Leishmania spp.

**PRINCIPLE**

*Leishmania* is found in macrophages around the point of infection.

**SPECIMEN**

Scrapings from infected areas.

**SAFETY**

Blood tubes should only be opened in a running biological safety cabinet. Any spills should be cleaned up immediately. Gloves must be worn when preparing samples and any contact with contaminated sharps should be reported. *Leishmania* can be transmitted from blood samples, therefore follow up of exposure is recommended.

**REAGENTS**

- Absolute methanol
- Giemsa stain.
- NNNN media (see appendix)

**PROCEDURES**

*This is considered to be a non-routine procedure therefore it should only be performed by experienced personnel.*

1) **Smears**

   a) Prepare smears - from touch preps of biopsy material or from an aspirate from the edge of a lesion.

   b) Dry and fix in methanol.

   c) Stain with Giemsa (as for malaria smears) and examine under oil immersion (X1000) for the presence of amastigotes.
**In Vitro Cultivation**

a) Inoculate biopsy material or aspirate from the lesion to prewarmed (room temperature) NNN media culture tubes and incubate at room temperature. Attending physician inoculates specimen at bedside.

b) Examine culture using a wet prep at 10, 14, and 21 days for the presence of promastigotes.

**QUALITY CONTROL**

- Make sure that media is not outdated or contaminated.
- Ensure that the microscope has been calibrated in the last year and that the results of the calibration are displayed on the microscope base.
- Include a control slide if Giemsa Stain is used. Red cells should stain grayish, white cell nuclei stain red-purple and cytoplasm stains bluish.

**REPORT**

Report the presence of *Leishmania* sp. promastigotes from culture and amastigotes from biopsies or aspirates. Definitive species identification requires isoenzyme analysis or molecular methods.

**AUTHOR**

Ian Crandall

**REFERENCES**