

THE RETAINED CAPACITY OF *LUTZOMYIA LONGIPALPIS* (LUTZ & NEIVA) TO TRANSMIT *LEISHMANIA CHAGASI* (CUNHA & CHAGAS) AFTER EIGHT YEARS (64 GENERATIONS) IN A CLOSED LABORATORY COLONY

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A closed Lutzomyia longipalpis colony, from Ceará has been used to transmit Leishmania chagasi isolated from a fox in Pará state. The last time this colony was successfully used in similar transmission experiments was eight years (64 generations) ago indicating that this colony of Lu. longipalpis has fully maintained its vectorial capacity in spite of such a long period of maintenance in the laboratory.

There are at least two morphological forms of *Lutzomyia longipalpis*: one in which the 4th abdominal tergite bears a pair of dorsolateral, pale spots, and another in which a second pair of spots is found on the 3rd segment (Mangabeira, 1969; Ward et al., 1983). The latter authors showed that these two forms were reproductively isolated and suggested that differences in their ecology and biting habits might influence the incidence of human visceral leishmaniasis in different geographical regions.

Lainson, Ward & Shaw (1977) reported the first experimental transmission of *Leishmania chagasi* by the bite of laboratory-bred *Lu. longipalpis* and later (Lainson et al., 1984; 1985) again transmitted the parasite by the bites of naturally infected *Lu. longipalpis*.

In the first transmissions these authors used a combination of the "2-spot" form of *Lu. longipalpis* from Morada Nova, Ceará and a strain of *Le. chagasi* isolated from a child in Bahia state. In the second experiments, they used naturally infected, "1-spot" *Lu. longipalpis*, captured in a focus of visceral leishmaniasis in Santarém, Pará state. They also presented biological and biochemical evidence showing that numerous isolates of *Le. chagasi* from man, *Lu. longipalpis*, foxes and dogs in Pará, and man in Bahia and Ceará were indistinguishable (Braga et al., 1985).

We report here experiments designed to see if our colony of *Lu. longipalpis* ("2-spot") still retained its vectorial capacity, and also if it would be possible to transmit *Le. chagasi* from a "1-spot" area.

MATERIAL AND METHODS

Strain MCER/BR/81/M6445 of *Le. chagasi* was isolated, in a hamster, from a fox (*Cerdocyon thous*) captured near Salvaterra, island of Marajó, Pará state in 1981 (Silveira et al., 1982); it had undergone seven passages in hamsters. The spleen was removed from a hamster five months post inoculation and triturated in 4 ml of sterile, defibrinated rabbit blood. On 31st August, 1984 approximately 200 female *Lu. longipalpis* from our ("2-spot") colony were fed on this suspension, through a chick membrane, maintaining this at 37°C with a Haake Model FE constant temperature circulator. They were offered a saturated sucrose solution throughout the experiment except for 28 hours prior to the feeds and maintained in net cages (Ward, 1977). The hamsters were examined for parasites three months later.

RESULTS

Totals of 28, 22 and 13 sandflies were successfully refeed on three hamsters on the 5th, 6th and 7th days post-infected feed respectively, and dissection of these confirmed a 100% infection rate, with massive infections of the proventriculus and midgut. Two hamsters showed amastigotes in smears of liver and spleen when the animals were killed three months later; unfortunately the 3rd animal escaped and was lost.

This work was supported financially by the Wellcome Trust (London) and the Fundação SESP (Rio de Janeiro). The Wellcome Parasitology Unit, Instituto Evandro Chagas, Fundação SESP, Caixa Postal 3, 66000 Belém, PA, Brazil.

Received for publication February 1st and accepted March 6th, 1985.

DISCUSSION

Our colony of *Lu. longipalpis* from Morada Nova, Ceará was in its 23rd generation at the time of the first experimental transmission by Lainson, Ward & Shaw (1977) and in its 87th generation at the time of the present experiment. Under our laboratory conditions the maintenance of *Lu. longipalpis* in a reproductively closed colony for eight years has not effected its ability to transmit *Le. chagasi*.

