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RESEARCH NOTE

A PRACTICAL METHOD FOR MASS BREEDING OF SANDFLIES IN THE LABORATORY: *LUTZOMYIA INTERMEDIA* (LUTZ & NEIVA, 1912) AND *LUTZOMYIA LONGIPALPIS* (LUTZ & NEIVA, 1912) (DIPTERA, PSYCHODIDAE)

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Laboratory colonies of sandflies may be used in various types of research in leishmaniasis, such as xenodiagnosis in infected humans or animals, experimental infections, the systematics of the parasites, the biology and genetics of the vectors. Such experiments may require a large number of sandflies and, therefore, the improvement of rearing techniques aiming at mass production with a reduced amount of labour is, obviously, important.

With this in mind we introduced the alterations mentioned below in the routine of our laboratory rearing of two species, *Lutzomyia intermedia* and *Lutzomyia longipalpis*.

Some of the main attempts of mass rearing of these sandflies were performed by Barretto (1942, Thesis, São Paulo University, 162 p.) with *L. intermedia,* and Sherlock & Sherlock (1959, *Rev. Bras. Biol., 19* :229-250) and Modi & Tesh (1983, *J. Med. Entomol., 20* :568-569) with *L. longipalpis,* all with some success.

In our case, to start the mass rearing we used sandflies from the colonies established in the Department of Entomology, Instituto Oswaldo Cruz (Rangel et al., 1985, *Mem. Inst. Oswaldo Cruz, 80* :219-226) but did some modifications concerning the maintenance of the immature stages – eggs and larvae – and fungi control.

Immature stages – In the original colony (Rangel et al. op.cit.) the eggs were placed in Petri dishes lined with plaster of Paris and damped with distilled water. 300 eggs were used per dish. After the hatching of 1st instar larvae, the dishes were observed daily, even during week-ends, for larval feeding, humidity and fungi control. Four technicians worked in the maintenance of the two colonies, each spending 45 hours per week.

In the new method the eggs are placed in plastic pots with cover (6,5 cm high X 16 cm diameter), both lined inside with plaster of Paris. The pots are placed into covered plastic boxes, lined in the botton with filter paper moistened with distilled water. The number of eggs in each pot should not exceed 2.500 per unit. The food for the larvae – Vitormonio (used for fish) – is given on the 5 th day after the hatching of eggs, and subsequently only twice a week for *L. longipalpis* and three times for *L. intermedia*, when we have the opportunity to observe the behaviour of larvae and the humidity level. We recommend to put a little food in the internal side of the cover pot, to be eaten by the larvae when dispersing inside the pot. When the pupae emerge we replace the cover pots with another nylon cover, fixed with adhesive tape, with a hole in the centre to allow the transference of the sandflies to the cage, using a Castro collecting tube.

With this method we verified that the output per pot is the same for both species, with only one well trained technician spending 25 hours per week. We obtain, approximately, 6.000 sandflies per generation. The life cycle of both species was the same as described by us (Rangel, 1985, Thesis, Rio de Janeiro University, IX + 154 p.) when the more complex method was used.

These alterations in the method of mass rearing, being practical and inexpensive, could be applied to several species of sandflies, including those with a more difficult adaptation in the laboratory.

Anti-fungi test – In some batches of blood-fed females we tested an anti-fungi product – Tropfish (used for fish) – diluted in the proportion of two drops per litre of distilled water. With this solution we moisten the plastic vials which harbour the females and observed them three times a week.

The growth of fungi is one of the worst obstacles for breeding sandflies. The presence of fungi on the eggs may be harmful because these are prevented from hatching. Using the anti-fungus solution we have a good performance for both species, without contamination, with a high percentage of ovipositions (70%)

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and a high number of eggs, without damage to the life cycle.

Another point to be considered is the growth of fungi on the larval food. According to Barretto (op.cit.), Sherlock & Sherlock (op.cit.) and Hertig & Johnson (1961, *Ann. Ent. Am., 54* :753-766) the larvae can get involved in the mycelia and spores, that would cause their death. We observed a good acceptance of fungi as food by the larvae, including the younger stages. We agree that some species of fungi may be dangerous for sandflies, but it is possible that others might be a good complement for their nutrition.

Eldridge at al. (1963, *Mosquito News, 23* :215-217) and Hertig (1964, *Bull. Wld. Hlth. Org., 31* :569-570) think that the sterilization of the material used in breeding could reduce the fungi growth. However, in our modification we did not need to use sterilized material, thus reducing the daily labour without damage for the colonies.

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