DEVELOPMENT AND SURVIVAL OF A NATURAL POPULATION OF *AEDES AEGYPTI*

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ABSTRACT. The development time of larvae of *Aedes aegypti* (L.) in the natural population was much longer than in the laboratory, primarily because less larval food was available. However, temperature also had an influence. Nevertheless, the survival of these stages in nature was high. Survival of the larval stages and egg production of the adult females were used to estimate average daily survival of adult females for different rates of population growth. Ovitraps, and animal- and carbon dioxide-baited traps were tested for effectiveness in collecting eggs or adults.

Before we can use new or integrated approaches to the control or management of total populations of vector species of mosquitoes, we must understand these populations, i.e., the dynamics, as they actually exist in nature. The development times and rates of survival of the various stages of mosquitoes are of particular importance. Once we can estimate these parameters, we can better understand the dynamics of populations, can construct models to predict growth at certain conditions, and can judge the feasibility of different approaches to control. Therefore, we arranged to study a population of *Aedes aegypti* (L.) found breeding (predominantly in washing machines) in a holding area for discarded appliances in the center of the city of Gainesville, Florida. Preliminary observations showed that the population was a naturally occurring one with all developmental stages of the insect present and was representative of *A. aegypti* that breed in containers in this urban area. Moreover, since the population persisted over several months, which indicated that it was cycling, we were able to study certain aspects of its dynamics. The present paper presents the results of the studies we made to determine the development times and rates of survival of the immature stages (egg through pupa) of the population and the egg laying potential of the adult females. These data were then used to estimate rates of survival of adult females that would be consistent with different rates of population increase. The results obtained with the ovitraps and adult traps used to sample the population are also reported.

METHODS

DEVELOPMENT AND SURVIVAL OF IMMATURE STAGES. Observations in the test area indicated that the immature stages of *A. aegypti* were breeding predominantly in rain water that had collected in discarded washing machines stored outdoors. This water did not contain an abundance of larval food; however, eggs, pupae, and all larval instars were found in most machines. We therefore investigated the development times and survival of these immature stages and compared them with: (a) the development times and survival in the same water with laboratory larval food added and (b) the development times and survival of larvae reared in the laboratory insectary. We also checked the number of adults emerging daily from other washing machines.

For the laboratory study of development, we collected adult females from the area, offered them blood from guinea pigs, and collected the eggs on wet filter paper. Known numbers of these eggs were conditioned for 6 days and hatched in the laboratory by flooding the eggs with water for 36 hours. When hatch occurred, 300 first stage larvae were placed in an enamel rearing pan containing 1 liter of distilled water and fed a mixture of equal parts of brewer's yeast and liver powder daily. The

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time required for development of all these that survived to the 4th larval, pupal, and adult stages and the number surviving were recorded.

For one field test, 4 galvanized iron tubs were placed on the ground near natural breeding sites. Then 10 liters of water were siphoned into each tub from washing machines that contained *A. aegypti*. Also, a small amount of detritus from the washing machines was added to each tub to approximate natural conditions. All immature stages of *A. aegypti* present in the tubs were removed, and the tubs were covered with screen enclosures and allowed to stand for 7 days to insure that no naturally occurring immature forms were present. Then, papers containing known numbers of eggs obtained as for the laboratory test were placed in each of the 4 tubs; the percentage egg hatch was checked after 36 hours; and the number of first-stage larvae in each of the tubs was adjusted to 400. In 3 tubs, no additional larval food was introduced; in the 4th tub, a combination of yeast and liver powder was added daily. The minimum and maximum water temperatures were recorded daily. Development times and survival were recorded from the first-stage larvae to the adult stages.

For the other field test, we picked third-stage larvae from washing machines and placed 150 in each of 3 other washing machines from which all immature stages had previously been removed. These latter machines were then covered with emergence traps, and the adults emerging daily were collected and recorded.

