

ience in assembling, numbers or codes are painted on the inside and outside surfaces of each panel. In practice, it took 3 men 4 hours to erect the hut, but only 2 hours to disassemble it. A large canvas overlapping the roof on all sides will prevent leakage during heavy rains.

During operation of the hut, the person infected with microfilariae slept from 9:00 p.m. to 6:00 a.m. Before he left in the morning, all mosquitoes in the hut were collected and dissected immediately. Three blood smears were taken just as he retired, to determine the average number of microfilariae per 20 cmm. of peripheral blood. Other smears may be taken during the night. For comparison, it is recommended that morning collections also be made when the hut has been unoccupied, because it was found that on occasions fully-fed newly infected mosquitoes flew into the hut when it was unoccupied. This necessitates using a correction factor

in interpreting the results when a person occupies the hut.

Depending on the nature of the investigation, statistical data are tabulated as regards pickup of microfilariae by mosquitoes which fed on the infected volunteer, evidence of prior infection, comparison of results of occupied and unoccupied hut, etc.

Literature on the use and assembling of Dexion may be obtained from Dexion, Inc., 39-27 59th Street, Woodside, Queens, New York, or from Dexion, Ltd., Maygrove Road London, N.W. 6, England.

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THE USE OF LIGHT IN REARING *ANOPHELES QUADRIMACULATUS* SAY FOR BEHAVIORAL STUDIES

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INTRODUCTION. Field tests in which sterile male *Anopheles quadrimaculatus* Say were released to evaluate their effect on natural population levels and reproductive potential have been conducted in two areas in Florida (Weidhaas *et al.*, 1962). Males were reared from a laboratory colony, sterilized with 12,000 r of gamma radiation, and released into the natural population. Sampling of females collected in or around the release area failed to demonstrate that the release of sterile males adversely affected the natural population or caused any appreciable degree of sterility in it.

Further studies (Weidhaas and D. A. Dame, in manuscript) were conducted to evaluate the effect of release of males, sterilized with chemicals, on the mating behavior of individuals in the natural population and in the laboratory colony. These studies demonstrated that there were most likely behavioral differences between the laboratory-reared and wild strains of mosquitoes. Three factors appeared to support this finding: (1) All attempts to mate wild individuals in the laboratory failed; (2) field tests indicated that males from the laboratory colony may not mate effectively in the field with wild

females, and (3) wild males mated with laboratory females in the field but progeny of wild males and laboratory females mated poorly in the laboratory. Therefore, it appeared that the laboratory strain and the wild strain were not morphologically isolated as far as transfer of the sperm was concerned, but behavioral differences may have prevented the strains from interbreeding.

In order to study the mating habits of the two strains, considerable time was spent watching both strains of mosquitoes in cages inside the laboratory and out-of-doors. Because the wild mosquitoes were not active until sunset, all observations were restricted to that time of day. Since observations made at sunset did not fit into the regular working hours, it was desired, if possible, to establish an artificial-lighting system which would permit observations on the mating habits during the regular working hours.

METHODS. Before any environmental chambers were constructed or any tests conducted, the rate of decrease in the level of illumination during sunset on a clear day (October 16, 1961) at Orlando, Florida, was determined with a Weston illumination meter (Model 756)¹ These measurements are presented in Table 1. Observations in the field indicated that wild mosquitoes became active about 45 minutes to 1 hour after sunset.

Two illumination boxes (24 x 18 x 30 inches high) were constructed so that the level of illumination could be regulated by fluorescent lamps attached to the ceilings of the boxes. General Electric¹ daylight fluorescent lamps were selected as the sources of illumination because their distribution of radiant energy is very close to that of the sun reaching the earth (Weitz, 1956). One of the boxes was equipped with lamps so that there were only two levels of illumination (low intensity), but in the other box there were

TABLE 1.—Levels of illumination during sunset on a clear day, October 16, 1961, Orlando, Florida.

Time (p.m.)	Foot-candles	Appearance of sky
5:15	115	Sun visible
5:20	115	"
5:25	70	"
5:30	45	"
5:35	29	"
5:40	20	Sunset
5:45	16	"
5:50	12	Horizon red
5:55	6	"
6:00	3	Red in sky gone
6:05	1.4	"
6:10	0.5	Horizon red
6:15	0.1	"

several levels (high intensity), which corresponded with those observed in the field during sunset. Table 2 presents the levels

TABLE 2.—Levels of illumination used in the two rearing boxes.

Time	Foot-candles	
	High-intensity box	Low-intensity box
2:00 p.m.—2:00 a.m.	0	0
2:00 a.m.—2:30 a.m.	0.25	0
2:30 a.m.—2:45 a.m.	2	0
2:45 a.m.—3:00 a.m.	20	10
3:00 a.m.—1:00 p.m.	120	10
1:00 p.m.—1:15 p.m.	20	10
1:15 p.m.—1:30 p.m.	2	0
1:30 p.m.—2:00 p.m.	0.25	0

of illumination used in the 2 boxes. One screen-wire mosquito cage (15 x 10 x 12 inches high) was placed in each of the boxes, which were maintained at approximately 75°–85° F. and 40–80 percent relative humidity.

