cabernet Sauvignon; annual branch; water loss; film-forming material

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以“粮改饲”为主线，促进绿色供给侧结构性改革向纵伸发展

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摘要：为了加快农业供给侧结构性改革步伐，以“粮改饲”为主线，加大青贮饲用玉米推广力度，结合工作实际总结出一些经验，最终实现种植养殖相结合、农机农艺相配套、传统绿色共融合的发展目标，促进全市粮食生产绿色增产模式进一步发展。

关键词：粮改饲；青贮饲用玉米；绿色；亮点

“以推进农业供给侧结构性改革为主线，围绕农业增效、农民增收、农村增绿，加强科技创新引领，加快结构调整步伐，加大农村改革力度，提高农业综合效益和竞争力，推动社会主义新农村建设取得新的进展，力争使农村全面小康建设迈出更大步伐”[1]。作为2017年中央1号文件提出的主要思路之一，“农村增绿”是一个新名词，更是一种新理念。丽江作为一个地处偏远的高原地区，“农村增绿”与供给侧结构性改革看起来似乎很遥远，但现实中生态的脆弱、环境的进一步恶化、结构性供应过剩更能给地方经济和社会发展造成更大的冲击和波动。如何科学的规划和制定绿色供给侧结构性改革的发展思路也成为当代农业人肩负的重要历史使命。丽江市农业局在认真解读中央1号文件的同时，紧紧围绕各级政府提出的农业发展思路，结合丽江农业发展现状，科学地做出了顶层设计。首先抓住“粮改饲”这条主线，逐渐实现了种植与养殖相结合、农机与农艺相配套、传统与绿色共融合的目标。通过农业相关部门两年多的努力，青贮饲用玉米绿色种植示范推广模式已取得了一定成效，促进全市农业供给侧结构性改革向纵伸发展。

1　结合实际，制定主线，推进农业供给侧结构性改革

1.1　领导重视，科学顶层设计

在2016年全市农业工作会议上，丽江市农业局局长和建华同志在传达丽江农业整体发展思路中，分析了如何充分利用当地草食畜牧的良好发展基础和丰富的资源优势，加大青贮饲料玉米新品种引种示范力度，是推进全市草食畜牧业产业发展，实现籽粒玉米向饲用玉米转变的一个有效途径。要求市种子管理站充分展示优质高产多抗青贮玉米品种生产应用技术成果，加大与畜牧站所、农机推广站和养殖大户合作，切实做好青贮玉米品种样板展示推广工作，以“粮改饲”工作为主线，推进全市农业供给侧结构性改革。

1.2　职能优势互补，形成发展合力

按照“三定、三配套”（定人员、定标准、定产量，良种良法配套、农机农艺措施配套、种植养殖联合配套）的要求，在样板示范点建设中实行领导、技术指导挂钩责任制。种子、畜牧、农机各站所实行分工协作，各相关人员严格履行工作职责，按照“五有、四统一、三到位”的示范标准抓好青贮饲料玉米样板展示推广工作，即：推广示范区建设有组织领导、有技术

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土地利用规划环境影响评价的相关问题分析

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【摘要】随着社会主义市场经济的发展速度越来越快，近几年城市化进程的加快可谓是无法阻挡，而我国虽然地大物博，但针对土地资源在选择的过程中也存在着日渐紧缺这一现状，土地利用规划对整个城市地区的生态环境、经济发展而言，都带来了十分重要的影响。在这种情况下，需要高度重视土地规划的环境评价、影响工作、分析土地规划，利用整个环境发展所带来的不同影响，更好地开拓土地的利用、规划现状，确保社会经济的发展能够逐步走向健康的状态。

【关键词】土地利用规划 环境影响 评价

引言：土地资源一直以来都是有限的，想要在当前高度开发一个地区的土地资源，并且有效地对该地区的土地进行规划，首先要了解的就是土地的利用以及总体规划是在一定的区域内，需要结合不同地区的自然条件、社会经济条件以及整体的经济发展三者相结合，整体协调土地供给以及该地区的土地总需求，确定或调整土地利用结构以及用地布局的宏观战略。土地利用规划评价影响也直接影响到了该地区对土地的应用以及该地区生态环境，需要做好综合评价，及时地考虑到其中可能存在的生态问题，针对这些问题进行分析，将评价结果最终提供给土地规划部门进行分析和参考，选择最具有针对性的措施，才能够确保社会发展中土地利用具有科学性以及有序性。

