



BANK OF STRAINS OF LEISHMANIA



by

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Promastigotes of 60 strains of Leishmania and amastigotes of 12 strains of Leishmania are held in liquid nitrogen in the Department of Parasitology of the London School of Hygiene and Tropical Medicine.

1. METHODS

Promastigotes have been maintained in Noguchi-Wenyon culture. Before freezing, the promastigotes were transferred to 4N cultures for which the base is Oxoid Nutrient Agar No. 2 with 10 per cent. defibrinated rabbit's blood. Five ml of the blood agar base is sloped in McCartney (28 ml) bottles. One ml of Hanks' balanced salt solution (BSS) is used as the liquid medium.

The promastigotes were harvested in November 1968, centrifuged at 2500 rpm for five minutes and resuspended in 0.5 ml of sterile liquid medium. The suspension was cooled to 4°C and 0.25 ml of sterile 15 per cent. dimethyl sulphoxide (DMSO) in Hanks' BSS at 4°C was added and mixed. This suspension was taken up in sterile capillary tubes 3 cm long. One cm of fluid was taken up. The end of the tube distal to the suspension was sealed very quickly in a gas/oxygen flame. The tubes were allowed to cool to 4°C and then placed, sealed end down, in sterile bijoux bottles with rubber cushions on the bottom. The tubes were centrifuged very briefly to throw the suspension to the bottom of the tube. The other end of the tube is then sealed as before. Ten such tubes were prepared for each strain. The tubes were marked with a very long broad black line for No. 50 onwards, a long broad black line representing 10 and a short broad black line representing 1. Thus No. 54 is  or No. 33  Each group of 10 tubes was placed in a plastic stoppered tube (36 mm long, 7 mm diameter), bored at each end to allow passage through of liquids and numbered as before. The plastic tubes were placed in the cooling apparatus of Goodwin & Thiel (1967)¹ and left for five hours. They were then transferred into liquid nitrogen.

The amastigotes were harvested in November 1968 by grinding the infected skin lesion or the infected spleen in cold five per cent. DMSO in Hanks' BSS. One ml of the suspension was placed into each of three ampoules (2 ml size) per strain. The ampoules were numbered and cooled as before and placed into liquid nitrogen.

2. STRAINS OF LEISHMANIA IN THE BANK

The following is the history of each strain and its position in the bank in November 1968.

¹ Goodwin, L. G., & Thiel, C. C. (1967) Trans. roy. Soc. trop. Med. Hyg., 61, 134

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PROMASTIGOTES

| No. | <u>Leishmania</u> species | History | Compartment | Cane No. | Position in cane |
|------|-------------------------------------|--|-------------|----------|------------------|
| L 1 | <u>L. braziliensis</u> | Isolated 11.11.60 from Cearo district, Brazil. Coehlo's strain No. 6. Kept in culture from 8.8.67 | 9A | 1 | 1 |
| L 2 | <u>L. donovani</u> | Isolated by Heish from <u>Phlebotomus martini</u> in Kenya. Kept in culture from 2.12.65 | 9A | 1 | 2 |
| L 3 | <u>L. donovani</u> | India. Called strain SC 23. Kept in culture from 5.4.63 | 9A | 1 | 3 |
| L 5 | <u>L. donovani</u> | Isolated from Nadir Moreira, ^o 9 years. Belo Horizonte, Minas Gerais, Brazil (Cafezinno). Coehlo's strain No. 8. Kept in culture from 17.4.67 | 9B | 12 | 1 |
| L 6 | <u>L. tropica</u> | Isolated from Rachel Palacci, Israel. Kept in culture from 18.4.68 | 9A | 1 | 4 |
| L 7 | <u>L. braziliensis</u> <u>sl</u> | Isolated from Dr J. J. Shaw, Panama. Shaw's strain SH 24. Kept in culture from 4.2.65 | 9A | 1 | 5 |
| L 9 | <u>L. braziliensis</u> | Isolated 1.2.61 from patient Manuel de Santos with leishmaniasis diffusa, Bahia, Brazil. Kept in culture from 14.5.63 | 9A | 1 | 6 |
| L 11 | <u>L. mexicana</u> | Isolated from <u>Nyctomys sumichrasti</u> via man. British Honduras. Lainson's strain, M379. Kept in culture from 17.4.68 | 9A | 3 | 1 |
| L 13 | <u>L. donovani</u> | Isolated 1961 from post-kala-azar dermal leishmaniasis. India. Kept in culture from 7.7.67 | 9A | 2 | 1 |
| L 15 | <u>L. braziliensis</u> | Isolated 16.8.63 from Raimundo Malo of Cametar, Para, Brazil. Case of leishmaniasis diffusa. Kept in culture from 8.8.67 | 9A | 2 | 2 |
| L 18 | <u>L. braziliensis</u> | Isolated 24.7.63 from case Ls 137/3N of Dr Herrer. Case of espundia, Peru. Kept in culture from 13.2.68 | 9B | 12 | 2 |
| L 19 | <u>L. donovani</u> | Isolated 10.8.63 from Jose Gomes da Silva of Sobral, Gara, Brazil. Kept in culture from 10.8.63 | 9A | 2 | 3 |
| L 21 | <u>L. braziliensis</u> | Isolated late 1962 from impetigo-like cutaneous leishmaniasis in Venezuela near Brazilian border. Patient Mario Perez. Kept in culture from 10th culture passage 16.9.63 | 9A | 2 | 4 |

