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棉铃虫 *HaHR3* 基因的克隆及其植物RNA干扰载体的构建

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Cloning of *Helicoverpa armigera* Gene *HaHR3* and Construction of Its RNA Interference Vector

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摘要 采用RT-PCR方法克隆了棉铃虫蜕皮调节转录因子 *HaHR3* 基因, 该基因含有1671 bp的完整开放阅读框。序列分析表明, *HaHR3* 与多种昆虫蜕皮调节转录因子高度同源。利用生物信息学方法对 *HaHR3* 的结构特征进行分析发现, *HaHR3* 具有蜕皮调节转录因子超家族的典型特征, 包括两个锌指结构和一个DNA结合结构域, 不存在信号肽序列和N糖基化位点。选择 *HaHR3* 基因的部分片段构建了RNAi中间载体pRNAi1017-*HaHR3*sa, 再将 *HaHR3* 正反向序列亚克隆至植物表达载体pCambia2300-35S-OCS, 成功构建了由35S启动子调控的 *HaHR3* 基因的正反向RNA干扰载体pCAM-RNAi-*HaHR3*。这一载体的成功构建为下一步通过植物介导的RNA干扰技术防治棉铃虫打下了坚实的基础。

关键词: 棉铃虫 蜕皮调节转录因子 结构分析 RNA干扰

Abstract: The molt-regulating transcription factor (hormone receptor 3, *HR3*) controls the expression of related gene clusters in the process of molting and plays a key role in the regulation of molt cascade reactions. The cDNA segments of *HaHR3*, *HR3* from *Helicoverpa armigera*, were cloned using RT-PCR. With an open reading frame of 1671 bp, *HaHR3* showed 98% identity with *HHR3* (AF337637) in homology comparison. The structure analysis indicated that *HaHR3* has the typical characters of the superfamily of molt-regulating transcription factors, including two zinc finger motifs and a DNA binding domain. Neither signal peptide sequence nor N-glycosylated site was found in the *HaHR3*. Based on the sequence of *HaHR3*, the RNAi vector pRNAi1017-*HaHR3*sa was constructed. By subcloning the sense-antisense sequence of *HaHR3* into the plant expression vector pCambia2300-35S-OCS, pCAM-RNAi-*HaHR3*, the RNAi vector of the *HaHR3* gene which is driven by the 35S promoter, was constructed. This provides a solid foundation for the application of the plant-mediated RNAi technique in the biological control of *Helicoverpa armigera*.

Keywords: *Helicoverpa armigera* *HaHR3* gene structure analysis RNA interference

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