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➤ 前沿资讯

1. 蔬菜花卉所百合课题组揭示百合腋生珠芽形成分子调控新机制

简介: 近日, 中国农业科学院蔬菜花卉研究所百合课题组在百合腋生珠芽形成分子调控方面取得新进展。相关研究内容“WUSCHEL-Related Homeobox Genes Cooperate with Cytokinin Signalling to Promote Bulbil Formation in *Lilium lancifolium*”在线发表于《植物生理学(Plant Physiology)》上。珠芽指贮藏养分、形态肥大的芽, 也叫零余子, 着生在植物地上部分的叶腋处。对百合属植物而言, 珠芽是其部分种类重要的繁殖器官。其中, 重要的食用和药用百合卷丹(*Lilium lancifolium*)是自然三倍体种, 不产生种子, 其种群自然传播繁殖完全依赖珠芽。珠芽依附于母体植株生长, 单株百合可产生多达上百粒珠芽。成熟珠芽自母体脱落后可发育成新的完整个体, 可以像种子一样繁殖传播, 且完整地保留了母体的特征而较少变异和退化。因此, 珠芽繁殖在百合籽球生产繁殖上有很广阔的应用前景, 掌握百合珠芽形成调控机制将有望在生产上发挥珠芽高效繁殖的优势。目前, 关于百合珠芽的形成仍处于初步阶段, 其分子调控机制尚不清楚。本研究基于前期卷丹珠芽形成的转录数据, 通过对差异表达基因进行qRT-PCR及荧光原位杂交, 发现L1WOX9和L1WOX11在可形成珠芽的叶腋处高表达, 且在珠芽形成过程中表达量持续升高。进一步通过瞬时过表达和VIGS技术对L1WOX9和L1WOX11在珠芽形成中的功能进行了研究, 发现过表达L1WOX9和L1WOX11可促进卷丹珠芽形成, 而沉默L1WOX9和L1WOX11, 卷丹珠芽的形成受到明显抑制, 表明L1WOX9和L1WOX11参与了百合珠芽形成的正向调控。前期研究表明, 细胞分裂素B类响应因子type-B L1RRs可正向调控百合珠芽形成。本研究克隆了L1WOX9和L1WOX11的启动子, 发现二者均包含大量的type-B L1RRs结合元件。经验证发现, type-B L1RRs能够与L1WOX9和L1WOX11的启动子结合并促进其转录。此外, 研究中还发现L1WOX11与细胞分裂素A类响应因子type-A L1RR9(细胞分裂素信号通路负调控因子)启动子结合并抑制其转录, 进一步加强细胞分裂素信号促进珠芽启动和形成。综上所述, 本研究首次揭示了WUSCHEL-related HOMEBOX家族中间进化支成员L1WOX9和L1WOX11介导细胞分裂素通路调控百合腋生珠芽形成的分子机制。不仅丰富了植物WOX转录因子调控腋生器官形成的理论, 也为人工调控百合珠芽形成, 显著提高百合繁殖效率提供了基因储备, 具有重要的科学意义和应用前景。

该论文以中国农业科学院蔬菜花卉研究所为第一完成单位和通讯单位。已毕业博士生何国仁为该论文第一作者, 杨盼盼助理研究员和明军研究员为共同通讯作者。该研究得到了国家自然科学基金(31902043, 32172612)、国家重点研发计划(2019YFD1001002)、贵州省科技计划项目(20201Y121)及中央级公益性科研院所基本科研业务费专项(IVF-BRF2021017)的资助和支持。(宣传信息员: 杨盼盼) 原文链接: <https://doi.org/10.1093/plphys/kiac259>

来源: 百合课题组 杨盼盼

发布日期: 2022-06-08

全文链接:

<https://ivf.caas.cn/xwdt/kyzj/6313a2403af041f480224322600c3454.htm>

2. 焦雨铃研究组合作研究发布拟南芥高质量参考基因组

简介: 拟南芥(*Arabidopsis thaliana*)作为被广泛应用的模式植物, 其基因组序列极大地加快了植物分子生物学研究。在首个基因组发布二十多年后, 仍然存在大量未填补的

