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论文

青稞 *hblt4.2* 基因的克隆及功能分析

何涛^{1,2}; 贾敬芬^{2*}

1青海大学生物科学系, 青海西宁810016; 2西北大学生命科学学院, 陕西西安710069

摘要:

根据大麦 *blt14.2* 基因序列设计引物, 用青稞的DNA为模板扩增低温反应基因的全长序列, 命名为 *hblt14.2*, 在 GenBank 登录号为 EF514912。该基因含 470 bp, 包括 249 bp 的开放阅读框、69 bp 的 5' 非翻译区和 152 bp 的 3' 非翻译区, 编码 82 个氨基酸残基, 富含 Gly、Ala、Leu 和 Val 氨基酸, 与其他冷诱导基因编码蛋白具相似性。与大麦 *blt14.2* 基因具 98.9% 同源性, 所编码的蛋白存在 2 个氨基酸的差别。将 *hblt14.2* 基因连接 CaMV35S 启动子和 NOS 终止子后定向插入质粒 pCAMBIA1301 的多克隆位点中, 构建植物表达载体 pCAMBIA-BI-hblt, 通过农杆菌介导转化烟草, 并对转基因烟草进行抗寒性检测。结果表明, 青稞 *hblt14.2* 基因与植物的抗寒性有一定关系。

关键词: 青稞 低温 *hblt4.2* 基因 功能分析

Cloning and Function Analysis of *hblt14.2* Gene in Highland Barley (*Hordeum vulgare* L. var. *nudum* Hook. f.)

1Department of Biosciences, Qinghai University, Xining 810016, China; 2College of Life Sciences, Northwest University, Xi'an 710069, China

1Department of Biosciences, Qinghai University, Xining 810016, China; 2College of Life Sciences, Northwest University, Xi'an 710069, China

Abstract:

The response to low temperature stress in highland barley indicates that there is a potential gene bank for cold resistance, which is helpful to develop the genes associated with resistance to cold and to survey the regulation mechanism. The objective of this study was to exploit a new gene with cold resistance, to validate its function. A pair of primer was designed based on the *blt14.2* gene sequence documented in GenBank, a low-temperature-responsive (LTR) gene *hblt14.2* was cloned by PCR from highland barley with GenBank accession number EF514912. Its length was 470 bp, containing 249 bp ORF, 69 bp 5' untranslated region (UTR) and 152 bp 3' UTR. Nucleotide sequence of *hblt14.2* gene shares 98.9% (only five base pairs difference) homology with that of *blt14.2* in barley, which encoded a protein of 82 amino acids. The protein was a highly hydrophilic small protein with rich Gly, Ala, Leu and Val amino acids, which were similar to proteins encoded by other LTR genes. The *hblt14.2* ligated with CaMV 35S promoter and NOS terminator was inserted into multiple clone site in plasmid pCAMBIA1301 to construct plant expression vector pCAMBIA-BI-hblt, which was transformed into tobacco mediated by *Agrobacterium tumefaciens*. Transgenic plants response to low temperature were investigated. The results suggested that the *hblt14.2* is related to cold resistance in highland barley.

Keywords: Highland barley Low temperature *hblt14.2* gene Function analysis

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通讯作者: 贾敬芬

作者简介:

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