ARTIFICIAL FEEDING APPARATUS FOR MOSQUITOES[1]

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Feeding of mosquitoes artificially on blood or other liquid substances through a synthetic or an animal membrane has been described by many investigators, including Whitman (1948), Greenberg (1949), Eyles (1951), and Rutledge *et al.* (1964). The principle of these techniques is based largely upon keeping the heparinized or defibrinated blood warm in a glass cylinder covered at the bottom with a membrane thin enough so that the mosquitoes can probe through it. Difficulties may be experienced in preparing or purchasing a satisfactory membrane, and in preventing sedimentation of the red blood cells. In this paper a simple technique is described for the preparation of a very satisfactory animal membrane and also a stirring device to be used with the feeding apparatus.

PREPARATION OF THE MEMBRANE. Of the many animal and synthetic membranes tested, including mouse skin, chicken skin, parafilm, dialyzing bag, and very thin rubber, the most satisfactory was one prepared from the diverticulum or crop of the chicken. This membrane is the elastic fibrous tissue separating the inner epithelial layer of the crop from its two outer muscular layers. The preparation requires only a gentle scraping of the inner epithelium and outer muscular tissue. The crop is sliced in two halves and some of the musculature is peeled off. One portion of the crop is then spread on the base of the thumb and over the fleshy part of the palm and scraped gently with the aid of a single-edged blade (Weck). When all the muscles are removed, the membrane is turned over and scraped on the inner side. At this time it becomes so transparent that any tissue still attached to it can be easily seen. Further cleaning of the small tissue fragments is done by a piece of "Kimwipes" tissue paper.

It is important to prevent the membrane from becoming dry during this process; this may be avoided by sprinkling a few drops of water on it from time to time. It is then spread over the mouth of a glass cylinder and secured with a rubber band. Each glass cylinder is made from a regular test tube with an open lip about 25 mm. in diameter. These test tubes are cut at the bottom to form a cylinder 17 cm. in length. The tubes bearing the membranes are placed in a large beaker and stored in a refrigerator. The beaker contains a wet cotton pad at the bottom to keep the membrane protected and moist. The tubes are checked for holes or leakage by filling them with distilled water or saline before they are used. A well-prepared membrane from a 1- to 2-month old chicken is permeable enough to keep its outside surface soft and moist when blood or other liquids are introduced into the tubes. Too much scraping is needed for the preparation of a membrane from a very old chicken, and on the other hand, a membrane from the crop of a very young chick is too thin and permeable to be of any practical use.

THE FEEDING APPARATUS. The feeding device of Greenberg (1949) was modified as described below (Fig. 1). It consists of two square-shaped, thin, pressed boards (30 x 30 cm.), one standing 30 cm. above the other, supported by four metal rods. There is a hole, 2 cm. in diameter, at each corner of the top board. Underneath each hole a heating unit, 6 cm. in length and diameter, is attached. Each unit is an asbestos-lined cylinder con-

[1] This study was supported in part by a research grant from the U. S. Public Health Service (AI-00351), National Institutes of Health, Bethesda, Maryland.