缺口区域。在常用的TAIR10/Araport11版本的基因组序列中，存在165个缺口。这些缺失区域可能由高度重复的序列组成，包括端粒、着丝粒、5S rDNA簇和含有45S rDNA的核仁组织区(NORs)。近年来ONT和PacBio等长读段测序技术的发展为组装高复杂度序列提供了有力工具。最近Science和GPB分别发表了两个高完整度基因组Col-CEN和Col-XJTU，填补了着丝粒等缺失区域。但这两个新的组装仍不完整且有相互矛盾之处。为了给植物学科研工作者提供一个更好的参考基因组，中国科学院遗传与发育生物学研究所焦雨铃研究组与中国科学院大学生科院汪颖研究组合作，结合长读ONT、高保真的长读PacBio HiFi和短读Illumina NovaSeq测序数据获得了接近完整的拟南芥Col-0生态型的参考基因组Col-PEK。Col-PEK组装填补了包括五个着丝粒在内各区域中的绝大多数缺口。例如，Science发布的Col-CEN中5号染色体中的缺口均已在Col-PEK中补齐。Col-PEK为目前最为完整的基因组组装，完成了1、3、5号染色体从端粒到端粒的完整组装，仅2号和4号染色体的多拷贝NORs区域尚不完全。Col-PEK组装总长度133.92 Mb，比TAIR10组装长14.77 Mb，即增加了12.4%的序列。在填补缺口之外，Col-PEK还修订了Col-CEN等组装中的拼接错误。Col-PEK组装具有很高的序列完整度，对Col-PEK的注释揭示了重复序列的分布规律，特别是着丝粒区域的CENH3结合区域分布规律和CEN180重复序列分布特征。对编码基因的注释还发现了145个新的“隐藏基因重复”。这些基因重复与已知基因序列高度相似，可能是由新近的串联重复等基因组扩增机制所产生。

Col-PEK组装补全了所有着丝粒序列及绝大部分其它缺口，纠正了之前的错误组装。对其初步分析展示了重复序列的分布规律，并揭示了一批新基因。Col-PEK参考基因组为国内外植物学科研工作者提供了新的参照序列和重要数据资源。

2022年6月1日，Molecular Plant杂志在线发表了题为“A near-complete assembly of an *Arabidopsis thaliana* genome”的研究论文(DOI:10.1016/j.molp.2022.05.014)，报道了Col-PEK组装。博士生侯学仁为该论文第一作者，汪颖副教授和焦雨铃教授为论文的共同通讯作者。遗传发育所程祝宽研究员和未来组汪德鹏博士参与了该研究。研究得到科技部重点研发计划的资助。

来源：中国科学院遗传与发育生物学研究所

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全文链接:

https://www.cas.cn/syky/202206/t20220602_4836740.shtml?from=singlemessage

➤ 学术文献

1. Identification of QTLs associated with curd architecture in cauliflower.(鉴定与花椰菜腐殖结构相关的QTL。)

简介: BACKGROUND Curd architecture is one of the most important characters determining the curd morphology of cauliflower. However, the genetic mechanism dissection of this complex trait at molecular level is lacking. Genes/QTLs responsible for the morphological differences between present-day loose-curd and compact-curd cauliflower haven't been well revealed.

RESULTS Herein, by using a common compact-curd parent and two loose-curd parents, we developed two double haploid (DH) populations including 122 and 79 lines, respectively. For each population, we decomposed the curd architecture concept into four parameters (basal diameter, stalk length, stalk angle and curd solidity), and collected

corresponding phenotypic data for each parameter across two environments. The Kosambi function and composite interval mapping algorithm were conducted to construct the linkage map and analyze the QTLs associated with curd architecture parameters. A total of 20 QTLs were detected with the minimum likelihood of odd (LOD) values ranging from 2.61 to 8.38 and the percentage of the phenotypic variance explained by each QTL (PVE) varying between 7.69 and 25.10%. Of these, two QTLs controlling stalk length (qSL.C6-1, qSL.C6-2) and two QTLs controlling curd solidity (qCS.C6-1 and qCS.C6-2) were steadily expressed in both environments. Further, qSL.C6-1, qSL.C6-2, qCS.C6-1 and qCS.C6-4 fell into the same chromosomal region of the reference genome, indicating that these loci are involved in pleiotropic effects or are tightly linked.

CONCLUSIONThe current study identified a series of QTLs associated with curd architecture parameters, which might contribute essentially to the formation of present-day loose-curd cauliflower that is widely cultivated in China. These results may pave the way for intensive deciphering the molecular mechanisms of curd development and for marker-assisted selection of curd morphology in cauliflower breeding.