EGG PRODUCTION OF ADULT FEMALES. The study of the egg production of the females of these populations was made with insects taken from the same sources studied previously. Thus, the males and females emerging from (1) laboratory rearing, (2) tubs with no food added, and (3) washing machines seeded with third-stage larvae were counted, and a representative sample of adult females from each source was weighed (before feeding). Then, 100 males and 100 females from each source were caged so we could determine egg production and female survival. The adults from the laboratory rearing were held in a cubic-meter cage in the laboratory; the adults from the two field sources were held in similar cages outdoors. All cages contained sugar-water and had guinea pigs placed in them daily as food sources. An oviposition substrate (moist filter paper) was also available at all times. Both the number of females surviving and the number of eggs laid were recorded daily for laboratory-reared adults; the number of eggs only was recorded daily for the field adults.

MODEL OF POPULATION PARAMETERS. Our studies produced single values for the survival of immatures and egg-laying potential of adult females (number of eggs per live female per day). We were unable to measure adult survival under actual field conditions. However, it was possible to estimate the average daily adult survival values necessary to maintain a stable population (rate of increase=1), an increasing population (e.g., rate of increase=2, 5, or 10), or a decreasing population (e.g., rate of increase=0.5) from 1 generation time to the next as follows:

$$R=[\Sigma p\times\frac{m}{2}]Si$$

where R = rate of increase
p = probability of survival of adult females=average daily survival
m = average number of eggs per live female per day
Si = survival of immatures

TEST OF TRAPS FOR SAMPLING FOR EGGS AND ADULTS. Twenty-two black jar ovitraps were placed at selected sites on the periphery and in interior of the breeding area and left in place for 3 consecutive weeks. Wooden paddles (tongue depressors) were placed in the jars for oviposition. The paddles were removed weekly and examined for eggs. New paddles and fresh water were added each week.

Also, 3 lard-can trapping devices baited

with either guinea pigs or chickens were placed on the periphery of the breeding area so that each animal was used on 3 occasions as bait.

Finally, octagonal-shaped plexiglass traps (Schreck *et al.* 1972) emitting carbon dioxide from an attached cylinder were used. The traps were set up at 4 pm and removed the next morning around 9 am. All trapped mosquitoes were collected, identified, and recorded.

RESULTS

DEVELOPMENT AND SURVIVAL OF IMMATURES. Table 1 summarizes the development and survival of immature stages of *A. aegypti* in the laboratory and in the field. In the laboratory, hatching of eggs (85%) and percentage and survival from first instar to adult emergence (0.97) were high, and development was rapid (9 days from 1st instar to complete adult emergence). In the field, the development time of larvae was longer, and the survival of immature stages and the percentage hatch were reduced. Availability of larval food appeared to be the major factor responsible for the differences: when food was added to breeding water in the field, the time for development was reduced from 39 to 13 days. However, temperature had some effect and was probably the major reason for the differences in development times between larvae reared in the laboratory and those reared in the field with extra larval food provided. For example, the average minimum and maximum daily temperatures of the water in the field were 23° and 29° C; the average water temperatures in the laboratory were relatively constant at 30° C. Although larval survival was lower in the field than in the laboratory, it was high (0.87 with food added and 0.67 without food added) despite the extended development time. (The duration of the pupal stage—2 days—was identical at all locations.)

TABLE 1.—Survival and development times of immature stages of *A. aegypti* in a field location in Gainesville, Florida, compared with survival and development of laboratory-reared mosquitoes.

Datum	Laboratory	Field with food added	Field with no food added
No. eggs	353	638	1914
% Hatch of eggs	85	62.7	62.7
No. 1st stage	300	400	1200
No. 4th stage	293	361	950
No. pupae	292	352	812
No. adults	290	347	798
Survival			
1st to 4th stage	.977	.903	.792
4th to pupa	.997	.975	.855
pupa to adult	.993	.986	.983
egg to adult	.822	.544	.417
1st stage to adult	.967	.868	.665
Development time (days) for 1st instar to adult			
Mean	8	12	24
Complete	9	13	39

A total of 288 adults emerged over 23 days from the 450 third-stage larvae placed in breeding water in washing machines. This time of development (approximately 4th instar to adult emergence) agrees closely with the 22 days observed in tubs (data not presented). However, survival was only 0.64, which was lower than the comparable value of 0.85 for the same stage in tubs. This decreased survival may have resulted in part from the transfer of larvae from one rearing medium to another.

