中国寄生虫学与寄生虫病杂志 2001 19 (2): 103-106 ISSN: 1000-7423 CN: 31-1248/R

实验报道

聚合酶链反应检测蚊体内马来丝虫幼虫的实验

王莹,戴晓冬,田晓光,崔昱,李杰,袁晓东,孙德建

大连医科大学寄生虫学教研室!大连116027(王莹;戴晓冬;田晓光;崔昱;李杰);宝生物工程(大连)有限公司!大连116600(袁晓东);中国预防医学科学院寄生虫病研(孙德建)

收稿日期 修回日期 网络版发布日期 接受日期

摘要

目的 建立一种特异、灵敏和简捷的聚合酶链反应 (PCR)检测蚊体内马来丝虫幼虫的方法。方法 在应用一种新的模板纯化处理技术 (Microcon 10 0)基础上 ,采用适应于检测我国马来丝虫的两套DNA扩增引物(P1、P2与P3、P4) ,对实验感染马来丝虫的中华按蚊进行扩增检测。结果 两套引物均可检出蚊体内不同发育期幼丝虫 (L1、L2 和L3) ,其灵敏度达 1只蚊体内 1/ 6 4条L1和 2 0 0只群体蚊中含有1只感染蚊 (体内有 1条L3)的水平 ,而对犬恶丝虫及未感染蚊却不能扩增出特异条带。结论 初步建立特异、灵敏和简捷的PCR检测蚊体内马来丝虫幼虫的方法。

关键词 马来丝虫 蚊媒 DNA PCR

分类号

Studies on Detecting Brugia malayi Larva in Mosquitoes by Polymerase Chain Reaction

WANG Ying [1],DAI Xiao dong [1],TIAN Xiao guang [1],CUI Yu [1],LI Jie [1],YUAN Xiao dong [2],SUN De jian [3]

1 Department of Parasitology;Dalian Medical University;Dalian 116027; 2 Takara Biotechnology Co. Ltd.;Dalian 116600; 3 Institu

Abstract

Objective To establish a specific, sensitive and simple assay for the detection of Brugia malayi larva in Anopheles sinensis .Methods Using a new DNA purification technique (Microcon 100) and two pairs of oligonucleotide primers (p1, p2 and p3,p4) suitable for detecting B malayi in seven areas in our country, the mosquito vectors infected by B malayi were detected by polymerase chain reaction(PCR).Results This PCR method could amplify separately a 322 basepair(bp) and a 155 bp DNA fragment and detect as few as 1/64 of one L 1 in 1 mosquito,the detectable limit was nearly 4 pg DNA of filarial larvae, and it could also detect 1 infected mosquito with one L 3 of B malayi in pools of up to 200 mosquitoes. In contrast,no such specific 322 bp or 155 bp DNA band was detected in Dilofilaria immitis and normal mosquito.Conclusion This PCR techique established for supervision of mosquito vector in B malayi endemic areas is specific,sensitive,and simple.

Key words Brugia malayi mosquito vector DNA PCR

DOI:

通讯作者

作者个人主页 王莹;戴晓冬;田晓光;崔昱;李杰;袁晓东;孙德建