

LEISHMANIA SPP. INFECTION IN FIVE HORSES FROM COSTA RICA

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INTRODUCTION

Leishmaniasis is one of the most prevalent parasitic public health problems worldwide. Is caused by a protozoal genus *Leishmania*. Aside from humans (cutaneous and visceral form), it has been published in dogs, cats and cows. In horses, it has been reported mostly in tropical and subtropical areas such as a Mediterranean basin, southern and central Europe and in America (Brazil and Puerto Rico) In the present study, five autochthonous cases of equine cutaneous leishmaniosis due to *Leishmania spp* complex species in Costa Rica are described.

RESULTS.

A- Clinical findings:

Signalment: there were four Andalusian and one American Pinto; four females and one male. Three horses were 2.5 years old or younger. One was 4 years and the other was reported as an adult one. The main clinical presentation was alopecic nodules or ulcerated and crust formation, affecting mostly the pinna (2 left, one right and one bilateral), with one case showing an ulcer lesion on the left neck. The presentation was chronic (one to two months). See Fig.1.



Figure 1. Three different clinical lesions (ears). A. Single ulcerated nodule (the hole is the punch site). B. There are several subcutaneous nodules, one is ulcerated. C. The inner pinna surface shows crust formation.

B- Histopathological findings:

The skin biopsies were fixed in 10 % buffered formalin. Then embedded in paraffin, sectioned at 5- μ m, and stained with hematoxylin-eosin. Besides in all cases special stains were done (Giemsa, P.A.S. and Fite). In three cases the dermis showed a diffuse cellular infiltration predominately with macrophages fill up countless microorganisms round to oval of 2 to 3 μ m morphology that pointed out the possibility of *Leishmania spp*. Moreover, multinucleated giant cells (MNGCs), lymphocytes and neutrophils were also presented. In the other two cases aside of the same inflammatory cellularity, there were coagulative necrotic areas. See Fig.2.