EGG PRODUCTION OF ADULT FEMALES. A total of 20,625 eggs was obtained in 39

days from the 100 laboratory-reared females caged in the laboratory with an equal number of laboratory-reared males. (At 39 days, when 7 females were still alive, observations were stopped.) Our first check for eggs was made 7 days after the mosquitoes were caged; thus, some eggs may have been laid earlier. The number of eggs laid per day per live female was variable (0–55) and no clear cut pattern or gonotrophic cycle was evident though there were peaks in daily oviposition. Therefore, we do not report the data for daily oviposition. If we assume that eggs were laid first on day 6 rather than on day 7, the average daily production of eggs per live female was 17. Those females and males emerging from natural breeding water (tubs and machines) without food added and held outdoors in field cages produced fewer total eggs, but they also died more rapidly. The 100 females emerging from washing machines produced 14,775 eggs over the 32-day maximum lifespan; the same number of females emerging from tubs produced 12,700 eggs over 28 days. However, among both groups held in the outdoor cages, there were definite ovipositional peaks at 4-day intervals. The females in these outdoor cages also began oviposition 2 days later than females held in the laboratory.

The average weight of females reared in the laboratory was 2.25 mg; the averages for those reared outdoors in tubs or washing machines without additional food were 1.40 and 1.34 mg, respectively.

The mortality of the laboratory-reared females was recorded daily, and the regression line of mortality over time (see Figure 1) was linear ($b=-2.00$; $r^2=0.98$). However, this type of female survival is probably more typical of caged insects than of those in field populations. The mortality of the field-reared females was not recorded daily, but regression lines for the mortality of females emerging from tubs or washing machines are assumed to be linear and are presented in Figure 1. Then with these regression lines and the known daily production of eggs, we could calculate the average daily production of eggs per live female as 14.5, 14.2, and 13.9 for those reared in the laboratory, those reared outdoors in tubs, and those reared outdoors in washing machines, respectively. The 14.5 calculated from the regression equation obviously agrees closely with the 17 calculated earlier.

MODEL OF POPULATION PARAMETERS AND GROWTH. The ratio of the number of females in a given generation or period of time to the number in the preceding equivalent unit is a measure of the growth rate of a population. It is also a function of survival and egg production. In the laboratory where survival can be maintained at high levels, rates of population growth should be high. For example, our laboratory-reared females laid 206 eggs per female or 103 female eggs per female. Then since 0.82 of these survived to emerge as adults, a single female produced 84 adult female progeny.

Such high rates of growth are generally not expected in the field because of reduced rates of survival. Our results verify this expectation. At the conditions existing, the survival of immature forms was 0.42 (0.627 of the eggs hatched and 0.67 of the resulting first-instar larvae emerged as adults). (Since we did not reflood the eggs, the actual survival could have been somewhat higher.) We therefore calculate that daily rates of survival of adult females of 0.61, .67, .73, .81, and .86 are required for growth rates of 0.5, 1.0, 2.0, 5.0, and 10.0.

SAMPLING FOR EGGS AND ADULTS. The 22 ovitraps collected a total of 1233 eggs over a period of 3 weeks. Thus, the average was only about 60 per trap and 2.7 per trap per day. Not all the traps, however, were positive at all times, and many paddles often had few (1–10) eggs. Traps set on the edge of the field closest to dwellings collected the most eggs.

A total of 28 female and 14 male *A. aegypti* was collected in the chicken-baited traps in three samplings. A total of 6 female and 2 male *A. aegypti* was collected from the traps baited with guinea pigs.

