

作物遗传育种·生物技术

石榴ISSR-PCR反应体系的优化研究^{*}

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摘要 以酸绿籽石榴为试材,对ISSR反应体系中的各个主要影响因子进行优化筛选。结果表明: 25 μL ISSR反应体系各组分的最适浓度分别为: 1×buffer (不含Mg²⁺), 1.5 mmol/L Mg²⁺, 0.2 mmol/L dNTPs, TaqDNA聚合酶1.5 U,引物0.3 μmol/L,模板40 ng。反应程序为: 94 °C预变性4 min, 94 °C变性45 s, 54 °C退火1 min, 72 °C延伸2 min, 40个循环,最后72 °C延伸10 min。该体系的建立为从DNA分子水平进一步研究石榴种质资源遗传多样性、系统分类、品种鉴定奠定基础。

关键词 [ISSR](#); [石榴](#); [优化体系](#)

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Optimizations of the Inter-simple Sequence Repeat Reaction System in Pomegranate

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Abstract

The optimization of the main influential factors of ISSR reaction system in pomegranate (*Punica granatum* L), Suanlvzi cultivar was studied. The results showed that the optimal concentrations of components in 25 μL reaction system were 1xBuffer (without Mg²⁺), 1.5 mmol/L Mg²⁺, 0.2 mmol/L dNTPs, 1.5 U Taq DNA polymerase, 0.3 μmol/L primer and 30 ng template DNA, respectively. The PCR programme was designed as following: pre-denaturing at 94 °C for 5 min, denaturing at 94 °C for 35 s, annealing at 36 °C for 35 s, extending at 72 °C for 1 min, total 40 cycles, then extending at 72 °C for 5 min. This system can be used in the further studies of genetic diversity, systematic classification and identification of *P. granatum* L. at DNA Level.

Key words [ISSR](#); [pomegranate\(*Punica granatum* L.\)](#); [optimization](#)

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