AN EFFICIENT METHOD FOR TRANSFERRING ADULT MOSQUITOES DURING FIELD TESTS

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To improve efficiency of field tests with sentinel mosquitoes, a method was developed for transferring specimens with a mechanical aspirator (Hausherr's Machine Works, Old Freehold Road—RD 1, Toms River, NJ 08753) similar to devices described by Carver (1967), Trpis (1968), and Jackson and Grothaus (1971). The objective was to transfer treated specimens from the standard mosquito exposure tubes contained in the World Health Organization (WHO) tests kits (Brown and Pal 1971) to collecting vials for observation. Since WHO exposure tubes are not designed for connection with a mechanical aspirator, an adapter was fabricated and tested (Fig. 1).

The adapter was fabricated by cementing a polypropylene funnel (65 mm top diam, 15 mm stem outer diam, and 18 mm stem length) to one of the threaded rings of the WHO exposure tubes. The funnel stem fits firmly into the 14 mm opening in the stopper-cap of the collecting vial (screen bottom, 27 mm outer diam) that accompanied the aspirator. Because the transfer method requires one aspirator and two identical adapters, an additional stopper-cap must be obtained from the manufacturer to fabricate a second adapter.

The screens in the WHO test kits that are inserted between the threaded rings and exposure tubes to prevent mosquitoes from escaping during tests were replaced with nylon netting (mesh repeats per inch: 20 × 28). The system worked best when a piece of netting was secured with a ring on one end of the tube and another piece of netting secured by a rubber band on the opposite end of the tube. The system depicted in Fig. 2 was used as follows: The end of the exposure tube with the netting secured by a rubber band was screwed into adapter 1 (Fig. 2A). The velocity of the air drawn through these connected pieces intensifies nearer the aspirator. Those mosquitoes near the distal end of the exposure tube were lightly blown closer to the aspirator where the air velocity was sufficient to retain them. The exposure tube ring and netting were removed from the opposite end of the exposure tube and replaced with adapter 2. The entire assembly was removed from the aspirator, rotated 180° and replaced into the aspirator (Fig. 2B). (Because the netting was removed before adapter 2 was connected to the exposure tube, the mosquitoes could be drawn freely from the tube into the collecting vial of the adapter.) Adapter 1 was removed and air blown lightly into the exposure tube to assist the aspirator in drawing the mosquitoes into the collecting vial of adapter number two (Fig. 2C). Once the mosquitoes were in the vial, the stopper-cap, funnel, and exposure tube were removed from the collecting vial (Fig. 2D). The air velocity was sufficient to hold mosquitoes in the collecting vial during capping. The aspirator switch was turned off between steps and after capping, prior to removal of the collecting vial from the aspirator.

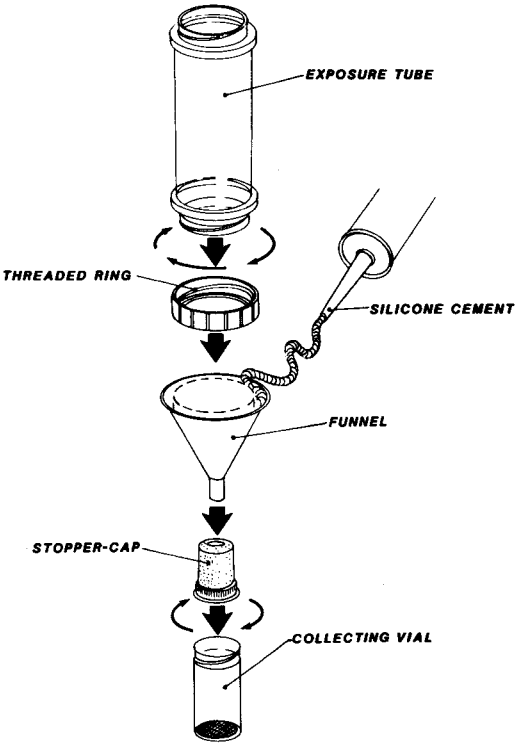

Fig. 1. Adapter components.

This method improves efficiency by permitting rapid transfer with a low degree of mortality. During field studies where 131 samples were transferred from treated exposure tubes

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[2] The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense.

to clean collecting vials, the average transfer time was 30 seconds per sample.

