



2022年第3期总38期

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## ▶ 前沿资讯

### 1. 研究破解小麦遗传转化中基因型依赖性难题

**简介:** 1月13日, 中国农业科学院作物科学研究所作物转基因及基因编辑技术与应用创新团队鉴定了一个与小麦植株再生相关的基因 TaWOX5, 利用 TaWOX5 基因克服了小麦遗传转化中的基因型依赖性难题。相关研究成果发表在《自然-植物 (Nature Plants)》上。据叶兴国研究员介绍, 遗传转化效率低和基因型依赖性制约小麦转基因研究与基因编辑研究及应用的主要障碍。近年来, 虽然小麦遗传转化效率显著提高, 但仅适用于少数模式小麦基因型, 还有许多小麦品种不适合进行转基因和基因编辑。而利用再生基因能够促进离体培养和转化的小麦组织再生, 提高转化效率并拓展可转化小麦品种的范围, 实现更多品种的转化和遗传改良。该团队历经13年研究, 对多个可能与小麦再生植株相关的基因进行了克隆和功能鉴定, 最后发现 TaWOX5 具有显著提高小麦再生效率和转化效率的作用。研究发现, 过表达 TaWOX5 基因不但显著提高了模式小麦基因型的转化效率, 而且显著提高了22个小麦推广品种或重要种质资源的转化效率, 尤其成功转化了顽拗型明星小麦品种, 转化效率提高了2-10倍。进一步验证发现, TaWOX5 基因在波兰小麦、栽培一粒小麦、黑麦、小黑麦、大麦和玉米等单子叶植物遗传转化中, 均可使转化效率显著提高。另外发现, TaWOX5 对小麦愈伤组织分化和再生植株根系生长没有副作用, 其宽叶表型有利于转基因植株识别以及无筛选标记转基因植株和基因编辑植株的快速鉴定。上述结果表明, TaWOX5 基因大幅度提高了小麦等植物的转化效率, 克服了基因型的限制, 简化了鉴定工作, 具有广阔应用前景。中国科学院种康院士高度评价了此项工作, 他解读到, 小麦等多倍体作物基因遗传转化成功率限制了转基因生物技术育种与产业化。基因型依赖性长期困扰植物遗传转化的瓶颈问题, 即便是烟草、拟南芥和水稻等模式植物的遗传转化, 也存在强烈的基因型依赖性, 限制了利用转基因技术对优良品种的遗传改良。与已报道的再生基因相比, TaWOX5不影响愈伤组织分化和再生植株根系发育, 不需要从愈伤组织中排除。有理由预测, TaWOX5基因在小麦等植物基于转基因和基因编辑技术进行的基因功能鉴定和新材料创制中具有广泛应用前景。中国科学院林鸿宣院士点评到, 遗传转化过程中的基因型限制性普遍存在于各种作物, 并成为限制转基因育种的瓶颈之一。小麦是六倍体作物, 遗传转化一直远远落后于其他主要农作物。叶兴国实验室长期致力于小麦遗传转化技术体系建立和再生基因挖掘, 最近取得了新突破, 基本解决了小麦遗传转化中基因型限制问题, 为小麦转基因育种和基因编辑育种提供了强有力的技术支撑。利用该基因还能显著提高大麦、小黑麦、黑麦、栽培一粒小麦和玉米等单子叶植物的遗传转化效率, 具有广泛的应用前景。该研究得到了国家自然科学基金、宁夏回族自治区重点研发计划和中国农业科学院科技创新工程资助。

**来源:** 中国农业科学院作物科学研究所

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<https://www.caas.cn/xwzx/tpxw/318874.html>

### 2. 睦晓蕾课题组在PNAS杂志在线发表了题为“Transcriptional control of local auxin distribution by the CsDFB1-CsPHB module regulates floral organogenesis in cucumber”的研究论文