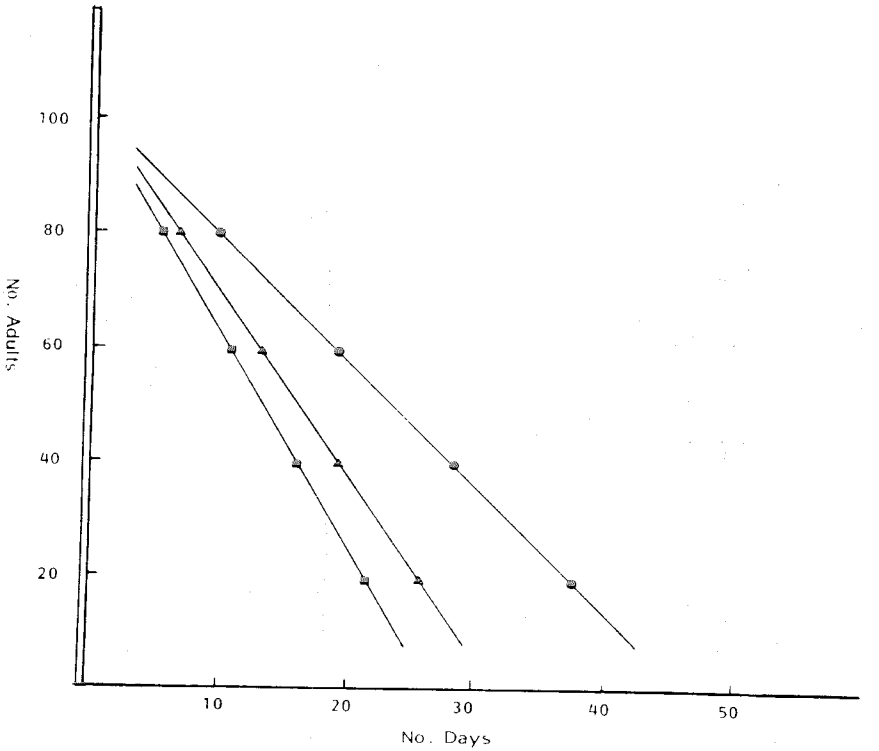

FIG. 1.—Regression lines for mortality of adult females from 3 sources (types of rearing). The lines for females from tubs and washing machines are hypothetical.

●=laboratory females
■=tub females
▲=emergence trap females

(Also, a very few *Culex quinquefasciatus* Say and *A. infirmatus* Dyar and Knab were sometimes found.) A total of 32 female and 12 male *A. aegypti* was collected in three samplings from the plexiglass traps baited with carbon dioxide. On two occasions, these traps were continued in operation during the day, but only very few mosquitoes were trapped during that time.

DISCUSSION

Aedes aegypti is a container breeding mosquito, and the population we studied may be typical of this mosquito in areas of similar climatic conditions. In natural breeding water, the immature stages required 39 days for complete (100%) development. The shortage of food appeared to be primarily responsible for this extended period since the addition of food decreased the time almost to that observed in the laboratory. Moreover, in natural breeding water without additional food, development time showed about a 2-fold increase from 1st to 3rd instar larvae and

a 5-fold increase in the 4th instar compared with the times in similar water with food added.

The high survival of the immature stages in nature indicates that the impact of density-regulating factors at this period were minimum in this population. For example, survival increased from 0.67 to 0.87 (1.3X) when food was added, but even if all immature forms had survived, the increase would have been only 1.5X. The number of immature forms per breeding site was low in this population, and we prepared the tubs with only 400 larvae in 10 liters (4 larvae/100 ml). If larvae occur naturally in more crowded conditions, the survival of these stages could be much lower. Southwood *et al.* (1972) found that in Thailand, survival of *A. aegypti* larvae ranged from 0.07 to 0.52 over a 10-month period and except in October (0.52) and February (0.26) was below 0.15. However, they did not indicate the density of larvae in the breeding containers. They also measured egg survival in the field and showed a range in egg hatch of 0.03–0.36, which was far less than the egg hatch of 0.62 that we observed.