| No. | <u>Leishmania</u> species | History | Compartment | Cane No. | Position in cane |
|------|-------------------------------------|---|-------------|----------|------------------|
| L 22 | <u>L. braziliensis</u> <u>sl</u> | Isolated 31.5.63 from child on Atlantic Coast of Costa Rica. Montero Gei's E10 strain. Kept in culture from 23.9.63 | 9A | 2 | 5 |
| L 23 | <u>L. braziliensis</u> <u>sl</u> | Isolated 1958 in San Jose, Costa Rica. Zeledon's O/CR strain. Kept in culture from 2.11.67 | 9B | 12 | 3 |
| L 26 | <u>L. tropica</u> | Isolated 4.3.65 by Dr Rahim from oriental sore, Baghdad, Iraq. Kept in culture from 4.3.65 | 9A | 2 | 6 |
| L 28 | <u>L. tropica</u> | Isolated 15.9.63 from Marmite Teleahou of Tikara village, Debra Sina, Shoa, Ethiopia. Patient with leishmaniasis diffusa. Fromentin's strain Ethiopian II. Kept in culture from 12.10.65. | 9A | 3 | 2 |
| L 29 | <u>L. tropica</u> | Isolated late 1965 from Wolde Sinbet of Ethiopia. Patient with leishmaniasis diffusa. Kept in culture from 24.11.65 | 9A | 3 | 3 |
| L 30 | <u>L. tropica</u> | Isolated late 1965 from Terunish of Ethiopia. Patient with leishmaniasis diffusa. Kept in culture from 22.8.66 | 9A | 3 | 4 |
| L 31 | <u>L. tropica</u> | Isolated late 1965 from Mussiy of Ethiopia. Strain from a tuberculoid lesion. Kept in culture from 24.11.65 | 9A | 3 | 5 |
| L 33 | <u>L. guyanensis</u> | Isolated 13.4.66 from 23629095 Private Coombes. Lesions acquired in Guyana December 1965. Kept in culture from 5.7.66 | 9A | 3 | 6 |
| L 34 | <u>L. guyanensis</u> | Isolated 27.4.66 from Private Howard, A. F. Lesions acquired in Guyana December 1965. Kept in culture from 5.7.66 | 9A | 9 | 1 |
| L 36 | <u>L. donovani</u> | From Kenya, kala-azar, isolated by Heisch. Kept in culture from 24.10.66 | 9A | 4 | 1 |
| L 37 | <u>L. donovani</u> | Isolated by Dr Heisch in Kenya. Strain S13. Kept in culture from 16.1.68 | 9A | 7 | 1 |
| L 39 | <u>L. braziliensis</u> <u>sl</u> | From man, Panama, Hertig's strain "Vega". Kept in culture from 16.1.68 | 9A | 4 | 2 |
| L 40 | <u>L. donovani</u> | From Sudan via WHO. Strain DH 177. Kept in culture from 24.10.66 | 9A | 4 | 3 |
| L 42 | <u>L. donovani</u> | From Sudan, isolated from <u>Genetta</u> . Kept in culture from 16.1.68 | 9B | 12 | 4 |
| L 43 | <u>L. donovani</u> | From Sudan, isolated from <u>Felis serval</u> . Kept in culture from 16.1.68 | 9A | 4 | 4 |
| L 46 | <u>L. donovani</u> | From Sudan, isolated from man. Strain No. 3. Kept in culture from 24.10.66 | 9A | 4 | 5 |

