

### 三七RNase-like 基因的克隆及表达分析

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### Isolation and Expression Analysis of RNase-like Gene from *Panax notoginseng*

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**摘要** 采用同源序列克隆法, 结合RACE技术, 从三七[*Panax notoginseng* (Burk.) F.H.]根茎总RNA中克隆次生代谢相关差异表达RNase-like 贮藏蛋白的编码序列, 并进一步以DNA为模板扩增全长基因, 获得一条开放阅读框为717 bp的cDNA序列, 命名为PMP (GenBank, KC751542), 相应基因全长1 074 bp。序列及其进化分析表明, 该基因包含3个外显子和2个内含子, 编码238个氨基酸, 相应蛋白质的分子量为27.47 kD, 含有核苷酸结合的保守区域, 属于RNase-T2超家族成员。三七PMP蛋白与人参RNase-like 贮藏蛋白高度同源, 序列相似性达95%。实时荧光定量PCR研究表明, 三七PMP基因在其根、茎、叶、花等器官中均有表达, 且3年生根中表达量最高, 暗示该基因可能参与三七皂苷次生代谢调控及其品质形成。

**关键词:** 三七 RNase-like 基因 克隆 表达模式

**Abstract:** Combining homology cloning approaches with RACE (rapid amplification of cDNA ends) techniques, the coding sequence of RNase-like major storage protein with differential expression was cloned from the total RNA of roots and stems of *Panax notoginseng*, then the full gene sequence was amplified from the total DNA. As a result, a cDNA sequence containing a 717 bp ORF (open reading frame) was cloned and named as PMP (GenBank, KC751542), together with a full-length DNA sequence of 1 074 bp. Analysis of sequence and its phylogenetic tree showed that PMP gene consisted of 2 introns and 3 exons, encoding a protein of 238 amino acids. The deduced protein, with a predicted molecular mass of 27.47 kD, contained two conserved domains of RNases, which belonged to the RNase-T2 superfamily member. The sequence showed 95% identity with that of RNase-like major storage protein in *Panax ginseng*. Real-time quantitative PCR showed that the expression level of PMP in 3-year-old root was higher than the other organs. The expression pattern of PMP showed notable correlation with that of main active components in *P. notoginseng*, which suggested that it might be involved in the regulation of secondary metabolism of notoginsenoside and the quality formation.

**Keywords:** *Panax notoginseng*, RNase-like gene, cloning, expression profile

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