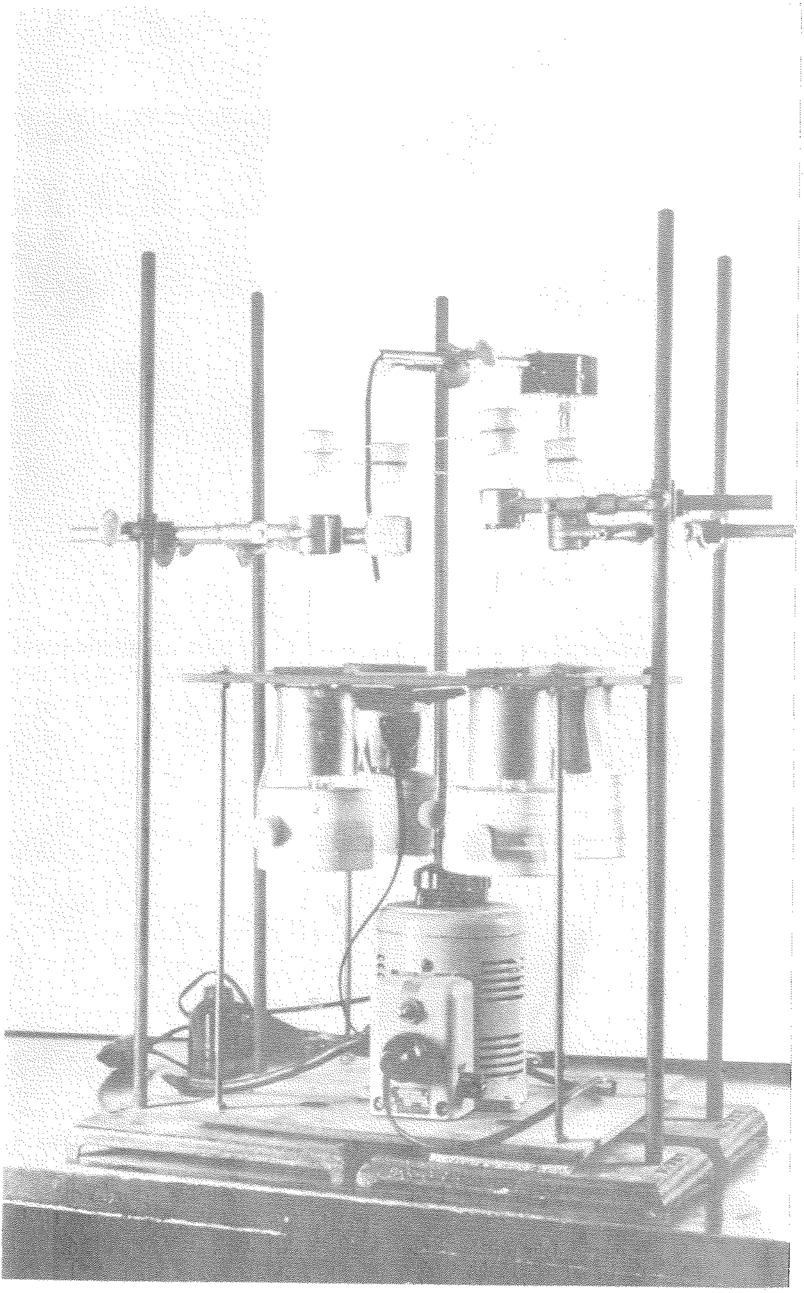

FIG. 1.—Artificial feeding apparatus with mosquito boxes attached.

taining a heating coil around a central hole, 2 cm. in diameter.

A rheostat connected to these four units controlled the desired temperature. The improvement in this apparatus was to add a glass stirring rod to each tube containing the suspension to be fed to the mosquitoes. The stirrers were driven by a small electric motor, regulated by a speed control unit. Each stirrer was made of a glass rod, 5 mm. in diameter and 250 mm. in length, and a glass tube 7 mm. in diameter and 120 mm. in length. The glass tube is inserted firmly up to its middle into a number 6 rubber stopper (Fig. 2, stopper #2), which provides a place for clamping the stirrer to a stand. One end of the glass rod is also forced into a narrow hole which is made in the center of another number 6 rubber stopper (stopper #1). The glass rod is then passed through the glass tube and the free end of it heated until it becomes red and soft. With a pair of pliers, pressure is applied immediately to the soft glass until a small disk about 8 mm. in diameter is formed. The glass rod should easily rotate inside the glass tube.

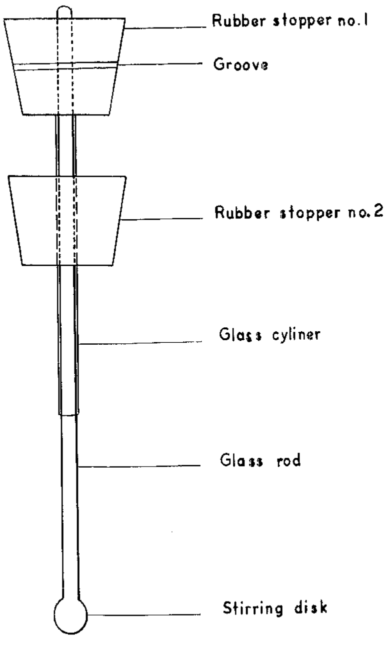

FIG. 2.—Stirrer of the artificial feeding device.