In each test the eggs were placed in the boxes, the larvae reared there, and the adults allowed to emerge until each cage had approximately the same number of adults. However, in test 2 with the laboratory strain, the mosquitoes were not placed in the cage until the adults were 2 days old. After the adults had been in the cage for at least 6 days and had an opportunity to feed on a guinea pig or

¹Mention of companies or products in this paper does not imply recommendation or endorsement by the U. S. Department of Agriculture over others not mentioned.

human arm, they were taken from the illumination box and placed in a cold room (34° F.) until immobilized. All of the females that had taken blood were placed in individual vials, returned to the illumination box, and allowed to oviposit. Those females that had not taken blood were dissected and the spermathecae checked for insemination.

Since wild mosquitoes are found in stumps, tree holes, and other such dark locations during the day, two 1-quart ice-cream cartons, painted black on the inside and provided with a 1-x 1½-inch hole in each end, were placed on the floor of each cage for the mosquitoes to hide in while the lamps were in operation. In one test (laboratory strain—test 4), the influence of these black cartons on the behavior of the mosquitoes was determined. Two mosquito cages were placed inside the high-intensity box, one containing an ice-cream carton and one without a carton.

RESULTS. Laboratory Strain—Test 1. The following observations were made during this test. (1) When the illumination was at the maximum level in both cages, about 95 percent of the mosquitoes were inside the ice-cream cartons. After illumination in the high-intensity box was reduced to about 2 foot-candles, the mosquitoes started coming out of the cartons. When illumination was reduced to 0.25 foot-candle, nearly all mosquitoes came out. The mosquitoes in the low-intensity box did not come out of the cartons until the lamps were turned off. (2) When the level of illumination was reduced to 0.25 foot-candle in the high-intensity box, mating was observed. Mating was so closely correlated with the level of illumination that it was possible to predict it within a 15-minute interval. Mating was not observed in the low-intensity box. (3) After the adults had been in the cage for at least 6 days, they were removed from the cage, counted, and the females checked for insemination, blood feeding, and oviposition. Table 3 presents these results. The percent insemination obtained in other laboratory tests with the laboratory

TABLE 3.—Results of three tests with Orlando laboratory strain of *Anopheles quadrimaculatus* mosquitoes caged in low- and high-intensity boxes with an ice cream carton available for resting (34–50 each of males and females per test).

Data obtained at end of test ¹	Percent in	
	High-intensity box	Low-intensity box
Test 1		
Females alive	83	89
Inseminated	100	56
Blood fed	90	66
Ovipositing	79	31
Test 2		
Females alive	76	51
Blood fed	53	24
Ovipositing	42	19
Test 3		
Females ovipositing	65	74

¹ Percent inseminated, blood feeding, and ovipositing based upon the number of females alive at end of test.

strain (unpublished data) has been about 65–80 percent; rarely does it exceed 80 percent.

Laboratory Strain—Test 2. Results were the same as those in test 1. The number of females that were inseminated, that blood fed, and that oviposited are presented in Table 3.

Laboratory Strain—Test 3. As soon as these adults emerged, it was noticed that they were behaving more like the wild strain. They were as active in the cage as the other laboratory mosquitoes but appeared to be seeking a place to get out of the cage and not flying around inside as happened in other tests with the laboratory strain. In addition, mating was not as evident as in the previous two tests. These results (Table 3) indicated that the conditions in the high-intensity box were not conducive to increasing the percent of females ovipositing.

Laboratory Strain—Test 4. The only observations made in this test were those on the percent of females that were inseminated, that blood fed, and that oviposited. Results (Table 4) indicated that the

TABLE 4.—Results of test with Orlando laboratory strain of *Anopheles quadrimaculatus* mosquitoes caged in low- and high-intensity boxes with and without ice-cream carton available for resting (50 each of males and females per test).

Data obtained at end of test ¹	Percent in cage	
	With carton	Without carton
Females alive	86	74
Inseminated	100	87
Blood fed	66	81
Ovipositing	56	65

¹ Percent inseminated, blood feeding, and ovipositing based upon the number of females alive at end of test.

presence of an ice-cream carton had no influence on insemination, blood feeding, and oviposition.

Wild Strain—Test 1. Eggs collected from wild females were placed in the high-intensity box and allowed to hatch. Following are the observations made on 99 adults: (1) About 95 percent of the adults were in the ice-cream cartons when the lamps were on; they did not come out until the level of illumination had been reduced to 0.5 foot-candle, which was a little lower than the level at which the laboratory mosquitoes became active; (2) mating was not observed; (3) none of the females was inseminated; and (4) the wild mosquitoes were as active in the cage as the laboratory strain but appeared to be seeking a place to get out of the cage and not flying around inside the cage as noted in most of the tests with the laboratory strain.

Since mating was not observed and dissection of spermathecae showed that none occurred, blood feeding and oviposition were not determined.

Wild Strain—Test 2. A second test was conducted with 104 adults, but the results were the same as those in the first test with the wild strain.

CONCLUSIONS. (1) The crepuscular activities of the Orlando laboratory strain of *Anopheles quadrimaculatus* Say can be shifted to any desired time of the day by use of an artificial illumination system such as used in the high-intensity box.

(2) Mating can be observed with the laboratory strain.

(3) The percent of laboratory females inseminated, blood feeding, and ovipositing may possibly be increased by using an artificial illumination system as in the high-intensity box.

(4) The use of ice-cream cartons as a resting place during the light period does not appear to affect the response of the laboratory strain.

(5) The environmental conditions used in the high-intensity box are apparently not conducive to mating for the wild strain.

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