

论著

PCR-RFLP和多重PCR技术检测常见病原性丝状真菌的实验研究

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摘要: 目的 建立检测常见丝状真菌感染病原菌的PCR-RFLP和多重PCR方法。方法 建立以PCR技术为基础的限制片段长度多态性(RFLP)方法,首先用真菌通用引物扩增丝状真菌的ITS区,然后用限制性核酸内切酶对PCR产物进行酶切。用4种丝状真菌的特异性引物建立多重PCR体系,用该体系检测单模板、双模板和三模板的扩增情况,并测定该体系的特异性和敏感性。结果 用PCR-RFLP技术能够鉴别5种常见丝状真菌,多重PCR能够根据扩增片段的不同鉴别菌种,在合适的反应条件下,对单模板、双模板和三模板均能扩增出目的片段。结论 PCR-RFLP和多重PCR技术能够快速鉴定丝状真菌感染病原菌,有临床应用的良好前景。

关键词: 丝状真菌 PCR-RFLP 多重PCR 分子鉴定

Experimental investigation on the detection of pathogenic molds by PCR-RFLP and multiplex PCR

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Abstract: Objective To establish diagnostic methods for pathogenic molds using PCR-RFLP and multiplex PCR. Methods A PCR-based restriction fragment length polymorphism method was developed. ITS region of molds was amplified with universal fungal primers, and for RFLP analysis with restriction enzyme. Multiplex PCR with 4 primer pairs was used to detect 1 template and 2 or 3 template mixtures. Sensitivity and specificity of multiplex PCR were measured. Results PCR-RFLP clearly differentiated the pathogenic molds. Multiplex PCR could amplify the corresponding 1, 2 or 3 DNA fragments. Five molds were identified through distinct amplicons. Conclusions PCR-RFLP and multiplex PCR are rapid methods for the identification of pathogenic molds.

Keywords: mold PCR-RFLP multiplex PCR molecular identification

收稿日期 2009-11-15 修回日期 网络版发布日期

DOI:

基金项目:

科技重大专项(2008ZX10004-002): 临床重点学科建设项目[2007-2009年度卫生部部属(管)医院临床学科重点项目(第7号)]; 卫生部公益性行业科研专项经费项目(200802032)

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