Zeitschrift: Acta Tropica

**Herausgeber:** Schweizerisches Tropeninstitut (Basel)

**Band:** 40 (1983)

Heft: 1

**Artikel:** Uptake of promastigotes of a lizard "Leishmania" sp. and "Leishmania"

donovani" by mouse peritoneal macrophages: short communication

Autor: Olobo, J.O. / Mutinga, M.J.

**DOI:** https://doi.org/10.5169/seals-313118

### Nutzungsbedingungen

Die ETH-Bibliothek ist die Anbieterin der digitalisierten Zeitschriften auf E-Periodica. Sie besitzt keine Urheberrechte an den Zeitschriften und ist nicht verantwortlich für deren Inhalte. Die Rechte liegen in der Regel bei den Herausgebern beziehungsweise den externen Rechteinhabern. Das Veröffentlichen von Bildern in Print- und Online-Publikationen sowie auf Social Media-Kanälen oder Webseiten ist nur mit vorheriger Genehmigung der Rechteinhaber erlaubt. Mehr erfahren

#### **Conditions d'utilisation**

L'ETH Library est le fournisseur des revues numérisées. Elle ne détient aucun droit d'auteur sur les revues et n'est pas responsable de leur contenu. En règle générale, les droits sont détenus par les éditeurs ou les détenteurs de droits externes. La reproduction d'images dans des publications imprimées ou en ligne ainsi que sur des canaux de médias sociaux ou des sites web n'est autorisée qu'avec l'accord préalable des détenteurs des droits. En savoir plus

#### Terms of use

The ETH Library is the provider of the digitised journals. It does not own any copyrights to the journals and is not responsible for their content. The rights usually lie with the publishers or the external rights holders. Publishing images in print and online publications, as well as on social media channels or websites, is only permitted with the prior consent of the rights holders. Find out more

**Download PDF:** 30.11.2025

ETH-Bibliothek Zürich, E-Periodica, https://www.e-periodica.ch

International Centre of Insect Physiology and Ecology (ICIPE), P. O. Box 30772, Nairobi, Kenya

# Uptake of promastigotes of a lizard *Leishmania* sp. and *Leishmania donovani* by mouse peritoneal macrophages

# **Short communication**

J. O. Olobo, M. J. Mutinga

# Introduction

Leishmania are parasitic protozoa. They exist as two well defined forms: the promastigotes in the insect vectors and culture media and the amastigotes in mononuclear phagocytes in mammalian hosts and tissue culture.

Apparently *Leishmania* species that can initiate infections in a given host have developed the ability to resist the effects of activated macrophages of that host, but are readily destroyed by cells of an unsusceptible host. For example, mouse macrophages do not destroy *L. tropica* which infects mice but do kill *L. enriettii*, which fails to infect mice (Mauel et al., 1978). There have been a number of studies on interactions of *Leishmania* from different geographical regions with peritoneal macrophages of different animal species (Chang and Dwyer, 1976; Handman and Spira, 1977). Similar investigations on *Leishmania* isolates from Kenya are scarce in literature. This work was aimed at elucidating more the uptake of promastigotes of lizard *Leishmania* sp. and *Leishmania donovani* by BALB/c macrophages.

#### Materials and Methods

Two Leishmania isolates were used in this study, a lizard and a human strain. The lizard Leishmania (Liz. 1, ICIPE 140) was isolated from Kacheliba, Kenya in 1976 through the inoculation of heart blood of a lizard Mabuya natalensis into NNN medium. After some unrecorded passages, the promastigotes were frozen as stabilates initially in CO<sub>2</sub> until 1979 when they were transferred to liquid nitrogen.

Leishmania donovani was isolated from a patient in Kenya with a clinical case of kala-azar in 1979. Splenic aspirates were cultured in NNN medium and the promastigotes were stabilated and stored in liquid nitrogen as ICIPE 126 after a number of unrecorded passages.

For these experiments, promastigotes of the two Leishmania strains were grown at 25°C in

Correspondence: Dr. J. O. Olobo, International Laboratory for Research on Animal Diseases (ILRAD), P.O. Box 30709, Nairobi, Kenya

Hepes buffered (25 mM) Dulbecco's Modified Eagle Medium (DMEM, Gibco) containing penicillin (100 units/ml), streptomycin (100  $\mu$ g/ml) and heat inactivated foetal calf serum (FCS, 20% v/v, Gibco). They were harvested at peak growth, washed and counted in a haemocytometer.