来源：BMC Plant Biology

发布日期:2020-01-01

全文链接:

http://agri.ckcest.cn/file1/M00/03/34/Csgk0YcC4jyAfqS_AB746EsYGjs259.pdf

2. A novel approach to carotenoid accumulation in rice callus by mimicking the cauliflower Orange mutation via genome editing. (通过基因组编辑模拟花椰菜橙色突变在水稻愈伤组织中积累类胡萝卜素的新方法。)

简介：BACKGROUND β -carotene (provitamin A) is an important target for biofortification of crops as a potential solution to the problem of vitamin A deficiency that is prevalent in developing countries. A previous report showed that dominant expression of splicing variants in the Orange (Or) gene causes β -carotene accumulation in cauliflower curd. In this study, we focused on a putative orthologue of the cauliflower Or gene in rice, Osor, and attempt to accumulate β -carotene in rice callus by modification of the Osor gene via genome editing using CRISPR/Cas9. FINDINGSCRISPR/Cas9 vectors for the Osor gene were constructed and transformed into rice calli. Some transformed calli showed orange color due to β -carotene hyper-accumulation. Molecular analyses suggest that orange-colored calli are due to an abundance of in-frame aberrant Osor transcripts, whereas out-of-frame mutations were not associated with orange color.

CONCLUSIONSWe demonstrate that directed gene modification of the Osor gene via CRISPR/Cas9-mediated genome editing results in β -carotene fortification in rice calli. To date, golden rice, which accumulates β -carotene in rice endosperm, has been developed by conventional transgenic approaches. Our results suggest an alternative approach to enhancing β -carotene accumulation in crops.

来源：RiceVolume

发布日期:2019-12-01

全文链接:

<http://agri.ckcest.cn/file1/M00/03/34/Csgk0YcC3wWAXqxPABk0u5azTD0926.pdf>

3. CRISPR/Cas9-mediated resistance to cauliflower mosaic virus. (CRISPR / Cas9介导的花椰菜花叶病毒抗性。)

简介：Viral diseases are a leading cause of worldwide yield losses in crop production. Breeding of resistance genes (R gene) into elite crop cultivars has been the standard and most cost-effective practice. However, R gene-mediated resistance is limited by the available R genes within genetic resources and in many cases, by strain specificity. Therefore, it is important to generate new and broad-spectrum antiviral strategies. The CRISPR-Cas9 (clustered regularly interspaced palindromic repeat, CRISPR-associated) editing system has been employed to confer resistance to human viruses and several plant single-stranded DNA geminiviruses, pointing out the possible application of the CRISPR-Cas9 system for virus control. Here, we demonstrate that strong viral resistance to cauliflower mosaic virus (CaMV), a pararetrovirus with a double-stranded DNA genome, can be achieved through Cas9-mediated multiplex targeting of the viral coat protein sequence. We further show that small interfering RNAs (siRNA) are produced and mostly map to the 3' end of single-guide RNAs (sgRNA), although very low levels of siRNAs map to the spacer region as well. However, these siRNAs are not responsible for the inhibited CaMV infection because there is no resistance if Cas9 is not present. We have also observed edited viruses in systematically infected leaves in some transgenic plants, with short deletions or insertions consistent with Cas9-induced DNA breaks at the sgRNA target sites in coat protein coding sequence. These edited coat proteins, in most cases, led to earlier translation stop and thus, nonfunctional coat proteins. We also recovered wild-type CP sequence in these infected transgenic plants, suggesting these edited viral genomes were packaged by wild-type coat proteins. Our data demonstrate that the CRISPR-Cas9 system can be used for virus control against plant pararetroviruses with further modifications.

来源：Plant Direct

发布日期:2018-03-01

全文链接:

http://agri.ckcest.cn/file1/M00/10/06/Csgk0GKsMP-APTcfAB4c114_11o394.pdf

4. A novel fluorescent biosensor for detection of target DNA fragment from the transgene cauliflower mosaic virus 35S promoter (一种检测转基因花椰菜花叶病毒35S启动子目标DNA片段的新型荧光生物传感器)

简介：In this paper, we reported a convenient fluorescence method for the detection of genetically modified organisms (GMOs). As it is known that the cauliflower mosaic virus (CaMV) 35S promoter is widely used in most transgenic plants (Schnurr and Guerra, 2000), we thus design a simple method based on the detection of a section target DNA (DNA-T) from the transgene CaMV 35S promoter. In this method, the full-length guanine-rich single-strand sequences were split into fragments (Probe 1 and 2) and each part of the fragment possesses two GGG repeats. In the presence of K⁺ ion and berberine, if a complementary target DNA of the CaMV 35S promoter was introduced to hybridize with

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Probe 1 and 2, a G-quadruplexberberine complex was thus formed and generated a strong fluorescence signal. The generation of fluorescence signal indicates the presence of CaMV 35S promoter. This method is able to identify and quantify Genetically Modified Organisms (GMOs), and it shows wide linear ranges from 5.0×10^{-9} to 9.0×10^{-7} mol/L with a detection limit of 2.0×10^{-9} mol/L.

