论著

约氏疟原虫卵囊黑化与卵囊内游离钙关系的研究

张锡林*,徐文岳,王英,段建华,况明书

第三军医大学病原生物学教研室,重庆 400038

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摘要

目的研究大劣按蚊蚊胃的约氏疟原虫卵囊黑化时卵囊内Ca~(2+)的变化,探讨疟原虫感染不易感蚊种时卵囊黑化与囊内Ca~(2+)的相互关系。方法以荧光染料Fluo-3/Am作卵囊内钙指示剂,应用共聚焦激光扫描显微镜(CLSM)观察不同时期约氏疟原虫卵囊内Ca~(2+)分布、变化以及检测的实验条件。结果3μmol/LFluo-3/Am同时加入1μl/ml25%PluronicF-127在37℃恒温下,孵育约氏疟原虫卵囊60min即可达到最佳的负载效果。Fluo-3/Am浓度的提高和孵育时间延长只能增加背景染色,若降低孵育浓度和缩短孵育时间则难于达到最佳的负载效果。约氏疟原虫成熟卵囊的囊内Ca~(2+)为(137.15±7.02)nmol/L,完全黑化卵囊为(18.44±1.75)nmol/L,并在囊壁出现明显的钙沉着。结论约氏疟原虫卵囊完全黑化时,囊内Ca~(2+)明显减少,并有外排现象。

关键词 约氏疟原虫 卵囊 黑化 Ca~(2+) 共聚焦激光扫描显微镜(CLSM)

分类号

Study on the Relationship between Intracellular Free Calcium and Melanization in Oocysts of Plasmodium yoelii

ZHANG Xi-lin, XU Wen-yue, WANG Ying, DUAN Jian-hua, KUANG Ming-shu

Department of Pathabiology, the Third Military Medical University, Chongqing 400038

Abstract

Objective To study the change of intracellular free Ca2+ in the oocyst when it melanized and to find out the relationship between the melanized oocyst and its intracellular level of free Ca2+ in a Plasmodium-refractory strain of Anopheles dirus. Methods The distribution and experimental condition of the intracellular free Ca2+ in oocyst of Plasmodium yoelii was measured with Ca2+ sensitive dye Fluo-3/AM and Plutonic F-127 under confocal laser scanning microscope (CLSM) at different time. Results The best load condition was that the oocysts were incubated in 3 μ mol/ml Fluo-3/AM adding 1 μ l/ml 25% Pluronic F-127 for 60 min at 37 $^{\circ}{\rm C}$. Fluorescent imaging of oocysts was affected by an increase or decrease of the concentration of Fluo-3/Am and incubation time. The distribution of intracellular free Ca2+ was heterogeneous in the oocysts. The mean value of Ca2+ in the mature oocysts was (137.15 \pm 7.02) nmol/L (X \pm S) but was (18.44 \pm 1.75) nmol/L in melanized oocysts with Ca2+ sedimentation in the wall of oocyst. Conclusion The results suggest that the level of the intracellular free Ca2+ in oocyst decreased and excreted during its melanization in a Plasmodium-reiractory anopheline mosquito species.

Key words Plasmodium yoelii oocyst nnelanization Ga2+ CLSM

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通讯作者

作者个人主

元 张锡林*;徐文岳;王英;段建华;况明书

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