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条锈菌诱导的小麦EF手钙离子绑定蛋白基因 *TaCab1*的功能初步分析

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Preliminary analysis on the role of an EF-hand binding protein gene *TaCab1* in wheat leaves challenged with *Puccinia striiformis* f. sp. *tritici*

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摘要 根据小麦EF手钙离子绑定蛋白(*TaCab1*)基因序列,利用WMD3软件设计特异的人工miRNA (amiRNA),构建VIGS沉默载体。利用amiRNA-VIGS体系,对小麦的*TaCab1*基因的功能进行了初步分析。利用Northern blot和实时定量PCR技术分别检测了amiRNA的积累及*TaCab1*的沉默效率,并利用显微观察技术统计条锈菌侵染小麦后的组织学差异。结果表明,amiRNA可以得到有效的积累,其靶标基因*TaCab1*可以得到有效的沉默。从表型上看,小麦叶片上条锈菌夏孢子的产孢量也在一定程度上有所降低。组织学观察发现当*TaCab1*被沉默后,寄主细胞的坏死面积在侵染后期明显增大,条锈菌的菌丝分枝数也明显增多,但菌丝长度明显变短。

关键词: 小麦 EF手钙离子绑定蛋白 amiRNA VIGS 功能

Abstract: Role of a calcium binding EF-hand protein gene *TaCab1* in the response of wheat to fungal infection was studied using a specific artificial miRNA (amiRNA). Corresponding amiRNA-VIGS vectors were constructed by the over-lap PCR method. Accumulation of amiRNA in leaves transformed with the amiRNA-VIGS vectors was detected by northern blot and the efficiency of the amiRNA in silencing the target *TaCab1* was evaluated by quantitative real-time PCR. Histological changes in wheat leaves challenged with stripe rust pathogen were analyzed by microscopic observation. It was shown that accumulation of amiRNA reduced the expression of *TaCab1* effectively in the transduced leaves. Along with some changes in histological characteristics, leaves expressing the amiRNA suppressed fungal sporulation but increased leaf necrosis. The growth of *Puccinia striiformis* f. sp. *tritici* was changed, showing an increase in hyphal branches and a decrease in hyphal lengths.

Key words: EF-hand binding protein amiRNA VIGS function

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