# NOTES ET INFORMATIONS

# THE ARTIFICIAL FEEDING OF LABORATORY REARED PALEARCTIC SANDFLIES (DIPTERA : PSYCHODIDAE) FOR STUDIES ON THE TRANSMISSION OF DISEASE AGENTS

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SUMMARY. Female sandflies of three *Phlebotomus* species were fed on membrane. The feeding apparatus is described and preliminary data obtained are also reported. A total 2 835 sandflies were fed : 2 596 *P. perniciosus*, 104 *P. perfiliewi* and 135 *P. papatasi*.

#### La nourriture artificielle des Phlébotomes (Diptera : Psychodidae) élevées dans le laboratoire pour les études de la transmission d'agents pathogènes

RÉSUMÉ. Les femelles de phlébotomes de trois espèces du genre *Phlebotomus* ont été nourries sur membrane artificielle pour étudier la transmission d'agents pathogènes.

L'appareillage est décrit et les résultats préliminaires sont rapportés. Un total de 2 835 phlébotomes ont été nourris : 2 596 P. perniciosus, 104 P. perfiliewi et 135 P. papatasi.

## Introduction

Several artificial feeding techniques have been used in studies on the capacity of haematophagus arthropods as vectors of infectious disease agents. Regarding phlebotomine sandflies (Diptera, Psychodidae) Adler and Theodor (1941) carried out for the first time experimental transmission of cutaneous leishmaniasis to man, using *Phlebotomus papatasi* infected with *Leishmania tropica* through a membrane. Later on, many authors have used artificial feeding techniques to feed or to infect sandflies (Adler, 1947; Heyneman, 1963; Schmidt, 1964; Gemetchu, 1976; Ready, 1978; Ward *et al.*, 1978).

This paper describes an apparatus which has been designed for artificial infection of sandflies; the apparatus can be introduced directly into the rearing cage of sandflies. Preliminary data on feeding tests with the apparatus are also reported.

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### Materials and methods

Feeding tube. The feeding tube apparatus (fig. 1) is made of heated-resistant glass to permit autoclaving. The feeder tube is  $32 \times 3.5$  cm, the bottom of which is drawn inward to form a cone-shaped cavity (fig. 1, A) and contains about 3 ml of liquid. The bottom of the tube has a circular groove (fig. 1, B) over which the membrane can be easily secured using a metal thread to close the base of the cone. The cavity is surrounded by a cylindrical water-jacket provided with a lower inlet (fig. 1, C) and an outlet (fig. 1, D) for connecting rubber tubing. The inlet and outlet tubes are connected to a constant temperature circulator. The feeder is supported inside the cage (fig. 2). The area of the feeding surface is  $3.5 \text{ cm}^3$ . High humidity is maintained, during the feeding test, by sealing the cage containing sandflies in a plastic bag with a swab of wet cotton wool. The feeding trials are carried out in the dark at the room temperature of  $20-22^{\circ}$  C.







FIG. 2. — Feeding apparatus with feeder connected to the termostat.

*Membrane*. The membrane used is the skin of five day old chicken freeshly prepared or kept frozen. Before use, the membrane is washed with physiological sterile saline to prevent contamination of food mixtures.

*Blood.* Defibrinated rabbit blood is used either fresh or refrigerated and mixed with promastigotes obtained from blood-agar media, or with leishmanial amastigotes from infected hamsters or with some Phleboviruses from 10 % suspension of infected suckling mouse brain. The blood and the membrane are heated at 34-35° C by the above mentioned thermostatic apparatus.

Sandflies. The sandflies used in the trials were from a laboratory colonies established in our department (Maroli, 1983; Maroli *et al.*, 1983 in press). The species were : *P. perniciosus*, now in tis 21th closed generation, *P. perfiliewi*, 13th closed generation and *P. papatasi*, 20th closed generation. The *P. papatasi* colony was started with larve provide by Dr. R. Lane, British Museum, Natural History, London, England. The original line of this colony was descended from *P. papatasi* samples collected in India.

### **Results and discussion**

Using the feeding apparatus previously described a total 2 835 sandflies were fed. The results include 16 trials with promastigotes or amastigotes of different *Leishmania* strains (1 046 *P. perniciosus* and 68 *P. perfiliewi* were fed) and 22 trials with different Phleboviruses (1 550 *P. perniciosus*, 135 *P. papatasi* and 36 *P. perfiliewi* were fed). The feeding period ranged between 1.30 hour and 3.00 hours, but it has been observed that a high percentage (70-80 %) of sandflies took their blood meal during the first hour of the experiments.

Regarding the species so fed, since it is the first time that *P. perfiliewi* has been recorded as laboratory reared in a closed colony, the total of 104 females engorged on membrane suggests that this species will readly feed on artificial apparatus if a suitable host is not available. *P. perniciosus* and *P. papatasi* were previously fed on a membrane by other authors.

Using this technique we obtained a high infection rates, as will be described in a subsequent paper in studies on the laboratory transmission of *Leishmania infantum* to *Rattus rattus* through the bite of *P. perniciosus* experimentally infected on membrane (Pozio *et al.*, 1985 in press). Moreover this technique was also used in laboratory studies on the behaviour of some phleboviruses. Susceptibility testing of *P. perniciosus* to laboratory infection with Toscana and Arbia viruses were also carried out (Ciufolini *et al.*, 1985).

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