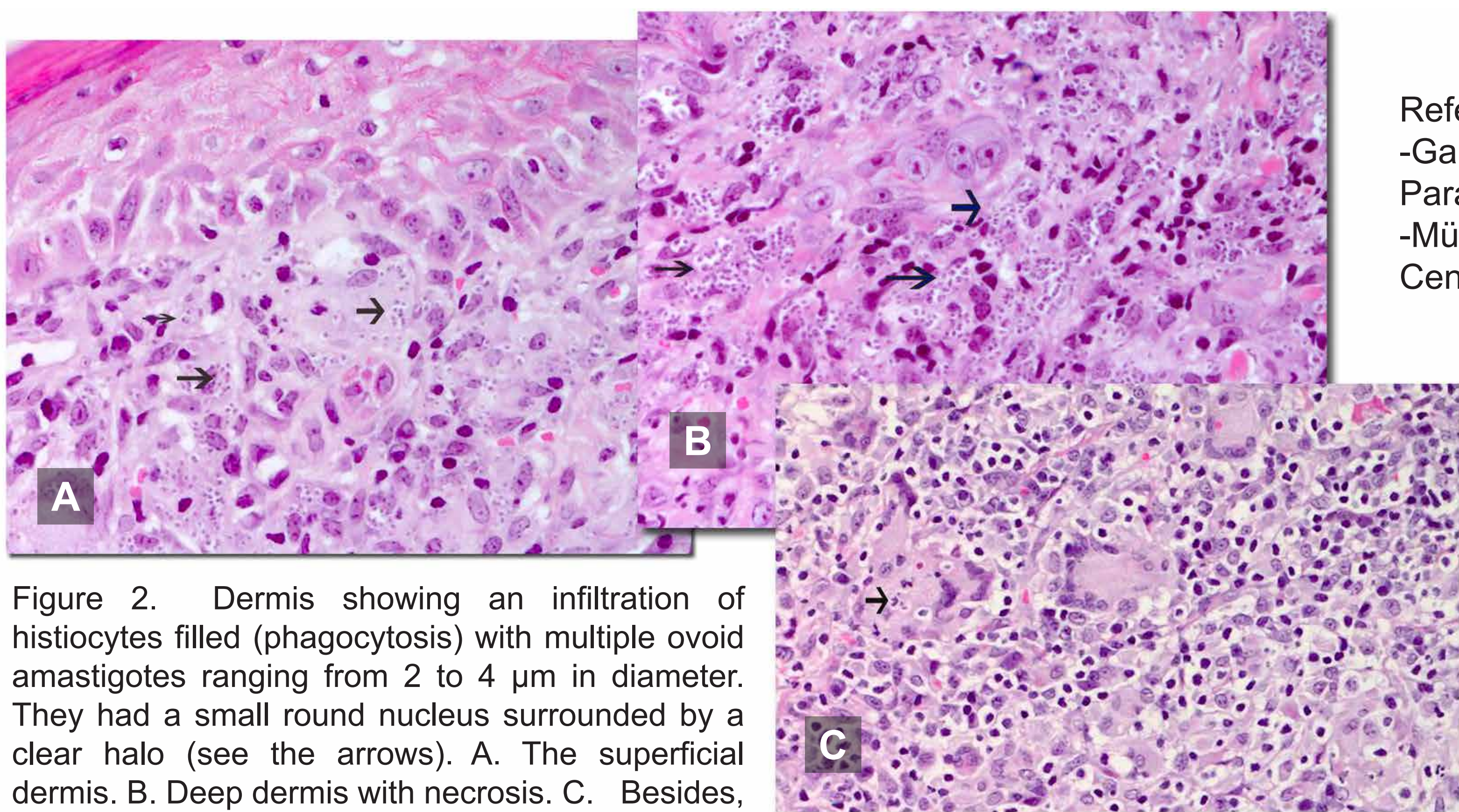


Figure 2. Dermis showing an infiltration of histiocytes filled (phagocytosis) with multiple ovoid amastigotes ranging from 2 to 4 μ m in diameter. They had a small round nucleus surrounded by a clear halo (see the arrows). A. The superficial dermis. B. Deep dermis with necrosis. C. Besides, multinucleated cells are also present.

The special stains help to eliminate a possible mycotic infection; also allowing the see better the morphology of the microorganisms.

C- Immunohistochemistry findings:

An immunohistochemical detection of *Leishmania spp*. was conducted in paraffin tissue section using a rabbit polyclonal antibody raised against *Leishmania donovani*, *L. infantum* and *L. amazonensis* and an established protocol for diagnostics at the School of Veterinary Medicine, University of Surrey, United Kingdom. Tissue sections were obtained for laser-capture microdissection (LCMD) followed by PCR and sequence analysis to identify the species of *Leishmania spp*. LCMD samples are being analysed for *Leishmania spp*. DNA detection and sequencing in order to identify the species and type circulating in these animals, although the quantity of extracted DNA is quite limited and different extraction/amplification protocols are currently being used. Nevertheless, strong positive reaction was observed within the cytoplasm of macrophages and MNGCs where amastigotes were clearly identified. See Figure 3.

