DIFFERENCES BETWEEN in vitro AND in vivo OBTAINED SCHISTOSOMULES1

Alan L. MELO (2), Conceição R. S. MACHADO (3), & Leógenes H. PEREIRA (2)

SUMMARY

The injection of cercariae of **Schistosoma mansoni** into the peritoneal cavity of naive mice induces cell adhesion to these larvae, and this adherence sharply decreases when the infecting larva changes to schistosomule. This procedure was used to detect differences between schistosomules obtained in vivo and in vitro. Reinoculation of schistosomules obtained in vivo into the peritoneal cavity of mice did not trigger cell adhesion. In contrast, adherent cells were found in 4 and 24-hour-in vitro schistosomules. Our data on schistosomules obtained in vitro indicate that more than 24 hours are needed for complete remotion of molecules involved in the phenomenon of cell adhesion.

KEY WORDS: Schistosoma mansoni; transformation; Schistosomula; in vivo, in vitro.

INTRODUCTION

Cercariae of Schistosoma mansoni after release from a molluscan host penetrate the skin of the vertebrate host in order to continue their life cycle. So, the parasite undergoes many structural and physiological changes, originating schistosomules which mature to adult worms adapted to the bloodstream.

The difficulties for the recovery of schistosomules from the skin led to the development of several techniques for transforming cercariae into schistosomula, in vitro. The schistosomule-host cell interactions have been also studied, mainly through in vitro experiments. However, schistosomules can be easily obtained in vivo by inoculating cercariae into the peritoneal cavi-

ty of mice^{2, 3, 4}. In this study, the kinetics of cell adhesion to **Schistosoma mansoni** larvae developing in the peritoneal cavity of naive mice was used to compare **in vitro** and **in vivo** obtained schistosomules.

Aiming at obtaining in vivo schistosomules, S. mansoni cercariae (LE strain) shed by laboratory reared and infected Biomphalaria glabrata were concentrated, and 0.5 ml of well water containing about 500 cercariae was injected in albino mice (males, weighing 18-22 g), intraperitoneally. The mice were killed by cervical fracture 4,24 hours and 7 days after cercarial inoculation. The larvae recovered by washing the peritoneal cavity with saline were concentrated by centrifu-

⁽¹⁾ Supported, in part, by the Parasitic Diseases Programme-WHO, and CNPq, FINEP, FAPEMIG, Brasil.

⁽²⁾ Departamento de Parasitologia, Instituto de Ciências Biológicas da Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brasil.

⁽³⁾ Departamento de Morfologia, Instituto de Ciências Biológicas da Universidade Federal de Minas Gerais. Belo Horizonte. MG. Brasil.

Address for correspondence: A.L.M. GIDE-ICB/UFMG — Caixa Postal 2486. CEP 30161 Belo Horizonte, MG, Brasil.

gation. The in vitro schistosomules (4 hours, 24 hours, and 7 days) were obtained according to RAMALHO-PINTO et al.5. The in vitro and in vivo obtained schistosomules were inoculated into the peritoneal cavity of naive mice (groups of 10 animals) and recovered 30 and 180 minutes later, as described above. Reinoculation of in vivo obtained schistosomules (4, 24 hours and 7 days old) did not trigger cell adhesion. In contrast, adherent cells, mainly neutrophils, were present in 87.7% (4 hours) and 85.2% (24 hours) of the in vitro obtained schistosomules, recovered 30 minutes after inoculation. All in vitro obtained schistosomules recovered 180 minutes after inoculation were free of host cells. Cell adhesion was not induced by 7-day-old schistosomules obtained in vitro (either after 30 or 180 minutes post-inoculation).

It seems very likely that the attachment and subsequent detachment of cells to S. mansoni larvae in vivo are both related to surface molecules that are lost during the process of transformation. The glycocalyx is the most probable structure involved in cell adhesion. In contrast to the in vivo obtained schistosomules, the in vitro obtained organisms were able to induce host cell adhesion when inoculated into the mice peritoneal cavity. Comparative studies on the development of schistosomules produced by several techniques showed that the degradation of glycocalyx is slowered in artificially derived organisms, and may be not complete until 24 hours1. Our data on schistosomules obtained in vitro indicate that more than 24 hours are needed for the complete remotion of molecules involved in the phenomenon of cell adhesion.

RESUMO

Diferenças entre esquistossômulos obtidos in vitro e in vivo.

Injeção de cercárias de Schistosoma mansoni na cavidade peritoneal de camundongos normais induz adesão celular a estas larvas. Esta aderência diminui acentuadamente quando as larvas infectantes se transformam em esquistossômulos. Este procedimento foi usado para detectar diferenças entre esquistossômulos obtidos in vivo e in vitro.

A reinoculação de esquistossômulos obtidos in vivo na cavidade peritoneal de camundongos não acarreta adesão celular. Por outro lado, células aderentes foram encontradas em esquistossômulos obtidos in vitro (4 e 24 horas, respectivamente). Nossos dados referentes a esquistossômulos obtidos in vitro indicam que mais de 24 horas são necessárias para a completa remoção de moléculas envolvidas no fenômeno de adesão celular.

REFERENCES

- COUSIN, C. E.; STIREWALT, M. A. & DORSEY, C. H.
 Schistosoma mansoni: comparative development of schistosomules produced by artificial techniques. J. Parasit., 72: 606-609, 1986.
- CRAM, E. B. & BOZICEVICH, J. Experimental Schistosoma mansoni infection by intraperitoneal route. Trop. Med. News. 1: 16-17, 1944.
- EVELAND, L. K. Schistosoma mansoni: conversion of cercariae to schistosomula. Exp. Parasit., 32: 261-264, 1972
- PEREIRA, L. H.; PELLEGRINO, J.; VALADARES, T. E.; MELLO, R. T. & COELHO, P. M. Z. — A new approach for screening prophylactic agents in schistosomiasis. Rev. Inst. Med. trop. S. Paulo, 16: 123-126, 1974.
- RAMALHO-PINTO, F. J.; GAZZINELLI, G.; HOWELLS, R. V.; MOTA-SANTOS, T. A.; FIGUEIREDO, E. A. & PEL-LEGRINO, J. — Schistosoma mansoni: Defined system for stepwise transformation of cercaria to schistosomule in vitro. Exp. Parasit., 36: 360-372, 1974.

Recebido para publicação em 27/06/1989.