Mice of inbread BALB/c strain used, were 7–8 weeks old at the commencement of the experiments. Peritoneal exudate cells were stimulated with 0.5 ml of sterile liquid paraffin. Three days later each mouse was injected intraperitoneally (i.p.) with  $2 \times 10^7$  promastigotes. The experimental mice were killed at 1 h intervals up to 4 h and at 24 h and their peritoneal cavities washed with 3 ml of DMEM containing heparin (5 units/ml) and antibiotics. Cells collected were pooled accordingly, washed and resuspended in Hepes buffered DMEM containing antibiotics and heat inactivated foetal calf serum. They were added in 0.5 ml volumes to  $18 \times 18$  mm coverslip in Petri dishes and incubated in a humid atmosphere for 2 h at 37° C. Non adherent macrophages were washed in cold phosphate buffered saline before the coverslips were dried, fixed and Giemsa-stained for light microscopy. The level of infection was determined by counting at least 400 macrophages per coverslip.

## **Results and Discussion**

The human and lizard Leishmania/mouse macrophage model system presented here was used to try to simulate the situation in nature where promastigotes of some Leishmania species invade macrophages when deposited in the skin of hosts by bites of sandflies and may undergo transformation to amastigotes which either survive or are killed. When  $2 \times 10^7$  promastigotes were injected intraperitoneally into BALB/c mice, the rate of infection of macrophages was  $8-10\% \pm 1\%$  for the lizard *Leishmania* sp. and  $7-8 \pm 1\%$  for *L. donovani* at 1–4 h, with an average of 2 amastigotes per cell for each of the isolates. In a few cases, mice infected with the lizard isolate had 8–12 parasites per macrophage. Liz. I amastigotes were observed to be round or oval in shape. No clear evidence of vacuole development around amastigotes of the lizard isolate was noticed as compared to L. donovani where there was some vacuole development (not shown). At 24 h, intact Liz. 1 amastigotes could not be seen but granules and clusters which appeared to be disintegrated parasites were numerous within the macrophages. L. donovani amastigotes were, however, clearly visible at this time with distinct outlines and the level of macrophage infection by this parasite isolate was similar to that found after 1-4 h of infection indicating that the parasites had probably not multiplied by this time.

These results suggest that the lizard parasites used in this study could infect warm blooded mammals in nature. But the transient parasitism means that finding the parasites in mammals would be a very rare event. It is common knowledge that prolonged in vitro cultivation of *Leishmania* promastigotes may influence their capability to infect macrophages, a factor which could have had some effects on the two parasite isolates used in this study. But efforts were made to propagate the parasites for the minimal time in vitro and promastigotes were obtained from the original stabilates for new experiments.

Earlier workers have reported similar findings of transient infections after some human volunteers and other lower warm blooded mammals were inoculated with promastigotes of reptilian origin (Manson-Bahr and Heisch, 1961; other literature cited in Belova, 1971). Mutinga and Ngoka (1980) reported apparently fewer cases of human leihmaniasis in certain areas in Kenya where many lizard *Leishmania* isolations had been made than in areas with no lizard *Leishmania*. They hypothetized that *Leishmania* of lizards may play some role in conferring immunity against human infections. But this remains to be thoroughly investigated. The transformation of this lizard *Leishmania* sp. into amastigote forms, in mouse macrophages which normally supports growth of *L. donovani*, is suggestive of some relationship between lizard and true mammalian *Leishmania*. More studies are being conducted in this laboratory to elucidate further the role of reptiles in *Leishmania* epidemiology in Kenya.

A

#### Acknowledgments

This study was supported in part by grants from the UNDP/WORLD BANK/WHO Special Programme for Research and Training in Tropical Disease and the International Centre of Insect Physiology and Ecology (ICIPE). We are grateful to Professor Thomas R. Odhiambo, Director of ICIPE, for his guidance and encouragement during this investigation.

- Belova E. M.: Reptiles and their importance in the epidemiology of leishmaniasis. Bull. Wld Hlth Org. 44, 553–660 (1971).
- Chang K. P., Dwyer D. M.: Multiplication of human parasites (*Leishmania donovani*) in phagolysosomes of hamster macrophages in vitro. Science 193, 678–680 (1976).
- Handman E., Spira D. T.: Growth of *Leishmania* amastigotes in macrophages from normal and immune mice. Z. Parasitenk. 53, 75 (1977).
- Manson-Bahr P. E. C., Heisch R. B.: Transient infection of man with a *Leishmania* (*L. adleri*) of lizards. Ann. trop. Med. Parasit. 55, 381–382 (1961).
- Mauel J., Buchmuller Y., Behin R.: Studies on the mechanism of macrophage activation. 1. Destruction of intracellular *Leishmania enriettii* in macrophages activated by cocultivation with stimulated lymphocytes. J. exp. Med. *148*, 393 (1978).
- Mutinga M. J., Ngoka J. M.: Suspected vectors of lizard leishmaniasis in Kenya and their possible role in partial immunization of the human population against *L. donovani* in kala-azar endemic areas. Insect Sci. Applications 1, 207–210 (1980).

