## CYTOMOPHOMETRIC AND MOLECULAR ANALYSIS OF Leishmania donovani IN CUTANEOUS LEISHMANIASIS PATIENTS IN SRI LANKA

## P. M. A. Gunasekara

Postgraduate Institute of Science (PGIS), University of Peradeniya, Peradeniya, Sri Lanka Department of Parasitology, University of Peradeniya, Peradeniya, Sri Lanka

*Leishmania donovani* Zymodeme MON-37 has been identified as the etiological agent of cutaneous leishmaniasis (CL) in Sri Lanka. There are two forms of the parasite. Intracellular amastigote form found in vertebrate host and the promastigote form seen in the sand fly vector. Diagnosis of CL in Sri Lanka is mainly based on microscopic detection of amastigotes in skin smears. Previous studies done in other countries have reported varied cytomophometrics of *Lieshmania* parasite. Therefore, this study was carried out to identify cytomorphometric features of *Leishmania* parasite in CL patients and to co-relate the morphological differences with the chronicity of the lesions and the demography of patients.

A hundred and thirty samples (n= 130) were stained with Giemsa and examined using the a light microscope under oil emersion (X100). Light microscopic photos were taken for comparison of cytomorphometric features of *Leishmania* parasite. Furthermore, PCRs were performed for the identification of parasite.

Two different morphological forms were identified. They were amastigote forms and promastigote-like structures. Out of the 130 cases, 84 (60.83%) showed presence of amastigote form. In 3 (2.31%) amastigotes were present in intracellular locations while 43 (33.08%) had extracellular presence of amastigotes. In 19 (14.62%) cases, amastigotes were observed in both intracellular and extracellular locations simultaneously. All amastigotes observed were varied from 2- 5 µm in size. Promastigote-like structures were seen in 65 (50%) cases and they were associated with amastigote forms in 19 (14.62%) cases. Out of the 130 cases, promastigote-like form alone (no amastigotes association) was observed in 46 (35.38%) cases. The promastigote- like structures had tail-like features. They lie within 6-35 µm in size. The presence of the amastigotes was predominant in lesions with less than 2 months duration and promastigote- like structures were more common in 8 to 12 months old lesions. This finding leads to suggest that cytomorphometric features are varied with duration of lesions. The findings based on PCR and DNA sequencing determined that the present isolate (SLisolate) was Leishmania donovani. This study will help to add an additional benefit to get a holistic approach for the traditional microscopic method in diagnosing CL in Sri Lanka. Other than dot and dash appearance, cytomorphometric features which support to diagnosis of CL must not be ignored as they may help to diagnose in clinically suspected CL patients.