一、土地利用规划环境影响评价的原则

在进行土地规划环境评价影响时，首先要明确七，评价内容的原则是什么，该原则是评价准确的基础，而评价内容不能够随便选取，也需要按照一定的原则、一定的规范进行选取。第一个原则就是土地利用规划的类型。任何一个城市地区其土地的利用类型是存在一定的区别的，例如有一些地区的土地规划是用来建造民房，而有一些则是建造工厂，不同的利用目的也会导致其对周围环境所带来的影响各不相同，再进行评价内容的选取时，首先要明确的就是该土地的使用目的，尽可能地明确这一目的对于环境所带来的影响，并且根据目的进行环境问题的选择评价内容。第二原则是土地利用规划所导致的环境问题，不同的土地由于其所在地理位置以及自然条件不同，在进行土地规划时，规划的内容、方式也不同，所导致的环境问题也不同，这就要求政府在进行选择评价内容时，对该规划有着更为明确且清楚的认知，能够清晰地了解和估计出该环境可能出现的环境问题有哪些，并且根据这些问题作为导向，以导向为基础制定相应的评价内容以及评价方法，提高该地区的环境保护整体效果。第三个原则是要求土地利用规划的空间体系能够与城市发展相适应。我国土地利用规划体系分为五个不同的等级，分别是：全国、省、市、乡、镇而不同的等级在开展土地利用规划时，其内容也存在着一定的区别，而随着等级越来越高，所考虑到的土地规划评价也愈发地有深度，其不仅仅要考虑到环境内容，与此同时，也需要考虑到当时所处在的空间，一旦进行土地规划其所带来的社会影响、经济影响等等不同的内容。为此，一定要确保土地利用规划与空间体系相适应，才能够提高该规划的整体效果。第四个原则是在进行土地利用规划时，也需要考虑到该规划后可能会对生态环境所带来的不同影响，我国在开展土地利用规划时，其目的之一就是解决我国目前相对较为严重的生态环境问题。在土地利用规划时，任何一个规划内容都是以这一目的作为最基本的原则进行服务，进行评价内容的选取时，一定要了解到该土地利用规划所带来的生态环境问题有哪些。

二、土地利用规划环境影响评价的内容

土地利用规划本身对环境的影响是多方面的，无论是好的还是坏的，土地利用都会给环境带来非常明显的改变，只是一种不可逆转的改变。为此，在对环境影响评价内容确定时，也需要从多方面进行考虑。第一土地利用规划对土壤本身所带来的影响，土地利用对土壤的影响可谓是最直接的，例如如果是农业用地，对土壤的影响非常大，农业用地会导致土壤本身的营养出现一定的破坏，而各式各样不同的人类活动也改变了土壤的盐碱化。随着技术的飞速发展，近几年在农业用地过程中，由于大量地使用农药，也对土壤带来了一定的负面影响。第二则是对水环境所带来的影响，土地利用对水资源存在着非常明显的影响，其中主要是从两方面的土地利用进行分析，分别是农业用地与工业用地，农业用地对水资源的影响主要体现在河流、湖泊形状的改变以及对城市水源地掠夺方面，工业用地则是对水资源影响。工业用地在进行水资源影响的过程中，其主要表现在废弃物以及污染物的排放方面，其极大地污染了水资源的使用。第三则是对生态系统的影响，土地本身是由多种不同的生态系统组成的一个相对较大的生态系统，而不同的土

城市规划与土地利用规划衔接问题分析

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摘要：城市化进程加速，对城市规划管理工作的有效性提出了更高要求。要想实现我国土地资源的高效利用，还需放大土地利用利用与城市规划间的密切关系，构成两方面相得益彰的协调局面。本文主要对城市规划与土地利用规划的衔接进行阐述，分析衔接中的问题，提出建设性的整改对策，希望对土地资源配置工作的转型升级起到积极参照作用。

