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蔬菜育种专题

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▶ 前沿资讯

1. How to make the gene-editing tool CRISPR work even better (如何让基因编辑工具CRISPR更有效力)

简介: 目前, 美国德克萨斯州大学奥斯汀分校的研究团队找到了决定性的证据, 证明在CRISPR基因编辑技术中首次发现且最常用的酶Cas9的效力和精确性都低于一种较少使用的CRISPR蛋白Cas12a。由于Cas9更有可能在编辑动植物基因组的时候找错目标, 因此科学家认为改用Cas12a会使基因编辑更加安全有效。研究成果发表在《分子细胞》杂志上。

该研究团队发现, Cas12a之所以更理想, 是因为它能像搭扣一样和基因组目标结合在一起, 而Cas9与目标结合的方式更像强力胶。两种酶都携带一小串基因编码, 这些编码写在RNA中, 与病毒DNA中的目标基因编码串相匹配。当酶遇到某种DNA时, 就开始尝试组成碱基对来与之连接——从一端开始不断检查一方(DNA)中的每个字母与另一方(RNA)中相邻字母的匹配度。

对于Cas9来说, 每个碱基对像强力胶水一样牢牢粘在一起。如果双方的头七个或八个字母匹配良好, Cas9就已经紧紧和DNA结合在一起了, 之后的字母Cas9就不再检查了。这意味着很容易忽视后面的不匹配情况, 导致找错目标。而Cas12a更像是搭扣。每个点的结合程度相对较弱, 需要两串编码中更多字母相匹配才能进行编辑, 因此更有可能找到预期编辑的基因组部分。Cas12a使碱基对的形成更加可逆, 即Cas12a更能有效检查每个碱基对, 匹配之后再检查下一个, 一直可以检查约18个字母。

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▶ 学术文献

1. Temperature and plant age drive downy mildew disease epidemics on oilseed Brassica napus and B-juncea (温度和株龄影响甘蓝型油菜和芥菜霜霉病的流行)

简介: Studies were undertaken on the effects of temperature (14/10 °C and 22/17 °C day/night) and plant age (15, 23, 31 and 40 day-old-plants) on the severity of downy mildew (*Hyaloperonospora parasitica*) on oilseed Brassica cultivars (temperature: Brassica juncea Montara, B. napus Atomic, ATR-Hyden, Hyola 432, Hyola 450 TT, Thunder TT; plant age: B. juncea Dune, B. napus Surpass 402 and Hyola 450 TT). For temperature studies, there were significant ($P < 0.001$) effects of temperature, cultivar, and cultivar x temperature interaction. On cotyledons of susceptible cultivars (B. napus Hyola 450 TT and Thunder TT), plants were symptomatic at 22/17 °C by 48 h post inoculation (hpi) and with abundant sporulation evident by 72 hpi, and with all cotyledons of B. napus Thunder TT collapsed by 7 days post inoculation (dpi). However, at 14/10 °C, there were no symptoms on the same cultivars until 5 dpi, and sporulation only observed at 7 dpi. Percent disease index values

(DI%) at 22/17 °C of *B. juncea* Montara and *B. napus* ATR-Hyden, Hyola 432, Atomic, Hyola 450 TT and Thunder TT were 4.5, 49.0, 51.4, 65.8, 86.3 and 96.0, respectively, with all except *B. juncea* Montara having significantly lower ($P < 0.001$) disease at 14/10 °C with DI% values of 2.8, 30.4, 27.9, 31.1, 44.4 and 76.4, respectively. For plant age studies, there were significant ($P < 0.001$) effects of plant age, cultivar, and cultivar x plant age interaction. DI% was significantly higher at 15 compared to 40 day-old-plants (dop) across all cultivars. *B. juncea* Dune showed greatest resistance, particularly on 40 dop, with DI% values of 25.8, 24.6, 22.9 and 7.5, for 15, 23, 31 and 40 dop, respectively. *B. napus* Surpass 402 showed high susceptibility on cotyledons of 15 dop but moderate resistance on leaves of other ages, with DI% values of 59.0, 31.2, 27.1 and 26.2 for 15, 23, 31 and 40 dop, respectively. *B. napus* Hyola 450 TT showed very high susceptibility at the cotyledon stage on 15 dop, but some resistance on 23 dop and more so on 31 and 40 dop, with DI% values of 84.0, 41.2, 35.4 and 32.9 for 15, 23, 31 and 40 dop, respectively. Together, these findings explain for the first time why development of downy mildew epidemics on susceptible cultivars occurs early in the growing season when warmer seasonal temperatures in autumn coincide with presence of seedlings; in contrast to later in the growing season on less susceptible older plants coinciding with cooler and less favourable winter temperatures. Increasing maximum and minimum temperatures associated with climate change have likely fostered the increased severity of downy mildew over the past 15 years.

