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技术与方法

# 一种单个胚胎的DNA模板制备方法

李军锋, 李海峰, 宋艳画, 孙 燕, 张家骅

西南农业大学重庆市牧草与草食家畜重点实验室，重庆 400716

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摘要 建立了一种简单处理单个卵子和早期胚胎制备DNA模板的方法——KOH/DTT-Triton X裂解法，并与TE-蛋白酶K法比较了PCR扩增效率。结果，采用KOH/DTT-Triton X裂解法处理单个卵子或2-细胞胚、8-细胞胚、桑椹胚、囊胚后，作为DNA模板直接进行PCR扩增线粒体DNA片段，3对引物的PCR扩增总成功率为100%（70/70），而TE-蛋白酶K法处理的单个卵子的PCR扩增总成功率为92.9%（65/70），二者差异显著（P<0.05）。但两种方法所制备模板的PCR假阳性率均为0。实验设计的KOH/DTT-Triton X裂解法是一种有效的单个早期胚胎的DNA模板制备方法，经一次PCR扩增即能获得清晰的目的DNA条带，能够满足早期胚胎遗传物质检测的需要。

关键词 卵子；胚胎；聚合酶链式反应；DNA模板制备

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# A Method for DNA Template Preparation

LI Jun-Feng, LI Hai-Feng, SONG Yan-Hua, SUN Yan, ZHANG Jia-Hua

Chongqing Key Lab of Forage & Herbivore College of Animal Science and Technology,

**Abstract**

We established a simple methodfor the preparation of DNA template from a single oocyte or early embryo by KOH/DTT-Triton X disintegration. The PCR amplification efficiency of DNA template prepared by this method was compared with that prepared by TE-proteinase K. Single oocyte, 2-cell embryo, 8-cell embryo, morula or blastocyst were separately treated by KOH/DTT-Triton X, then the DNA template was directly used to amplify mitochondrial DNA segment by PCR. The overall PCR success rate of the 3 pairs of primers was 100% (70/70), while the overall PCR success rate of single oocyte treated by TE-proteinase K was 92.9% (65/70). Difference between the two results was significant (P<0.05), and the PCR false positive rates in both groups were 0. The designed KOH/DTT-Triton X disintegrate method was efficient to the preparation of DNA template of a single early embryo. It needed only one cycle of PCR amplification to get clear aimed DNA stripe and the efficiency was high enough to meet the need of early embryonic genetic material detection.

**Key words** oocyte embryo PCR template of DNA preparation

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通讯作者 张家骅 j.zhang@swau.edu.cn