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**简介:** 2021年2月23日, 中国农业大学园艺学院睦晓蕾课题组(博士研究生聂静为文章第一作者)在PNAS杂志在线发表了题为“Transcriptional control of local auxin distribution by the CsDFB1-CsPHB module regulates floral organogenesis in cucumber”的研究论文。该研究发现, 一个黄瓜半胱氨酸蛋白酶抑制剂相关(cystatin-like)蛋白CsDFB1, 可以调节花分生组织中生长素生物合成和极性运输关键基因的表达, 从而调控黄瓜花器官和果实的发育。黄瓜属于葫芦科植物, 是世界上主要的蔬菜作物之一, 也是我国重要的设施蔬菜作物, 在全国各地广泛种植。果实是黄瓜的主要经济库器官, 花器官(果实)的正常生长发育对黄瓜产量和品质形成极为重要, 其内在调控机制一直是黄瓜等葫芦科蔬菜作物研究的焦点内容。植物生长素在花器官起始和发育中发挥了关键作用。一般认为, 生长素生物合成和极性运输相互协调, 从而在分生组织中建立并维持合适的生长素浓度梯度(gradient), 以确保包括花器官在内的多种植物器官的正确起始和发育。但是, 黄瓜花分生组织中生长素生物合成和极性运输的调控机制, 之前尚无明确报道。该课题组前期研究中, 针对黄瓜果实发育早期韧皮部组织, 通过激光捕获显微切割结合RNA-seq技术, 获得了一个未知功能基因Csa7G067350。利用反向遗传学技术, 沉默该基因导致黄瓜花瓣、心皮等花器官, 及其果实中维管束数量显著增加, 并伴随着茎尖生长素的明显积累; 而过表达该基因植株茎尖生长素积累减少, 不仅降低花器官数量, 维管分化发生紊乱, 甚或出现器官融合现象而导致果实畸形(图1)。据此, 将该基因命名为CsDFB1 (DEFORMED FLORAL BUD1)。植物半胱氨酸蛋白酶抑制剂(plant cystatins)已被证明在防御反应中发挥重要作用。该课题组后续研究表明, CsDFB1基因编码一个半胱氨酸蛋白酶抑制剂相关蛋白, 但是CsDFB1蛋白可以与HD-ZIP III转录因子CsPHB (PHABULOSA) 互作, 调控CsPHB对生长素生物合成关键基因CsYUC2 (YUCCA2)的转录激活, 及其对生长素外排载体基因CsPIN1 (PIN-FORMED1)的转录抑制, 从而影响植株顶端分生组织生长素分布和花器官发育(图2)。该研究揭示了CsDFB1-CsPHB分子模块通过调节黄瓜局部生长素的分布, 从而调控黄瓜的花器官发生和维管分化的作用机制。这一发现为植物分生组织中生长素分布调控植株生长发育的相关机制提供新的认知, 也为黄瓜分子育种提供了新基因和新思路。文章链接:  
<https://doi.org/10.1073/pnas.2023942118>

**来源:** 中国农业大学园艺学院

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## ► 学术文献

### 1. Overexpression of the VaERD15 gene increases cold tolerance in transgenic grapevine (过量表达 vaerd15基因提高了转基因葡萄的耐寒性)

**简介:** In plants, the Early Responsive to Dehydration (ERD) genes are rapidly activated in response to drought and other abiotic stresses. Here, we characterize the VaERD15 gene from the cold-resistant grapevine *Vitis amurensis* accession 'Shuangyou', and its counterpart VvERD15 from the cold-sensitive *V. vinifera* cultivar 'Red Globe'. To evaluate a role for VaERD15 in promoting cold tolerance, we expressed the VaERD15 in transgenic *V. vinifera*

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'Red Globe' and evaluated its influence on type and cold stress-related physiological and biochemical indices. We also assessed the response of the ERD15 promoter and its deletion fragments from 'Shuangyou' and 'Red Globe' to low temperature, ABA, SA and PEG. Additionally, several proteins interacting with VaERD15 were identified and the expression profiles for the respective genes were evaluated in response to low temperature. We found that the VaERD15 protein contains a PAM2 domain and localizes in the nucleus and cell membrane. Under low-temperature stress, the expression of ERD15 in 'Shuangyou' was greater than that in 'Red Globe'. Transgenic 35S::VaERD15 plants showed greater cold tolerance and had lower electrical conductivity and malondialdehyde content than non-transgenic controls, while the contents of proline, soluble sugar, soluble protein and the activities of SOD, POD and CAT were higher than those in non-transgenic controls. The ERD15 promoter and its deletions could respond to low temperature, ABA, SA and PEG, but the degree of response to the treatments varied. Proteins interacting with VaERD15 were identified as xyloglucan endotransglucosylase/hydrolase protein 7, glutamine synthetase, 40S ribosomal protein 23 and disulfide-isomerase LQY1, and the corresponding genes responded to cold temperatures to varying degrees.

来源: Scientia Horticulturae

影响因子: 3.463/Q1

发布日期: 2022-02-05

全文链接:

<http://agri.ckcest.cn/file1/M00/0F/F0/Csgk0GHqcsyANBJbAQAHWmoCV1U136.pdf>

## **2. An integrated analysis of transcriptome and metabolome provides insights into the responses of maize (*Zea mays* L.) roots to different straw and fertilizer conditions (转录组和代谢组的综合分析为玉米根系对不同秸秆和肥料条件的反应提供了深入的见解)**