Sheppard *et al.* (1969) calculated rates of survival every 2 weeks for adult *A. aegypti* in the study area used by Southwood *et al.* They found a range of 0.783–0.959 but all but 2 values were greater than 0.82. Their values for survival of adults combined with our data on survival of immature stages would indicate growth rates greater than 5-fold. However, in a tropical area such as Thailand, *A. aegypti* would breed continuously, and regulation of a population would most likely relate to the effect of density-dependent factors on the survival of immature stages due to food. In contrast, the cool weather of the northern part of Florida causes a break in mosquito breeding there. Also, Southwood *et al.* showed both low survival of immature stages and a mean development time of 18.57 days as opposed to our observations of high survival and a mean development time of 24 days. The length of development time would not have as much effect on population size in a continuously breeding population as on a population that is interrupted by adverse climatic conditions. For example, in our area, there are 4 months with temperatures favorable to the development of *A. aegypti,* and our observations were made at the peak of the favorable conditions. A mean development time of 24 days for the immatures allows about 5 immature cycles per year. We believe that the factors which limited population growth in our study area were the combination of long development times and the cold weather of winter.

Adult females emerging from the field population weighed less than those reared in the laboratory, a difference attributable to larval nutrition (Colless and Chellapah 1960). However, Miura and Takahashi (1972) showed a correlation between the size of females, the amount of blood ingested, and the number of eggs laid. Thus, we expected that the smaller females emerging in the field would produce fewer eggs than the larger females from laboratory rearing. However, our data did not show this effect. Since we did not observe the daily mortality of females caged in the field, we do not know whether the difference in egg production between the laboratory and field was caused by size or survival.

The various trapping devices we used did not collect large numbers of adults or eggs. However, we did not have estimates of the absolute densities of the population, so we cannot estimate their efficiency.

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SPERMATOGENESIS IN *AEDES ALBOPICTUS* (SKUSE) [1]

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ABSTRACT. Chromosome behavior during spermatogenesis in *Aedes albopictus* was studied and compared with results of similar studies. Fourth instar larvae, pupae, and adults of the Mauritius strain were dissected at intervals of one hour after these stages had begun. Squash preparations stained with 2% lacto-aceto-orcein were utilized exclusively. Leptotene and zygotene stages were not observed. The chromosomes first became visibly distinct in pachytene. In early pachytene the chromosomes were usually closely paired, but some asynaptic regions were observed. A prometaphase stretch was not seen. At anaphase I the separation of the dyads was asynchronous with the smallest pair often separating first. In prophase II polarization of the chromosome arms was observed. Separation of the chromosomes at anaphase II was synchronous, and the chromosomes were individually recognizable. The developing spermatids gradually became elliptical and contained alternating regions of light and dark staining material. The elliptical spermatids then elongated to form mature sperm.

INTRODUCTION. Work on spermatogenesis in mosquitoes appears to have been neglected when compared to the literature available in such areas as taxonomy, genetics, control, and ecology. Most early studies involving meiosis or spermatogenesis utilized the *Culex pipiens* complex (Stevens 1910, Lomen 1914, Taylor 1914, Whiting 1917, Moffett 1936, Grell 1946, Callan and Montalenti 1947, and Jost 1971). Other earlier studies included the following species: *Culex tarsalis* (Stevens 1911); *Anopheles punctipennis* (Stevens 1911); *Culiseta incidens* (Stevens 1911); *Corethra plumicornis* (Frolowa 1929); *Anopheles maculipennis* (DeBuck and Swellengrebel 1935); and *Theobaldia longiareolata* (Callan and Montalenti 1947). Most recent studies include the following species *Anopheles stephensi* (Rishikesh 1959); *Aedes aegypti* (Akstein 1962); *Culiseta inornata* (Breland *et al.* 1964); *Aedes aegypti* (Krafsur 1964); *Aedes aegypti* (Mescher and Rai 1966); *Aedes dorsalis* (Mukherjee and Rees 1970); and a very brief description of meiosis in *Aedes albopictus* (Jost 1971). In addition, Krafsur and Jones (1967) described the process of spermiogenesis in *Aedes aegypti*.

MATERIALS AND METHODS. All specimens utilized in this study belonged to the Mauritius strain of *Aedes albopictus* currently maintained at the Mosquito Genetics Laboratory, Georgia Southern College. The colony originated from specimens collected by the second author in Mauritius during March of 1970. Eggs of this strain were hatched in deoxygenated water. Larvae and pupae were raised in distilled water in white enamel pans (23 cm x 35 cm x 5 cm) at

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