The degree of mortality due to stress during transfer in *Aedes aegypti* (Linnaeus) and *Aedes taeniorhynchus* (Wiedemann) was determined by comparing mortality of specimens transferred 0, 1 and 2 times. Adult mosquitoes were chilled and 20 females placed into WHO exposure tubes then transferred to collecting vials (transferred 1 time). Control mosquitoes were placed directly into collecting vials (transferred 0 times). Specimens transferred twice were rechilled in the collecting vials, returned to the exposure tubes and transferred a second time. Controls were rechilled a second time and kept in the collecting vial. The mosquitoes were provided sugar water while in the collecting vials and percent mortality recorded 12 hours

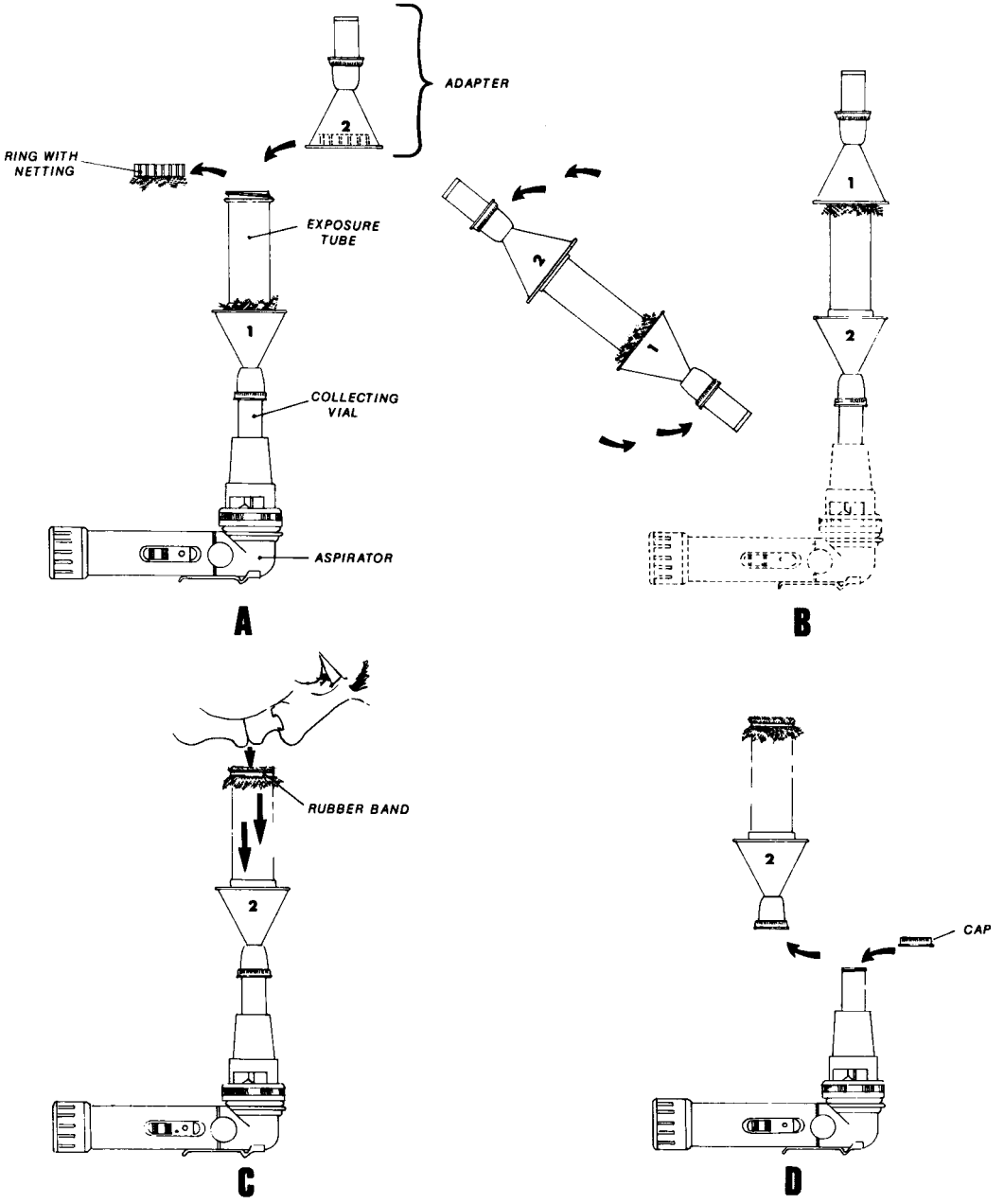

Fig. 2. Method for transferring adult mosquitoes from WHO test kit exposure tubes to collecting vials.

after transfer. Three replicates of each treatment with each species was conducted.

