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大豆ISSR-PCR反应体系的优化

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摘要： 以黑龙江省大豆为材料，利用正交试验设计，对大豆ISSR-PCR反应体系中的5种主要因素(Mg2+、引物、dNTPs、模板DNA量、Taq酶量)在4个水平上进行体系优化。结果确定了大豆ISSR-PCR反应的最佳体系(25 μL)为：Mg^{2+}浓度1.85 mmol·L-1、dNTPs浓度1.2mmol?L-1、引物浓度1.2μmol?L-1、模板DNA60 ng、Taq酶量0.7 U。利用该最佳体系，选取引物855对25份材料进行扩增，以验证该体系的稳定性。建立了适于大豆的ISSR-PCR反应体系，为利用ISSR标记技术开展黑龙江省大豆遗传多样性分析提供了依据。

Abstract: Orthogonal design was adopted to optimize ISSR-PCR amplification system on soybean in five factors (Mg2+,dNTPs,primer,DNA template and Taq DNA polymerase)at four levels respectively, with 25 soybean germplasm from Heilongjiang Province as material. The most suitable ISSR-PCR system for soybean was established, namely 25 μL reaction system containing 1.85 mmol·$L^{-1}$$Mg^{2+}$,1.2mmol·$L^{-1}$dNTPs,60 ng DNA template, 1.2μmol·L^{-1}primer, and 0.7 U Taq DNA polymerase. Underthe optimized reaction conditions, 25 soybean were easily amplified with primer 855. The result provided a standardizing program for the analysis of interspecies genetic diversity of soybean of Heilongjiang Province.

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