Two narrow strips of adhesive tape are wrapped around the uppermost rubber stopper. These strips are about 3 mm. apart, leaving a groove between them. The stirrers are held in the center of the heating cylinders by four clamps and four stands. A belt made of a narrow cord is placed in the grooves of the stoppers and is connected to a light duty stirring motor with a speed control unit (Eastern model #1, Arthur H. Thomas Co.). The original stirring rod of the motor is replaced by a regular syringe adaptor, which serves as a pulley, driving the belt and turning the stirrers simultaneously. The position and height of the stirrers in the heating units can be adjusted by raising or lowering the positions of the clamps on the stands. The tension of the belt is regulated by changing the position of the stand which is holding the motor.

At the time of feeding, the stirrers are lowered into the heating units, and extend a few millimeters beyond the bottom openings of the cylinders. The tubes bearing the membranes are then inserted from below into the cylinders until the membranes stand 2–3 mm. from the flattened ends of the stirrers. Each tube is held in this position by being passed through a tight-fitting hole of a flat cork which is placed on the top board of the feeding device. The corks are 50 x 50 mm., with a central hole slightly smaller than the outer diameter of the feeding tubes. Following adjustment to proper feeding tube temperature and speed of stirrers, blood or other liquid material to be fed is placed in the tubes. The mosquitoes are held in ½ pint ice-cream cartons, the top and bottom of which are covered with a fine mesh nylon gauze. These are positioned so that the membrane rests on the top gauze layer. After each feeding, the tubes are washed and, if necessary, can be stored and used repeatedly.

SUMMARY. A description is presented

of a modification of the Greenberg feeding device which has been useful in feeding blood and other suspensions to mosquitoes. The improvements consist especially in the use of a membrane prepared from the crop of the chicken, and in the addition of a stirring mechanism to prevent sedimentation of suspended materials.

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EFFECTIVENESS OF CARBON DIOXIDE AS A MOSQUITO ATTRACTANT IN THE CDC MINIATURE LIGHT TRAP

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INTRODUCTION. Various models of light traps have been employed to determine adult mosquito population indices and to collect large numbers of specimens for disease transmission studies. Since phototropism is exhibited to varying degrees among mosquito species, a light source in a trap has not always proved to be the ideal attractant.

Carbon dioxide released from either compressed gas cylinders or dry ice has been used successfully as an attractant for collection of mosquitoes. Headlee (1934), Reeves (1951), and Reeves and Hammon (1942) conducted field studies showing that various mosquito species were attracted to carbon dioxide gas.

Results of tests [reported by Newhouse et al. (1966)] indicated that the total number and variety of mosquitoes collected with light traps baited with carbon dioxide was significantly greater than those traps unbaited.

Procurement and economics are of primary consideration when using carbon dioxide; therefore, it is essential to determine the minimum amount of carbon dioxide which will increase the mosquito collection rate.

MATERIALS AND METHODS. The study was conducted for 8 weeks at Edgewood Arsenal, Maryland, in a wooded area near the Post Pumping Station. Six CDC Miniature Light Traps were suspended from tree limbs in an area of approximately 3,200 square yards. Four of the light traps were equipped to release carbon dioxide from 50 pound compressed gas cylinders at the rate of 250 c.c., 500 c.c., 1000 c.c., and 2000 c.c. per minute respectively. The fifth light trap was operated with no light source and set to release 500 c.c. per minute. The sixth light trap (control) was operated with a standard light source but with no carbon dioxide. Light traps were operated 5 to 7 nights per week; however, all traps were not operated simultaneously due to un-