关键词：城市规划；土地利用规划；衔接；问题

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城市规划是指城市发展的计划与战略部署。土地利用规划是确保土地资源合理开发利用而展开的一种技术或工程等措施的长期土地的配置或布局。我国虽然土地资源丰富，但用于农耕与城市建筑的用地面积相对较小，还需综合各地区的用地需求进行统筹安排，以推动社会的可持续发展。土地利用规划与城市规划存在局部和点的关系，城市规划则是土地利用规划的一部分，而后者需从整个区域的长期发展入手考虑，难免会出现衔接性的问题，要想实现两者间的协调发展，还需加强研究。

一、城市规划与土地利用规划间的关系

1.基本任务

首先从土地利用总体规划的基本任务入手分析，要想实现城市土地资源的合理利用，推动社会经济的有序发展，还需从全局的角度出发，充分考虑土地的长远持续利用目的，加强耕地面积保护的同时，采取统筹协调的方式，综合城市发展中的用地需求，科学划分各种规模的布局与面积。由此可见土地利用规划工作展开的重要意义。其次从城市规划的基本任务入手分析，主要是指整体规划城市的未来发展方向，具体安排城市的各项建筑，通过规划各种用地的建设地点与面积大小，满足用地需求的同时，实现建设上流线管理问题的有效协调，从而推动城市社会经济迅猛发展。

2.关系

在定义城市规划时，需参照城市发展中的相关内容，因为城市规划的主体，主要是对满足城市发展所需而提供服务，有目的的构建推动城市经济进一步发展的合理计划。城市规划涉及的内容广泛，需合理安排各项建设发展计划，从而为城市经济稳定发展提供基础保障。对土地利用规划的定义，需从国家的角度出发，要求基本生产用地能够满足居民生活的需要，再合理分配剩余的土地资源，实现土地资源的高效开发利用，实现土地资源利用的效益最大化，从而切实发挥土地开发利用对社会经济发展的推动作用。土地利用规划与城市规划间，存在面与点、总体与分支的关系。城市规划的工作开展，为土地利用规划提供服务，任何城市规划涉及到的内容，都需在土地利用规划的基础上展开。但实际上，受规划工作人员的素质水平与各部门的经济利益等因素影响，两者间的关系尚未明确，在关系处理中存在诸多问题，尤其是各管理部门为获取自身的利益而出现的土地资源不合理征用的问题，会对各项工作展开与社会经济的稳定发展带来一定的阻碍即影响。

二、城市规划与土地利用规划间的衔接问题

土地利用规划是土地利用方面的长期指导，土地利用规划于城市利用规划均是整治国土开发及深化城市建设的策略。但实际上，当前的城市规划在建设中存在诸多问题，包括城市规划规模盲目扩大，脱离城市定位等实际情况；布局方面缺乏空间的合理性，导致城市特色不突出，影响城市的稳定长远发展。除此之外，在第二轮土地利用总体规划的编制工作中，由于各城市的上层国土规划的划分不明确，土地规划缺乏丰富的理论指导，导致土地利用规划与城市规划间的矛盾突出。

1.研究侧重点间的差异

从土地利用规划工作入手分析，其目的是在确保耕地面积的基础上，增加可利用田地，实现土地资源的高效利用。在田地重组工作中，需从整体发展的角度进行规划与统筹发展，合理协调生态建设与社会发展间的矛盾，使其处于均衡发展的状态与局面。科学发展观贯穿规划工作的全程，对政府各部门的职能发展提出了更高要求。在土地利用规划工作中，普遍存在重视社会经济发展、忽视生态工程建设的问题。而在城市规划工作中，生态工程也是不能忽视的问题，需加强各方面工作的协调。

2.用地规模的平衡问题

改革开发后施行的土地家庭联产承包责任制，使得农民具备田地的使用权但无所有权，可带动农村生产积极性的提升，加速农业的发展进程。但用于农耕的田地面积相对局限，城市建设用地与农民生产用地间的均衡发展，还需在农村与城市间的土地规划中，加强对划分指标的界定与明确。我国城市规划建设的规模逐步扩大，促使大量耕地土地变为了产业园基地与工业园及物流园等工业用地。透支利用土地资源，易引起城市规划用地滞后于城市化发展进程速度的问题。城市建设用地需符合国家规定的现行标准，实现对现有建设用地的高效利用，减少对农用地的占用。在城市化发展的进程中，城镇发展用地的需求量不断增大，农民耕地数量随之减少，也是标准规范尚未成型与成熟应用的直接体现。

