

牡丹 *Ty3-gypsy* 类反转录转座子反转录酶序列的克隆及分析

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Cloning and Analysis of Reverse Transcriptase of *Ty3-gypsy-like* Retrotransposons in Tree Peony (*Paeonia*)

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摘要 根据*Ty3-gypsy*反转录转座子反转录酶的保守序列设计简并引物, 从中原牡丹 (*Paeonia suffruticosa* Andrews) 品种‘洛阳红’和野生种卵叶牡丹 (*Paeonia qui* Y. L. Pei et D. Y. Hong) 中扩增出430 bp左右的目标片段。目的条带经回收、克隆、测序及相关生物信息学软件进行序列分析后, 获得了13条来自牡丹的*Ty3-gypsy*反转录转座子反转录酶序列。这些核苷酸序列具有较高的异质性, 主要表现为缺失突变, 序列长度变化范围为412 ~ 446 bp, 同源性范围为71.5% ~ 94.8%。翻译成氨基酸后, 有12条序列出现1 ~ 9个不同程度的终止密码子突变, 3条序列出现移框突变。其核苷酸序列经过系统聚类后可分为6个家族。将其氨基酸序列与已登录的不同物种*Ty3-gypsy*反转录转座子反转录酶的氨基酸序列进行聚类分析, 结果表明与其他植物具有较高的同源性, 表明它们间可能存在着*Ty3-gypsy*反转录转座子的横向传递。

关键词: 牡丹 *Ty3-gypsy* 类反转录转座子 反转录酶 异质性

Abstract: Using degenerate oligonucleotide primers corresponding to conserved domains of the *Ty3-gypsy-like* retrotransposon reverse transcriptase, a fragment of 430 bp was amplified by PCR from the genomic DNA of tree peony (*Paeonia suffruticosa* Andrews ‘Luoyang Hong’) and *Paeonia qui*. The amplicons were recovered and cloned into pMD-18T vector after purification, positive clones were selected and identified by colony PCR, then sequenced and analyzed. Thirteen different sequences of reverse transcriptase from tree peony ‘Luoyang Hong’ and *Paeonia qui* were obtained and six clusters were identified with high heterogeneity through phylogenetic analysis after alignment analyses of their nucleotide sequences. These sequences showed high heterogeneity, mainly characterized by deletion mutations. The length of the nucleotide sequences varied from 412 to 446 bp, and homology ranged from 71.5% to 94.8%. When translated into amino acids, twelve sequences presented stop codon mutation, and three sequences presented frameshift mutation. A phylogenetic tree was constructed based on the amino acid sequences from other species, indicating that horizontal transmission of retrotransposon has occurred among the plants in the past.

Keywords: tree peony, *Ty3-gypsy-like* retrotransposons, reverse transcriptase, heterogeneity

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