First successful in vitro culture of autochthonous Leishmania sp. in Sri Lanka

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Summary
Since the first autochthonous case of cutaneous leishmaniasis in Sri Lanka was reported in 1992 (1) attempts to culture the causative organisms have been unsuccessful. We report the first successful isolation of the local Leishmania sp. by in vitro culture, which would pave the way for species and strain identification.

Introduction
Whether cutaneous leishmaniasis is endemic in Sri Lanka is still uncertain in spite of the abundance of sandfly vectors in some parts of the island (2). A few cases of the infection in people returning from the middle east have been reported in the past (3). The first autochthonous case in Sri Lanka was reported in 1992 from the Southern Province of the island (1). Another case was reported from the central region (4). Since then a few more cases of cutaneous leishmaniasis have been diagnosed in our department indicating a scattered distribution of the parasite and its vector, but the diagnosis of these was based on the identification of amastigote forms in direct smears made from lesion aspirates or biopsy specimens.

Case reports
Three patients with typical leishmaniasis lesions from the National Hospital of Sri Lanka were referred to us. Two of them were men soldiers who had served for several months in the jungles of the North Central Province. The third was a schoolgirl from North Western Province. None of them had travelled abroad. All had lesions on the head, mainly on the face. No lymphadenopathy or hepatosplenomegaly was detected. The formol-gel test done on two patients was negative.

Materials and methods
Three standard media (5), NNN (Nicolle, Novey and McNeil), USAMRU (Difco Blood agar) and Evan’s modified Tobie’s, were used for culture. As overlay for Evan’s medium 1% glucose saline with nystatin (250 iu/ml) was used. Saline aspirates from the lesions were inoculated into each medium. Culture tubes with agar slopes were kept in the dark in an air-conditioned room at a temperature range of 25 to 27°C. On the fifth day smears made from cultures with Evan’s medium were positive for promastigotes. The other two media became positive on the ninth day. Parasites were seen in all 3 culture media tested with isolates from 2 patients. The cultures of the third were found to be contaminated and could not be used. A few stabilates made from positive cultures were stored at -70°C for future use. Presently animal inoculation in hamsters and mice is being done to make species and strain identification.

Comment
Successful isolation by in vitro culture of the causative organism of autochthonous cutaneous leishmaniasis in Sri Lanka would pave the way for definite species and strain identification. This is important for the development of control methods and proper treatment of locally acquired cutaneous leishmaniasis in Sri Lanka, the incidence of which seems to be increasing.

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References