3.用地管理体系不健全

我国农业大国，农业产品数量与种类较多，与农业政策的带动引导有着直接关系。城市化建设进程加速，耕地资源局限，与不断增大的田地需求量产生了冲突，也是城市化与农业用地间的管理体系不完善、耕地占用的监督机制不健全的直接体现，未建立长效的管理机制，耕地随意转让或占用等现象普及，不利于土地资源的利用价值提升。我国的土地使用中，对土地使用信息的掌握少，甚至存在土地无记载与信息缺失的问题。土地信息的更新时间成本高且更新程序繁琐，也是土地管理制度不规范的直接体现。

4.发展与优先资源保护间的矛盾

城市的总体规划工作展开，通常对现有区域内的资源整合优化与高效利用，实现经济发展水平的不断提升，或是调动外来资源，夯实城市发展建设的基础。而土地利用总体规划的工作展开，通常采取沿上至下的开发控制模式，强调土地资源开发利用与保护的并重，颁布了各种耕地转用与开发补充的控制政策。但在城市规划的进程中，普遍存在重视利用资源发展经济的问题，促使城市规划与土地利用规划在衔接中必然会出现错位的情况，也是急需解决的问题。

三、解决城市规划与土地利用间衔接矛盾的策略

1.贯穿可持续发展原则

从可持续发展原则入手分析，是指对土地资源的合理开发利

土地资源管理及其可持续利用研究

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摘要：自从进入21世纪以来，各个国家的耕地面积都在逐渐缩小，而土地属于不可再生资源，是我们生存的基本条件，对我们人类来说有着非常重要的影响。人们在地球上正常生活以及社会经济的可持续发展，土地的开发与利用有着无法衡量的贡献。本篇文章先讲述了土地资源开发出现的问题，后又及时提出了土地资源管理可持续发展的应用策略，实现土地资源与国民经济的可持续发展。

关键词：土地资源管理；可持续利用研究

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土地资源是人类生存与经济发展的前提，与此同时，也是我国步入家到小康的首要条件。但是，由于我国土地资源逐渐减少，人口越来越多，导致土地供应不足以及价格逐渐上升，土地规划也产生不合理的现象，使土地资源的使用率没有有效发挥。所以需要对我国土地进行整体规划，促使可以产生更多使用有效的土地资源，以此来缓解我国人口多土地资源少的现象。提升土地资源使用率，早日完成农业现代化发展，让我国社会经济可持续发展。

一、土地资源开发出现的问题

当下我国人口多、土地资源少两者间的矛盾逐渐上升，人口数量在不断增加，而土地面积却在逐渐下降，人均土地面积也在逐渐减少，土地资源的使用也是越来越紧张，约束着我国的经济发展以及社会发展。最近这几年，我国的经济建设获得飞快地发展，然而在经济建设的过程当中，使用的土地面积也在不断增加，经济建设促使我国原先就紧张的土地资源更是雪上加霜，供应矛盾不断白热化，使用土地缺口持续创新。

土地资源在开发过程中，因为对整体规划的认识度不够，导致规划不够仔细周密，不具有科学性以及没有科学、合理的使用规划。这也可能是由于我国当下的土地开发还处在刚开始阶段，许许多多的问题都没来得及仔细研究。

土地规划过程中的灵活度不够，有一部分规划项目相对来说比较呆滞、死板，设计方案也非常单一、枯燥，灵活性不是很强，根本无法满足当下市场经济的特点，所以说在某种程度上缺少一定的竞争力，到最后可能影响到相关土地规划的完成与实现。另外，土地资源在使用的时候，出现的土地破坏情况也是非常严重的，农药以及化工业产生的废水没有经过相关的处理就排放到就近的土壤里面，直接导致土地质量遭到破坏，严重影响到我国土地的整体质量。农民在耕地的过程中也出现一些不合理的问题，没有经过科学、合理地处理土地以及养地的关系，严重导致土壤当中的肥料含量直线下降以及土壤中的物理性逐渐降低，这样长期下去就会导致产生一些中低级的产田。