来源：Biosensors and Bioelectronics

发布日期:2013-06-15

全文链接:

http://agri.ckcest.cn/file1/M00/10/06/Csgk0GKsLcqAJVm_jAAXYtXkuexQ982.pdf

➤ 相关专利

1. 一种利用转基因技术培育短蔓花期延迟西瓜新种质的方法

简介：本发明涉及一种利用转基因技术培育短蔓花期延迟西瓜新种质的方法，本发明还涉及构建RNAi的方法，具体通过用DHpart27RNAiFADp1P4载体构建含西瓜Cla97C04G079450基因干扰片段的重组干扰载体，然后采用农杆菌介导法将构建的RNAi载体转化西瓜，筛选得到短蔓、花期延迟的西瓜新种子，本发明首次公开利用转基因方法成功得到具短蔓、花期推迟并且西瓜品质优的种质资源的技术，并且得到的短蔓西瓜，短蔓程度更高，在实际应用中能够更加节约土地资源。

来源：佰腾网

发布日期:2022-03-08

全文链接:

http://agri.ckcest.cn/file1/M00/10/06/Csgk0GKsPbmAAI93AA0J3_j0ngtg256.PDF

2. 茄子果色上位P基因位点候选基因及其应用方法

简介：本发明公开了茄子果色上位P基因位点候选基因；同时公开了一种茄子Sm ANS-1突变型基因，该Sm ANS-1突变型基因相比已知的茄子SmANS基因序列(EU809469和KT727966)，包含1处无义突变，导致翻译的提前终止，使花青苷合成酶的功能丧失。本发明还公开了茄子果色上位P位点候选基因在茄子果色上位性遗传中作用及在果色改良中的应用。

来源：佰腾网

发布日期:2021-04-20

全文链接:

<http://agri.ckcest.cn/file1/M00/03/34/Csgk0YcC7T2ANQwrABHbq00TecE158.PDF>

3. 拟南芥叶绿素b合成基因CAO在番茄中的应用

简介：本发明涉及一种拟南芥叶绿素b合成基因CAO在番茄中的应用，在番茄中过量表达AtCAO，以改变番茄的叶绿素含量及叶绿素a/b比例，研究创制出的新种质的光合系统的变化，叶绿素荧光参数和光合作用效率的变化；并分析新种质的农艺性状的改变，从而获得整体光合作用效率得到显著提高且产量或品质得到显著提高的蔬菜株系，从而为设施园艺蔬菜提供能够适应弱光环境的优良种质资源。培育出新的能够适应设施栽培弱

光环境的番茄新品种。该研究的实施，将有望将来大幅度提高设施番茄的产量及种植效益，满足消费者在不同季节对番茄的需求，增加菜农的经济收益。

来源：佰腾网

发布日期:2021-01-22

全文链接:

http://agri.ckcest.cn/file1/M00/03/34/Csgk0YcC726ATh_xAYc3oVeKgY641.PDF

4. 一种创制枯萎病抗性西瓜种质材料的方法

简介：本发明涉及一种创制枯萎病抗性西瓜种质材料的方法，属于分子育种技术领域，首先针对西瓜的Clpsk1基因的第一个外显子，设计基于CRISPR/Cas9的sgRNA序列，然后构建含有编码所述sgRNA序列的DNA片段的pRGEB32-CAS9-gRNA-Clpsk1载体；利用农杆菌介导法将基因编辑载体导入受体西瓜品种苏蜜1号，获得转化植株；对转化植株进行Clpsk1基因编辑检测和枯萎病抗性鉴定，所得阳性植株即为具有枯萎病抗性的西瓜种质材料。本发明方法可快速高效地获得西瓜抗病新种质，极大地缩短了育种周期，提高了育种效率，获得的材料不含有任何外源基因，除提高了西瓜抗枯萎病的能力，其他农艺性状不受影响。

来源：佰腾网

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全文链接:

<http://agri.ckcest.cn/file1/M00/10/06/Csgk0GKsM6eAMkIPABE1-hr0HIY451.PDF>