| No. | <u>Leishmania</u> species | History | Compartment | Cane No. | Position in cane |
|------|------------------------------|--|-------------|----------|------------------|
| L 47 | <u>L. donovani</u> | From Sudan, isolated from <u>Arvicanthus</u> strain VPRP 65. Kept in culture from 24.10.66 | 9A | 4 | 6 |
| L 48 | <u>L. donovani</u> | From Sudan, isolated from <u>Phlebotomus orientalis</u> . Strain DD 1021. Kept in culture from 16.1.68 | 9A | 5 | 1 |
| L 49 | <u>L. donovani</u> | From Sudan, isolated from <u>Phlebotomus orientalis</u> . Strain DD 1051. Kept in culture from 16.1.68 | 9A | 5 | 2 |
| L 50 | <u>L. donovani</u> | From Sudan, isolated from <u>Rattus</u> . Strain F25. Kept in culture from 24.10.66 | 9A | 5 | 3 |
| L 51 | <u>L. donovani</u> | From Sudan, isolated from <u>Acomys</u> . Strain 27A. Kept in culture from 30.1.68 | 9A | 5 | 4 |
| L 52 | <u>L. donovani</u> | From Sudan, isolated from <u>Acomys</u> . Strain 433. Kept in culture from 24.10.66 | 9A | 5 | 5 |
| L 53 | <u>L. infantum</u> | From Cyprus. Isolated by Dr Shaw 11.6.63 from Panagista Iginatiou. Kept in culture from 19.4.67 | 9B | 12 | 5 |
| L 54 | <u>L. infantum</u> | From Malta. Isolated from man. Strain 33. Kept in culture from 30.1.66 | 9A | 5 | 6 |
| L 55 | <u>L. infantum</u> | From South of France. Isolated by Dr Rioux from a dog. Strain SV. Kept in culture from 24.10.66 | 9A | 6 | 1 |
| L 56 | <u>L. infantum</u> | From South of France. Isolated by Dr Rioux from man. Strain SXI. Kept in culture from 24.10.66 | 9A | 6 | 2 |
| L 58 | <u>L. tropica</u> | From Iran. Isolated from leishmaniasis recidiva. Kept in culture from 24.10.66 | 9A | 6 | 3 |
| L 59 | <u>L. tropica</u> | From Algiers, isolated from man. Dr Rioux's strain SIX. Kept in culture from 30.1.68 | 9A | 6 | 4 |
| L 60 | <u>L. tropica</u> | From India, isolated from man by Brig. Sinton. Strain 23. Kept in culture from 24.10.66 | 9A | 6 | 5 |
| L 62 | <u>L. tropica</u> | From Iraq, isolated from dog in Baghdad. Strain Laurie II. Kept in culture from 24.10.66 | 9A | 6 | 6 |
| L 63 | <u>L. tropica</u> | From Iraq, isolated from case of leishmaniasis recidiva. Kept in culture from 24.10.66 | 9A | 7 | 2 |
| L 64 | <u>L. tropica</u> | From Iraq, isolated from case of leishmaniasis recidiva. Strain SAV. Kept in culture from 24.10.66 | 9A | 7 | 3 |