二、土地资源管理可持续发展的应用策略

我国当下的土地资源在开发过程中产生了许许多多的问题，比如说规划不合理，直接导致土地资源的使用率非常低下，所以，应当开展科学合理的规划布局，尽最大的努力有效发挥出土地管理的作用。各个地区都应当根据当地的真实情况对土地进行整体详细规划，每个地域的情况都是不一样的，需要仔细完成各个区域规划的任务，仔细认真地进行审批，并科学、合理地制定用地制度。与此同时对土地使用详情展开一系列的追踪，加强对土地使用的监测，有效提升土地使用率，早日实现社会与经济效益。

我国人口日益增多，但是人均耕地面积是很小的，人地两者间的矛盾逐渐上升，所以需要规划生育问题，控制人口出生率，持续提升人口质量，可以有效减小人地矛盾，让土地能够长期使用。提高我国土地的农作物产量，尽量减少土地污染现象，必须加强土地的生态规划与建设，避免产生中低产田。积极主动地鼓励我国农民进行科学合理施肥，尽量避免使用农药，大力提倡绿色农业、无化肥农业，创造农业生态相关系统，从基础上就消除污染源。

大力提倡创建土地规划队伍，建立相应的土地规划机构，提升工作人员的规划水平与规划质量。作为规划工作者需要不断提升自身的知识与技能，提升理论文化素养，规划的好与坏，都离不开相关工作人员去完成与实施。为了适应我国当下情势的严峻挑战，有效解决一系列突发性的问题，就必须大力提倡建设规划队伍。根据我国社会经济的快速发展形势，应当积极主动地对土地规划工作人员提出更加严格地要求与更难的任务。

加强我国人民的土地保护观念，与此同时树立土地资源可持续利用观念，科学、合理地安排与使用土地并保护耕地，我国的国土保护部门应当有效提升相关工作人员的文化素质，加强规划工作的科学合理性以及操作性。

应当严格管控与建设用地规划，创建一套完整的保护土地机制。农民在耕地的过程中，应当对土地肩负起保护作用，科学合理地改良土地，有效避免水土流失的现象。另外，在土地规划时应当具备一定的科学性，合理使用土地面积，投入大量的资金与科技力量，减少土地使用紧张的现象，进一步提升土地资源使用率，争取早日实现土地资源可持续发展，以此来保证我国的国民经济与社会经济的可持续发展。

三、结束语

根据以上内容的分析，我们从中可以发现土地资源的正确开发以及使用，对我国未来的经济发展有着非常重要的意义。因此，我们应当学习与吸收国外的经验与教训，加强土地规划管理，重视高科技技术的使用，创造一个具备科学、合理、有效、实用与完善的土地规划管理系统，以此来提升我国土地资源的使用率。作为土地资源管理工作人员应当积极主动地发挥自身的智慧与工作激情，不断努力克服在土地资源管理工作当中碰到的各种疑难问题，并进行一系列科学、合理的规划，促使我国的未来发展更加美好。

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Hans汉斯

农田生态系统恢复力评价与调控机制

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摘　要

科学认识和评价农田生态系统恢复力并构建调控机制，对于合理调控农田生态系统良性发展、促进农田资源可持续利用、高效生态农业发展、保障国家粮食安全和生态安全意义重大。农田生态系统恢复力研究包括农田生态系统恢复力的内涵及构成分析、影响机理与指标体系构建、定量评价方法和调控机制4部分内容。基本思路是以农田生态系统恢复力“内涵界定→构成分析→影响机理→定量评价→调控机制”为主线开展评价并构建起科学的调控机制。评价方法包括数据获取处理、指标体系构建、赋权和评价模型构建、阈值和标准确定、定量评价5个关键环节。调控机制应基于评价结果，结合政府部门和农业经营主体分级分层宏观微观管理需求建立。研究结果可为农田资源持续利用、高效农业发展，以及生态空间格局再造、资源环境协调、生态可持续发展提供科学依据。

关键词

恢复力，生态系统，农田，评价，调控机制

Evaluation and Regulation Mechanism of Farmland Ecosystem Resilience

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特此通知

《城市建筑》编辑部

2021 年 08 月 02 日