简介: Returning straw back to the field with an appropriate amount of fertilizer has been proven to be beneficial in the promotion of soil fertility, crop yield, and the sustainability of an agricultural system. However, little is known regarding the molecular and metabolic understandings of enhanced crop growth in a straw return system. By performing an integrated transcriptomic and metabolomics analysis, we investigated the effects of straw removal and straw return with two chemical fertilizer rates on root morphology in a mesh bag field experiment on maize in northwest China. Our results showed that straw return played a role in root thickening (bigger root diameter), while fertilization enhanced root branching (more root number). Different from straw return alone, straw return with fertilizer significantly decreased the root/shoot ratio ( $p < 0.01$ ) and increased the total root number ( $p < 0.01$ ), and root total nitrogen concentration ( $p < 0.001$ ). Root morphological responses were tightly associated with transcriptional regulation (e.g., root dry weight, Mantel's  $r = 0.6468$ ,  $p < 0.001$ ) and metabolism modulation (e.g., root/shoot ratio, Mantel's  $r = 0.7229$ ,  $p < 0.001$ ), mainly including the activation of genes related to phenylpropanoid and flavonoid metabolic pathways (e.g., BGLU, PAL and POD), the fine regulation of genes associated with auxin synthesis, homeostasis, flow and signaling (e.g., YUC, AUX1 and SAUR), and cell wall

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remodeling (e.g., CESA and EXPA), and the regulation of transcription factors and genes that function in the development of roots (primary, seminal, and lateral), root hairs, root tips, and root caps (e.g., BBM, LRP1 and RTCS), and the accumulation of different metabolites (e.g., indole, esculin, and sucrose) under straw return with fertilization. We revealed important insights into the growth-promoting and morphology-modifying effects of straw return, as well as its highly novel effects on the transcriptional regulation and metabolism reprogram of maize root, which may help to elucidate how maize roots respond under straw return conditions to optimize soil resource acquisition and then guide the suitable utilization of crop residue resources in the agricultural system.

来源: Environmental and Experimental Botany

影响因子: 5.545/Q2

发布日期: 2022-02-01

全文链接:

<http://agri.ckcest.cn/file1/M00/0F/F0/Csgk0GHqc7KAKp6dAMUPuV08PFo656.pdf>

### **3. Susceptibility and tolerance of fenugreek (*Trigonella foenum-graecum* L.) to salt stress: Physiological and biochemical inspections (胡芦巴对盐胁迫的敏感性和耐受性的生理生化观察)**

简介: Greenhouse experiments were conducted to evaluate the effects of soil salinity on some physiological and biochemical attributes of fenugreek (*Trigonella foenum-graecum* L.) and its tolerance for salt using some empirical models. The treatments included different salinity levels of irrigation water (0.5, 2, 4, 6, 8, 10 and 12 dS/m) which were arranged in the form of a randomized complete block design with three replicates. Some physiological and biochemical properties were measured at the vegetative stage and trigonelline at seed ripening. As the results showed, salinity reduced leaf area, relative water content, water use efficiency, and leaf water potential, but it increased leaf thickness, greenness index, canopy temperature, sodium, ash, electrolyte leakage, malondialdehyde (MDA) and the other aldehydes. Salt stress also reduced photosynthetic pigments, chlorophyll a/b ratio, leaf soluble protein, potassium, K<sup>+</sup>/Na<sup>+</sup> ratio, photochemical efficiency of photosystem II (PSII) and canopy air temperature difference (CATD) index. Like catalase (CAT) and ascorbate peroxidase (APX) activities, the activity of superoxide dismutase (SOD) was increased up to 6 dS/m and then decreased. Furthermore, proline concentration was increased up to 8 dS/m and decreased at higher salinities. The increased salinity enhanced seed trigonelline up to 6 dS/m and then decreased it. According to a linear model, the salinity threshold tolerance and the yield reduction slope were 1.28 dS/m and 4.9%, respectively. Nonlinear models, however, showed a yield reduction of 10%, 25% and 50% at the soil salinity of 3.38, 6.28 and 11.67 dS/m, respectively. The salinity tolerance index was found to be 12.24. Therefore, fenugreeks can be considered moderately sensitive to salinity at the vegetative growth stage. It may also be grown on slightly to moderately saline lands. Some characteristics may be considered as selective criteria to identify the cultivars of fenugreek that tolerate salt stress.

来源: Environmental and Experimental Botany

影响因子: 5.545/Q2

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全文链接:

<http://agri.ckcest.cn/file1/M00/03/1E/Csgk0WHqc7KAVarAAB67hx0bncY749.pdf>

#### **4. A WRKY transcription factor PbWRKY40 from *Pyrus betulaefolia* functions positively in salt tolerance and modulating organic acid accumulation by regulating PbVHA-B1 expression (转录因子的 pbwrky40通过调节 pbvha-b1的表达, 对梨的耐盐性和有机酸的积累具有正向调节作用)**