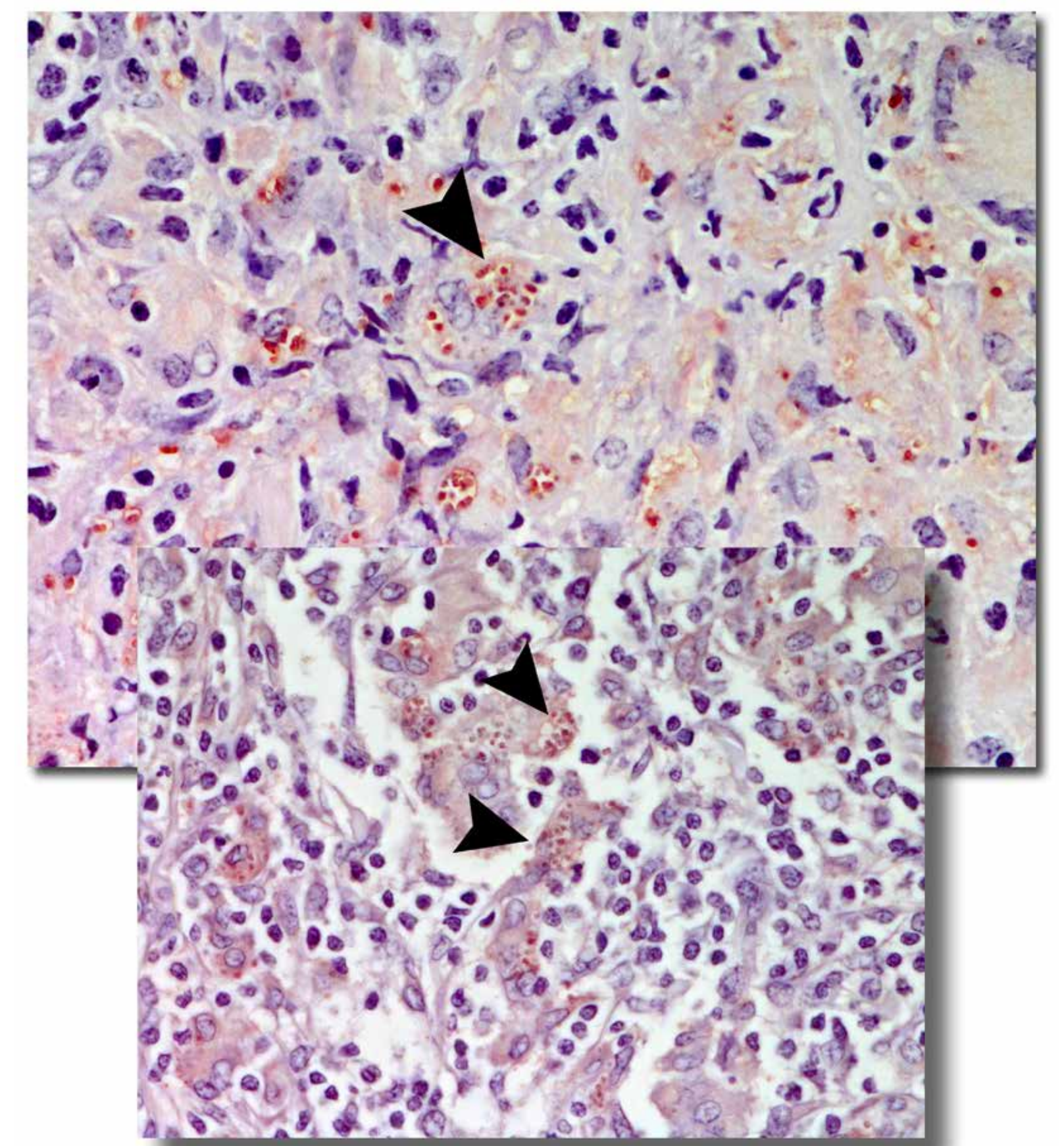


Figure 3. Immunoreactivity to *Leishmania spp*. amastigotes is observed in macrophages, within the inflammatory cell infiltrate and in macrophages within granulomas as well as of extracellular stages (arrowheads).

DISCUSSION AND RESULTS:

Humans leishmaniosis is present in the southern USA, Mexico, Central and South America, with Brazil and Peru reporting the majority of cases. In Costa Rica, New World cutaneous leishmaniosis is common in rural areas, but can also be acquired in semi-urban and urban areas. It also takes place in rainforests and arid areas. The etiological agent most frequently involved in cutaneous leishmaniosis is *L.(Viannia) panamensis*, although *L. (V.) braziliensis* has been reported.

This report shows the importance of *Leishmania spp*. as a causative agent of equine cutaneous disease in the new world and the importance as a possible emerging pathogen. The role of equines in the epidemiology of Leishmaniasis in the New World should be studied in depth. In horses, aside from South American countries mostly Brazil, the diseases has been published in Puerto Rico, been these five cases reported in Central America.

Finally, this report enhances the awareness of Leishmaniasis in the differential diagnosis of chronic equine dermatitis.

References:

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