

琯溪蜜柚及其早熟红肉突变体成熟果实SSH文库的构建及初步分析

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Construction and Analysis of Suppression Subtractive Hybridization Library of Red-fleshed Sweet Pomelo

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摘要 分别以琯溪蜜柚及其突变体红肉蜜柚(成熟期早20~25 d)的成熟果肉cDNA作驱动子和检测子,利用结合分子像选择技术的SSH技术,构建了红肉蜜柚和琯溪蜜柚的正向差减cDNA文库。通过PCR检测cDNA克隆外源插入片段,其大小介于100~1 000 bp。通过点杂交差异筛选文库,得到102个表达增强2倍或以上的克隆并测序,结果表明这102个克隆代表44个Unigenes,通过序列同源性比对分析,发现获得的Unigenes在功能上主要涉及信号传导、蛋白质合成、应激反应、转运等代谢反应。

关键词: 琯溪蜜柚 突变体 抑制差减杂交 差异表达基因

Abstract: A forward subtractive cDNA library was constructed through suppression subtractive hybridization (SSH) method with mirror orientation selection, which was performed using the cDNA from wild type 'Guanxi' pomelo (*Citrus grandis*) as driver, those from its red-flesh mutant as tester. PCR analysis showed that most of the clones with insertions ranged from 100 bp to 1 000 bp. After differential screening through dot hybridization, a total of 102 transcripts represented 44 Unigenes with signal ratios stronger than two times were selected for 2 additional screenings. Homology analysis showed that they were found to be involved in diverse biological processes, such as signal transduction, protein synthesis, stress response as well as transport metabolism.

Keywords: words: *Citrus grandis* (L.) Osbeck, mutant strain, suppression subtractive hybridization (SSH), differentially expressed sequence

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