An analysis of data, Chi Square and Fisher's Exact Tests, failed to detect any statistically significant differences ($P \leq 0.05$) in the average percent mortality of *Ae. aegypti* versus *Ae. taeniorhynchus*. The number of times mosquitoes were transferred did not significantly affect mortality of either species. *Aedes aegypti* showed 6.7% mortality on one transfer and 8.3% mortality on two transfers. *Aedes taeniorhynchus* showed no mortality on one transfer and only 1.7% mortality on two transfers.

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References Cited

Brown, A. W. A. and R. Pal. 1971. Insecticide resistance in arthropods. W. H. O. Monogr. Ser. 38, p. 55.

Carver, H. D. 1967. A portable aspirator for collecting mosquitoes. Mosq. News 27:428–429.

Jackson, S. C. and R. H. Grothaus. 1971. A combination aspirator and killing tube for collecting mosquitoes and other insects. Mosq. News 31:112–113.

Trpis, M. 1968. A suction apparatus for collecting mosquitoes and other insects. Mosq. News 28:647–648.

THE EFFECT OF DIFFERENT METHODS OF OVIPOSITION INDUCEMENT ON EGG FERTILITY RATES IN A *SIMULIUM DAMNOSUM* THEOBALD COMPLEX SPECIES (DIPTERA: SIMULIIDAE)

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Most gravid females of the species of African Simuliidae so far tested will oviposit in captivity, but the conditions required vary with the species (Raybould and Grunewald 1975). Lewis et al. (1961) found that gravid *Simulium damnosum* s.l. can be induced to lay eggs by immersion in water or decapitation. These methods have since been used successfully with all members of the *S. damnosum* complex investigated, although for convenience some workers have crushed the head rather than removing it entirely. Although most females can be induced to oviposit in the laboratory with these techniques, many eggs laid by inseminated females fail to develop (Wenk and Raybould 1972).

New, effective techniques for inducing oviposition using artificial twilight conditions have recently been developed (Cupp et al. 1981, Simmons and Edman 1981, 1982). These methods simulate natural conditions more closely than the earlier techniques. Simmons and Edman (1982) reported that most eggs laid by *S. damnosum* s.l females stimulated by their technique were fertile. They speculated that the more natural oviposition stimuli may have produced higher egg fertilization rates.

Since it is important to obtain maximum egg fertility rates for laboratory colonization, investigations were carried out on the *S. damnosum* complex species *Simulium squamosum* (Enderlein) to compare the proportion of fertile eggs laid following two different methods of oviposition inducement: the Simmons and Edman (1982) technique and the immersion method. The latter was chosen for comparison rather than decapitation because it does not damage the fly.

The *S. damnosum* complex females used were caught at Tsatsadu Falls, near Hohoe in the Volta Region of Ghana, when the biting population present was probably entirely *S. squamosum*.

Biting females were allowed to feed to repletion on man in the field and tubed separately in 4.5 × 1.5 cm plastic (not glass) tubes with push-in caps. They were kept cool before and during transportation. In the laboratory each fly was maintained in a separate tube, fed on sugar solution and protected from attack by ants (Raybould et al. 1982). Three or occasionally four days after feeding, each fly was induced to oviposit by one of the following methods:

THE IMMERSION METHOD. To determine whether the time of oviposition inducement affected egg fertility rates, about half the flies were stimulated during the day and the other half in the evening at about 1930 hr under artificial lighting. Each female was placed in a separate 4.5 × 1.5 cm plastic tube lined with a rolled up piece of fine silk-netting and filled with water to a depth of about 1 cm. The tube was tilted and rotated in such a way as to immerse the fly and induce it to oviposit on the netting, but not on the inside of the lid or the bottom of the tube. The netting with the eggs attached was placed in a container of well aerated water until the next day when the number

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