来源: European Journal of Plant Pathology

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<http://agri.ckcest.cn/ass/7edda6b8-113b-4a44-a8a2-3cb448c263ce.pdf>

2. Jasmonic Acid-Mediated Aliphatic Glucosinolate Metabolism Is Involved in Clubroot Disease Development in *Brassica napus* L. (茉莉酸介导的脂肪族硫代葡萄糖苷代谢参与甘蓝型油菜根肿病发展过程)

简介: Glucosinolate (GSL) is associated with clubroot disease, which is caused by the obligate biotrophic protist *Plasmodiophora brassicae*. Due to the complicated composition of GSLs, their exact role in clubroot disease development remains unclear. By investigating clubroot disease resistance in cruciferous plants and characterizing the GSL content in seeds, we can determine if clubroot disease development is related to the components of GSLs. The difference in the infection process between *Matthiola incana* L. (resistant) and *Brassica napus* L. (susceptible) was determined. Root hair infection was definitely observed in both resistant and susceptible hosts, but no infection was observed during the cortical infection stage in resistant roots; this finding was verified by molecular detection of *P. brassicae* via PCR amplification at various times after inoculation. Based on the time course detection of the contents and compositions of GSLs after *P. brassicae* inoculation, susceptible roots exhibited increased accumulation of aliphatic, indolic, and aromatic GSLs in *B. napus*, but only aromatic GSLs were significantly increased in *M. incana*. Gluconapin, which was the main aliphatic GSL in *B. napus* and present only in *B. napus*, was significantly increased during

the secondary infection stage. Quantification of the internal jasmonic acid (JA) concentration showed that both resistant and susceptible plants exhibited an enhanced level of JA, particularly in susceptible roots. The exogenous JA treatment induced aliphatic GSLs in *B. napus* and aromatic GSLs in *M. incana*. JA-induced aromatic GSLs may be involved in the defense against *P. brassicae*, whereas aliphatic GSLs induced by JA in *B. napus* likely play a role during the secondary infection stage. Three candidate MYB28 genes regulate the content of aliphatic GSLs identified in *B. napus*; one such gene was BnMYB28.1, which was significantly increased following both the treatment with exogenous JA and *P. brassicae* inoculation. In summary, the increased content of JA during the secondary infection stage may induce the expression of BnMYB28.1, which caused the accumulation of aliphatic GSLs in clubroot disease development.

来源: Frontiers in Plant Science

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3 . Integrated genomic and transcriptomic insights into the two-component high-affinity nitrate transporters in allotetraploid rapeseed (对异源四倍体油菜籽中双组分高亲和力硝酸盐转运蛋白的基因组学和转录组学研究)

简介: **Background and aims** The two-component high-affinity nitrate (NO_3^-) transport system (THATS) proteins (NRT2/NAR2) play key roles in the efficient nitrogen (N) uptake and transport under N limitations. We aimed at uncovering the core THATS gene(s) regulating N use efficiency (NUE) in allotetraploid rapeseed (*Brassica napus* L.). Methods Genomic information, high-throughput transcriptome sequencing and gene co-expression network were integrated to identify and characterize the core THATS genes.

Results We identified 17 BnaNRT2 and eight BnaNAR2.1 homologs spanning across the rapeseed genome. Copy number and gene presence/absence variations of BnaNRT2s/BnaNAR2.1 s, undergoing strong purifying selection, occurred. The over-representation of Dof, MYB and WRKY cis-regulatory elements and the enrichment of CpG islands, and protein phosphorylation sites indicated the importance of transcriptional and epigenetic regulation in the BnaNRT2 activities, respectively. qRT-PCR assays and high-throughput RNA-seq revealed that both BnaNRT2s and BnaNAR2.1 s were expressed preferentially in the roots; and they showed significantly differential expression under different N forms or different levels of NO_3^- supply. A gene co-expression network identified BnaC8.NRT2.1a and BnaC2.NAR2.1 as the core THATS genes.

Conclusions The core THATS members can serve as elite gene resources for crop NUE improvement. The transcriptomics-assisted gene co-expression network analysis provides novel insights regarding the rapid identification of central members within large gene families of plant species with complex genomes.

来源: Plant and Soil

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<http://agri.ckcest.cn/ass/7c600a38-bc20-4e9a-a44e-812fc447ffc2.pdf>

4. Quantitative trait locus analysis of seed germination, seedling vigour and seedling-regulated hormones in Brassica napus (甘蓝型油菜种子发芽、幼苗活力及幼苗调节激素的QTL分析)

简介: Good germination and seedling vigour are major breeding targets in winter oilseed rape (*Brassica napus*), because seedling vigour and prewinter crop establishment are closely associated with postwinter growth and yield. Here, we identified quantitative trait loci (QTL) related to germination, seedling vigour and seedling-regulated hormones in a doubled haploid (DH) mapping population from a cross between winter oilseed rape parents with high vigour (Express 617) and low vigour (1012-98). By phenotyping in a climate-controlled glasshouse, we identified a total of 13 QTL on nine chromosomes for germination and seedling-related traits at 7 and 14 days after sowing (DAS), explaining up to 11.2% of the phenotypic variation for seedling vigour. Forty-seven metabolic QTL on 15 chromosomes were identified for auxin, abscisic acid (ABA) and dihydrophaseic acid (DPA) at 5 and 12 DAS, explaining up to 49.4% of phenotypic variation in seedling hormone composition. Multitrait QTL hot spots contribute to our understanding of the genetics and metabolomics of germination and seedling vigour in *B. napus*, and represent potential targets to breed high-vigour cultivars.

来源: Plant Breeding

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