In addition, the results presented here indicate the relative ease of transmitting suprapylarian leishmaniasis compared to peripylarians (Ryan, Lainson & Shaw, 1985). The transmission of *Le. chagasi* from a "1-spot" *Lu. longipalpis* area, by the "2-spot" form of *Lu. longipalpis* adds weight to the argument that we are dealing with the same parasite in endemic areas throughout Brazil (Braga et al., 1985).

RESUMO

Lutzomyia longipalpis foi alimentado através de membrana com uma suspensão de macedado de fígado e baço em sangue desfibrinado de coelho. Este material foi originário de um hamster infectado com *Leishmania chagasi*, realimentado em hamsters limpos, transmitindo os parasitos em duas ocasiões. Esta mesma colônia de *Lu. longipalpis*, do Ceará, foi usada para a primeira transmissão há oito anos e 64 gerações atrás e não teve a capacidade vetorial diminuída.

ACKNOWLEDGEMENTS

The authors are grateful to José Itamar de Almeida, Iorlando da Rocha Barata, Augusto Francisco Nascimento Filho, Antonio Julio de Oliveira Monteiro and Suêd de Nazare Freitas Silvas for invaluable technical assistance.

REFERENCES

- BRAGA, R.R.; LAINSON, R.; SHAW, J.J.; RYAN, L. & SILVEIRA, F.T., 1985. Leishmaniasis in Brazil. XXII. Characterization of *Leishmania* from man, dogs and the sandfly *Lutzomyia longipalpis* (Lutz & Neiva) isolated during an outbreak of visceral leishmaniasis in Santarém, Pará state. *Trans. Roy. Soc. Trop. Med. & Hyg.* (in press).
- LAINSON, R.; SHAW, J.J.; RYAN, L.; RIBEIRO, R.S.M. & SILVEIRA, F.T., 1984. Presente situação da leishmaniose visceral na Amazônia, com especial referência a um novo surto da doença, ocorrido em Santarém, Estado do Pará, Brasil. *Bol. Epid. FSESP*, núm. esp. Julho, 1984, 8 pp.
- LAINSON, R.; SHAW, J.J.; RYAN, L.; RIBEIRO, R.S.M. & SILVEIRA, F.T., 1985. Leishmaniasis in Brazil. XXI. Visceral Leishmaniasis in the Amazon region and further observations on the rôle of *Lutzomyia longipalpis* (Lutz & Neiva, 1912) as the vector. *Trans. Roy. Soc. Trop. Med. Hyg.* (in press).
- LAINSON, R.; WARD, R.D. & SHAW, J.J., 1977. Experimental transmission of *Leishmania chagasi*, causative agent of neotropical visceral leishmaniasis, by the sandfly *Lutzomyia longipalpis*. *Nature*, 266 :628-629.
- MANGABEIRA, O., 1969. Sobre a sistemática e biologia dos *Phlebotomus* do Ceará. *Rev. Bras. Malar. Doenç. Trop.*, 21 :3-26.
- RYAN, L.; LAINSON, R. & SHAW, J.J., 1985. The experimental transmission of *Leishmania mexicana amazonensis* Lainson & Shaw, between hamsters by the bite of *Lutzomyia furcata* (Mangabeira). *Trans. Roy. Soc. Trop. Med. Hyg.* (in press).
- SILVEIRA, F.T.; LAINSON, R.; SHAW, J.J. & PAVOA, M.M., 1982. Leishmaniasis in Brazil: XVIII. Further evidence incriminating the fox *Cerdocyon thous* (L) as a reservoir of Amazonian visceral leishmaniasis. *Trans. Roy. Soc. Trop. Med. Hyg.*, 76 :830-832.
- WARD, R.D., 1977. The colonization of *Lutzomyia flaviscutellata* (Diptera: Psychodidae), a vector of *Leishmania mexicana amazonensis* in Brazil. *J. Med. Ent.*, 14 :469-476.
- WARD, R.D.; RIBEIRO, A.L.; READY, P.D. & MURTAGH, A., 1983. Reproductive isolation between different forms of *Lutzomyia longipalpis* (Lutz & Neiva), (Diptera: Psychodidae), the vector of *Leishmania donovani chagasi* Cunha & Chagas and its significance to Kala-azar distribution in South America. *Mem. Inst. Oswaldo Cruz*, 78 :269-280.