简介: The V-type-H<sup>+</sup>-ATPase gene (VHA-B1) plays an important part in plant stress responses. However, the transcriptional regulation of VHA-B1 in plants under abiotic stress conditions remains poorly understood. Here, a Group II WRKY gene (PbWRKY40) was obtained by yeast one-hybrid (Y1H) screening, using the promoter of PbVHA-B1 as bait. A Y1H assay, electrophoretic mobility shift assay (EMSA), and transient expression analysis further proved that PbWRKY40 could bind to the W-box element of PbVHA-B1 in the promoter region. PbWRKY40 had a complete open reading frame of 963 bp, encoding a 320 amino acid peptide located in the nucleus. PbWRKY40 was greatly induced by salt stress. Overexpressing PbWRKY40 in *Arabidopsis thaliana* and pear callus showed stronger resistance to salt stress compared with the WT. Transgenic lines had a higher germination percentage, root growth, chlorophyll content, and total organic acid content than the WT, while electrolyte leakage, Na<sup>+</sup>/K<sup>+</sup> ratio, and MDA content were lower than the WT. Compared with the WT, the silenced lines were more sensitive to salt stress and accumulated less organic acid. In addition, knockdown of the PbWRKY40 gene in *P. betulaefolia* also decreased the expression of PbVHA-B1. Taken together, our study demonstrated that PbWRKY40 functions in salt tolerance and organic acid accumulation, at least partly, by regulating the expression of PbVHA-B1.

来源: Environmental and Experimental Botany

影响因子: 5.545/Q2

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全文链接:

<http://agri.ckcest.cn/file1/M00/03/1E/Csgk0WHqcpWABqDkAB-sD8Uw3gM905.pdf>

#### **5. High fruit setting rate without male participation: A case study of obligate apomixis in *Rhomboda tokioi* (Orchidaceae) (无雄性参与的高坐果率——以长菱形花序无融合生殖为例)**

简介: Seed production is the key factor for maintaining plant population renewal and development. Because of pollination limitations and other factors, orchids, especially rewardless ones, generally have low fruit set rates. In *Rhomboda tokioi*, however, the natural fruit setting rate is very high, more than 90%. Common reasons for such a high natural fruit setting rate include high pollinator efficiency, autonomous selfing, seed formation through apomixis and so on. To test these possibilities, we carried out field observations, morphological examinations, and breeding system experiments. The results showed no

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migration of pollinia into or out of flowers and suggested that the structure of the floral organs hinder autonomous selfing. Interestingly, the fruit setting rate of this orchid was still higher than 90% when the gynostemium was removed prior to flowering. Additionally, after hand pollination, the pollen tube stopped growing at a late stage and could not enter the ovule, indicating the seeds were produced by obligate apomixis. Morphological experiments showed that a megasporocyte first forms a chain tetrad and then develops into a mature embryo sac, while the embryo of the offspring developed from unfertilized egg cells. More interestingly, the offspring had the same number of chromosomes as the female parent. According to the existing theory of apomixis, we can infer that the reproductive model of *R. tokioi* is *Allium*-type apomixis.

来源: Flora

影响因子: 2.088/Q3

发布日期: 2021-10-01

全文链接:

<http://agri.ckcest.cn/file1/M00/0F/F0/Csgk0GHqcZmAmd4APZTBjgBn1o175.pdf>

## ➤ 相关专利

### 1. 突变型纤维素酶及其在玉米青贮中的应用

简介: 本发明提供了一种纤维素酶突变体,其在SEQ ID NO. 1的基础上加入了T49K、A66P、C81A、F145S和D146G突变以提供更好的低温和低pH适应性;本发明还提供了该纤维素酶突变体在玉米青贮中的应用。

来源: 佰腾网

发布日期: 2021-01-05

全文链接:

<http://agri.ckcest.cn/file1/M00/03/1E/Csgk0WHqd5mABX7xAAcbgIMkHsw991.PDF>

### 2. 一种提高双功能纤维素酶的甘露聚糖酶活的方法及纤维素酶突变体RMX-M和应用

简介: 本发明涉及农业生物技术领域,具体涉及一种提高双功能纤维素酶的甘露聚糖酶活的方法及纤维素酶突变体RMX-M和应用。本发明通过氨基酸序列如SEQ ID NO: 1所示的野生型纤维素酶的E218位点实施定点突变获得E218H突变体。结果表明,以羧甲基纤维素钠和角豆胶为底物两种底物测定时,酶促反应的最适pH值不变,最适温度降低5℃;以角豆胶为底物时,突变体的甘露聚糖比活力比野生型提高了约80%;在以羧甲基纤维素钠为底物时,与野生型RMX的纤维素比活相比,突变体RMX-M的比活略有降低,实现了在纤维素酶活损失较小的基础上提高了其降解半纤维素底物甘露聚糖的能力。

来源: 佰腾网

发布日期: 2020-11-10

全文链接:

<http://agri.ckcest.cn/file1/M00/0F/F0/Csgk0GHqeDKAesF6AAynVumUUmU681.PDF>