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棉花GhCCR4基因的瞬时表达研究

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Functional Analysis of GhCCR4 Gene in the Epidermal Cells of Cotton Ovules

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摘要 利用棉纤维发育调控基因瞬时表达体系，分析目的基因在棉花纤维发育过程中可能存在的功能。实验构建了由CaMV35S启动子驱动的pGUS-CCR4融合瞬时表达载体，使用基因枪轰击法转化棉花胚珠，确定了转化0 DPA胚珠的最佳条件：轰击压力为1350 psi，轰击距离为9 cm，轰击次数为2次。GUS组织化学染色结果表明，*GhCCR4*基因在棉花纤维伸长期和次生壁增厚期持续表达。不同发育时期纤维长度测量结果发现，在8 DPA时转*GhCCR4*基因纤维长度和对照相比没有明显差别，但在27 DPA时纤维长度明显短于对照。纤维透射电镜切片观察发现，转*GhCCR4*基因的细胞次生壁与对照相比增厚达17 %。

关键词： 瞬时表达 棉纤维 *GhCCR4* 透射电镜

Abstract： In this paper, the role of *GhCCR4* in cotton fiber development was examined. Because the generating of transgenic cotton needs a long period of time and the transformation efficiency is still low, transient expression assay system was chosen for test of *GhCCR4* function in cotton fibers by using cotton ovule culture and biolistic transformation techniques. Transient expression vector of cotton *GhCCR4* gene was constructed. The transient expression vector pGUS-CCR4 is driven by CaMV35S promoter with GUS reporter gene and *GhCCR4* gene. The result showed that the highest transformation efficiency of GUS was obtained when 0 day of anthesis (0 DPA) ovules were applied. The optimal conditions for bombardment include helium pressures of 1350 psi, distance of 9 cm and bomb times of twice. Histochemical staining showed that high level of *GhCCR4* gene expression was detected during the fiber rapid elongation stage and the fiber secondary wall thickening stage in cultured ovules. Measurement of fiber length in different developmental stage showed that the fiber length of the transgenic plants at the stage of 8 DPA was not different as comparing with that of the wild-type plants. However, the fiber length in the transgenic plants reduced 19% comparing with wild-type plants at the stage of 27 DPA. Transmission electron microscopy(TEM) demonstrated that the wall thickness of transgenic fiber increased by 17% of that of the wild type.

Keywords： transient expression cotton fiber *GhCCR4* TEM

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