| No. | <u>Leishmania</u> species | History | Compartment | Cane No. | Position in cane |
|------|------------------------------|--|-------------|----------|------------------|
| L 65 | <u>L. tropica</u> | From Israel, isolated from man. Strain Ein Geddi II. Kept in culture from 24.10.66 | 9A | 7 | 4 |
| L 66 | <u>L. tropica</u> | From Israel, strain Yatvata. Kept in culture from 30.1.68 | 9A | 9 | 2 |
| L 67 | <u>L. tropica</u> | From Israel, strain 8 Ber. Kept in culture from 24.10.66 | 9A | 9 | 3 |
| L 69 | <u>L. tropica minor</u> | From Turkestan, USSR. Kept in culture from 24.10.66 | 9A | 7 | 5 |
| L 70 | <u>Leishmania sp.</u> | Isolated by Dr Heisch from <u>Tatera</u> in Kenya. Strain T1. Kept in culture from 30.1.68 | 9A | 9 | 4 |
| L 71 | <u>Leishmania sp</u> | Isolated by Dr Heisch from <u>Tatera</u> in Kenya. Strain T3. Kept in culture from 24.10.66 | 9A | 7 | 6 |
| L 75 | <u>L. tropica</u> | Isolated November 1965 by Dr Rahim from man in Baghdad, Iraq. Strain IR3. Kept in culture from 20.12.66 (9 tubes only) | 9A | 8 | 1 |
| L 76 | <u>L. tropica</u> | Isolated December 1965 by Dr Rahim from man in Baghdad, Iraq. Strain IR5. Kept in culture from 20.12.66 (8 tubes only) | 9A | 8 | 2 |
| L 78 | <u>L. infantum</u> | Isolated 17.1.67 by Dr Walters from man in South of France. Kept in culture from 17.1.67 | 9A | 9 | 5 |
| L 81 | <u>L. tropica</u> | Isolated January 1967 by Dr Rahim from a case of leishmaniasis recidiva in Baghdad, Iraq. Kept in culture from 31.3.67 | 9A | 9 | 6 |
| L 82 | <u>L. donovani</u> | Isolated in 1967 from kala-azar case in Humera district, Ethiopia. Kept in culture from 7.6.67. Strain HV3 | 9B | 10 | 1 |
| L 83 | <u>Leishmania sp</u> | Isolated from lizard, USSR | 9B | 10 | 2 |
| L 85 | <u>L. tropica</u> | Isolated 18.9.67 by Dr Bryceson from leishmaniasis diffusa patient Tsegai Wolde Gabriel of Ethiopia. Kept in culture from 14.10.67 | 9B | 10 | 3 |
| L 86 | <u>L. tropica</u> | Isolated 5.8.67 by Dr Bryceson from oriental sore patient Kedir Abdul of Ethiopia. Kept in culture from 28.9.67 | 9B | 10 | 4 |
| L 88 | <u>L. enriettii</u> | Kept in culture from September 1968 | 9B | 10 | 5 |
| L 92 | <u>L. donovani</u> | | 9B | 10 | 6 |

Time kept in culture means the time the promastigotes have been in culture prior to freezing down while in the reference centre. Position in the cane refers to the positions from bottom to top.

To remove promastigotes:

- (1) Take cane out, remove plastic container, take off cap, remove one tube, replace plastic container and cane.
- (2) Put tube in water at room temperature.
- (3) Dry tube and nick with diamond pencil in the middle of liquid column.
- (4) Place tube in ether for one minute.
- (5) Remove tube to sterile filter paper in sterile petri dish with sterile forceps.
- (6) Remove when dry from ether with sterile forceps and put in NNN medium.
- (7) With sterile forceps or rod break the tube.

AMASTIGOTES
(in 2 ml ampoules)

| No. | <u>Leishmania</u> species | Maintained <u>in vivo</u> in: | Compartment | Cane No. | Position in cane |
|------|---|----------------------------------|-------------|-------------|---------------------|
| L 1 | <u>L. braziliensis</u> | Hamster nose | 9B | 11 | 1, 2, 3 |
| L 2 | <u>L. donovani</u> | Hamster spleen | 9B | 11 | 4, 5 |
| L 7 | <u>L. braziliensis</u> | Hamster nose | 9B | 12 13 | 5 1, 2 |
| L 11 | <u>L. mexicana</u> | Mouse skin | 9B | 13 | 3, 4, 5 |
| L 15 | <u>L. braziliensis</u> | Hamster nose | 9B | 14 | 1, 2, 3 |
| L 18 | <u>L. braziliensis</u> | Hamster nose | 9B | 15 | 1, 2, 3 |
| L 25 | <u>L. tropica major</u> (from Turkestan USSR) | Mouse skin | 9B | 15 16 | 4, 5 1 |
| L 31 | <u>L. tropica</u> | Hamster nose | 9B | 16 | 2, 3, 4 |
| L 33 | <u>L. guyanensis</u> | Hamster nose | 9B | 16 17 | 5 1, 2 |
| L 82 | <u>L. donovani</u> | Hamster spleen | 9B | 12 | 2, 3, 4 |
| L 88 | <u>L. enriettii</u> | Guinea-pig nose | 9B | 17 | 3, 4, 5 |

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