HUMAN CUTANEOUS LEISHMANIASIS DUE TO A NEW ENZYMATIC VARIANT OF *LEISHMANIA (VIANNIA) BRAZILIENSIS* OCCURRING IN PERNAMBUCO, BRAZIL

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Both American cutaneous (ACL) and visceral (AVL) leishmaniasis are endemic in Pernambuco State, Brazil (R. Badaró, 1987, In B. C. Walton et al., eds. *Proc. Intern. Workshop*, Ottawa, Canada, June, 1-4). In the past, ACL was a zoonosis, occurring in people having contact with primary (Atlantic) rain forest. Environmental changes, due to the opening of forested lands to new agricultural and natural resource development, modified the epidemiology of the disease and new endemic areas were reported.

Between 1979 and 1986, a total of 58,913 cases of cutaneous leishmaniasis were reported in Brazil; only 728 (1.2%) of these patients were from Pernambuco (R. Badaró, 1987, loc. cit.). However, between 1989 and 1991, aproximately 1,900 new cases were recorded in the state (FNS, Ministry of Health). Here we extend these studies, describing ACL in Pernambuco caused by infection with *L. (V.) braziliensis*.

The study area where the patients originated is located in the municipality of Cortes, in the south-east region ("Zona da Mata") of Pernambuco, Brazil (at 8° 29'S and 35° 35'W). During an epidemic outbreak of ACL in this area in 1991, we examined several confirmed human cases of the disease. Diagnosis was based on detection of amastigotes in lesion biopsy smears, by the microscope, and isolation of the parasite in culture media, the position

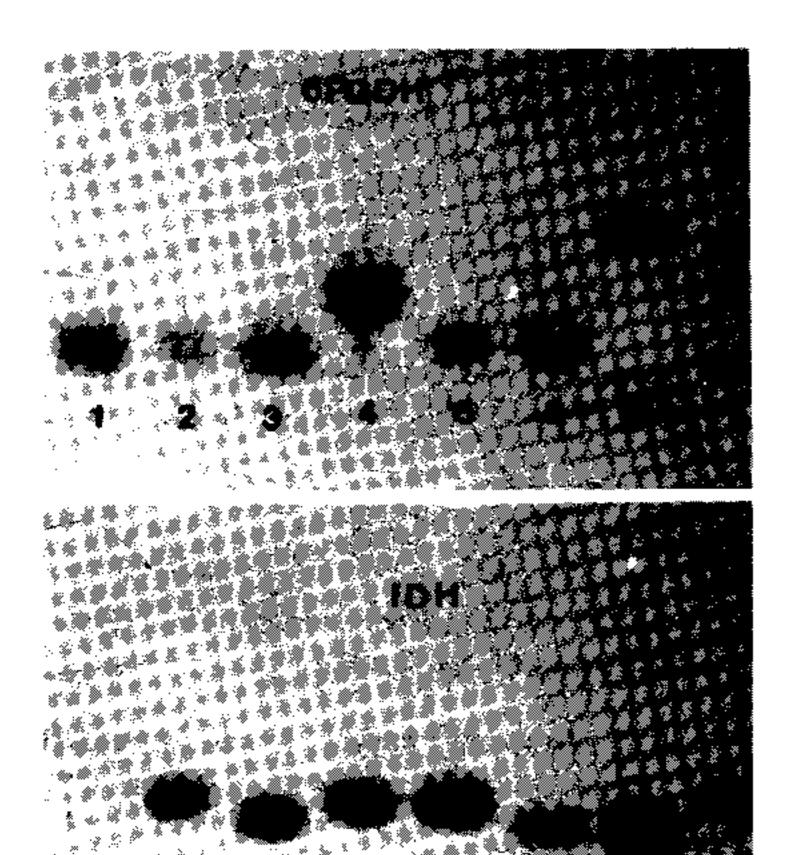
tive Montenegro intradermal reaction and clinical appearance of the lesions.

A dog serological survey was undertaken in the area of this study. About 18.5% of the canine population showed anti-Leishmania antibodies, as revealed by an indirect immunofluorescent assay, but none of the examined animals had skin ulcers. In addition, a preliminary survey for suspected vectors indicated that Lutzomyia whitmani is the most commonly found sandfly species and may be implicated in the transmission of this parasite (S. P. Brandão F° & M. L. B. Oliveira, 1992, Mem. Inst. Oswaldo Cruz, 87, Supl. II: 211).

Here we have compared two leishmanial parasites (designated with stock codes MHOM/ BR/91/JRS and MHOM/BR/91/SSS), isolated from humans in the study area, with other Leishmania species. On the basis of enzyme profiles the stocks were characterized as a new variant of L. (V.) braziliensis. Results of electrophoretic analysis were based on 18 enzyme loci and the development conditions for these enzymes have been reported previously (E. Cupolillo et al. 1992, Am. J. Trop. Med. Hyg., in press). The two strains from Pernambuco showed similar electromorphs (bands of enzyme activity as revealed by electrophoresis) in all enzymes examined, and they were grouped into a new zymodeme (IOC-45). The enzyme profiles of this zymodeme had similar electromorphic profiles to L. (V.) braziliensis stricto sensu except for the enzymes Isocitrate dehydrogenase, IDH-NADP and Phosphoglucose mutase, PGM (Fig.) which could distinguish this new enzymatic variant from either the WHO L. (V.) braziliensis reference strain (MHOM/BR/75/M2903) or other variants (G. Grimaldi et al., 1991, Am. J. Trop. Med. Hyg., 44: 645-661; E. Cupolillo et al., 1992, loc. cit.).

This work was supported in part by the Brazilian National Council of Scientific Development and Technology (CNPq), and UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases.

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Photographs of agarose gels stained for activity of 6-phosphate dehydrogenase (6PGDH), showing no differences among $Leishmania\ braziliensis$ strains; and Isocitrate dehydrogenase (IDH) showing differences between the new zymodeme (slot 1 & 3) and L. braziliensis reference strain (slot 2).

The following strains from Brazil were analyzed in this study: 1. MHOM/BR/91/JRS, L. braziliensis variant from Pernambuco; 2. MHOM/BR/75/M2903, L. braziliensis from Pará; 3. MHOM/BR/91/SSS, L. braziliensis variant from Pernambuco; 4. MHOM/BR/75/M4147, L. guyanensis from Pará; 5. MHOM/BR/90/HF-3, L. braziliensis from Espírito Santo; 6. MHOM/BR/91/AJF, L. braziliensis from Rio de Janeiro; 7. IFLA/BR/67/PH8, L. amazonensis from Pará.

This is the first report on the characterization and identification of leishmanial parasites from Pernambuco. The physical geography and ecology of this region is similar to that already described for other endemic areas of ACL, where the original forest environment has been altered (W. Mayrink et al al., 1979; Ann. Trop. Med. Parasitol., 73: 123-137; A. Falqueto et al., 1991, Mem. Inst. Oswaldo Cruz, 86: 499-500).

These data support other investigations, showing that ACL due to L. (V.) braziliensis occur in old well established communities, in non-forested areas, where the maintenance cycle seems to involve domestic animal reservoirs and phlebotomine sand flies with peridomiciliary habitats. However, the demonstration of a new enzymatic variant of this parasite suggests that a natural enzootic focus of the infection may also occur in the region. In this respect, it should be mentioned that there are sugar-cane cultures and remainders of Atlantic forest surrounding the area of study. In addition, wild rodents captured in this area were found to be infected with Leishmania (unpublished results).