AEDES TRISERIATUS: PERSISTENCE OF NULLIPAROUS FEMALES UNDER FIELD CONDITIONS¹

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ABSTRACT. Eighteen (67%) of 27 marked female Aedes triseriatus recaptured at ages 9–13 days were nulliparous. Fifty-five (96%) of 57 marked females recaptured at ages 15–31 days were uniparous (51) or biparous (4), indicating that nearly all females either obtained a bloodmeal or died before attaining 15 days of age. Sixty bloodseeking uniparous females

ranged from 9-31 days of age at time of recapture; the mean age of 17 of these females from a group that was sampled throughout the period when uniparous individuals were present was 15.6 days. One marked female, 19 days of age and uniparous, was recaptured in a woodlot separated from the experimental woodlot by 425 m of cropland.

As part of an effort to accumulate information on the population structure and behavior of *Aedes triseriatus* we conducted a small experiment to determine how long *Ae. triseriatus* may persist in the field in the nulliparous condition. The experiment was designed also to yield preliminary information on the age of uniparous females seeking a 2nd bloodmeal and to give some indication of how much time is spent in the search for hosts, mates and oviposition sites.

MATERIALS AND METHODS

The basic procedure was to release a known number of marked mosquitoes in a small woodlot, presumably isolated enough from other woodlots to prevent emigration, and then to recover and dissect as many as possible in order to relate their physiological age to their calendar age in days. The woodlot was 0.9 ha in size, bordered on the north by a highway, and on the remaining sides by cropland. The nearest wooded areas were a 2 ha woodlot 425 m to the southwest and a narrow band of trees along a fencerow 235 m to the west. That the woodlot chosen was a suitable habitat for A. triseriatus was confirmed by the finding of a large

basal treehole that supported a resident population. This treehole was plugged and no other treeholes were found.

The experiment was conducted during late July and August, 1977. Mosquitoes were released on July 25 and on alternate days thereafter through August 4, a total of 6 releases. The released mosquitoes were the F₁ progeny of adults reared from larvae collected from natural treeholes in a non-LAC endemic area. Upon emergence as adults the sexes were separated to prevent mating in the laboratory. Every 2 days during the release sequence 100 females and an appropriate number of males were drawn from the accumulated emergents, the females marked with fluorescent dust, and both sexes released in the center of the woodlot. The ratio of released males to females was based on seasonal ratios observed in a natural population (Scholl and DeFoliart 1978) and varied from 1.5 to .26 males to each female.

Briefly, the marking process entailed placing the females in a 4-liter ice cream carton which had a hole in the side 2 cm above the bottom and Saranwrap stretched across the top (Sinsko and Craig 1979). By removing the center cardboard from the lid, the remaining rim served to hold the plastic wrap in place. Six pure colors of fluorescent pigment (Dayglo Co., Cleveland, Ohio) were available and a different color was used for each release. With the mosquitoes inside the container, pigment dust was bounced off the

¹ Research supported by the College of Agricultural and Life Sciences, University of Wisconsin, Madison, and by NIH Grant AI-07453.

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plastic at the top by means of a powder insuffulator (De Vilbiss #119).

Visual observations in the woodlot indicated that there were few resident or transient mammals or birds. Therefore, 3 large New Zealand white rabbits were caged in the area to insure ample availability of hosts. Two days before beginning efforts to recover released mosquitoes, the rabbits were removed to enhance the attractiveness of the collectors as bait

Eight days after the last release, biting collections were initiated in an effort to retrieve as many marked mosquitoes as possible. Collections were attempted from sunrise to sunset on August 12, 14, and 16; thereafter, late afternoon collections were made every other day through September 5, or until no marked females were recovered in 3 successive collections.

Recovered females were captured individually in test tubes and returned to the lab at the end of each collecting day. After being anesthetized with CO₂, each was examined for fluorescent pigment with longwave ultraviolet light. Each female was then dissected to determine fertilization success and physiological age. Parous and nulliparous individuals were distinguished by the tracheal method of Detinoya (1962) and by the dilatation tech-

nique of Polovodova (1949). The latter technique was used to determine the number of completed gonotrophic cycles.

RESULTS AND DISCUSSION

Only 2 of 57 mosquitoes captured when 15 days of age or older were nulliparous (Table 1), indicating that most of the mosquitoes either obtained a bloodmeal or died before attaining that age. Some nulliparous mosquitoes were removed by collections in the 3 age groups sampled prior to age 15 days (Release Groups #4–6), but this bias was not present in groups in which sampling began on the 15th day or later (Release Groups #1–3). The oldest nulliparous mosquito recaptured was 17 days old.

From the data (Table 1) it is obvious that, under the conditions of this experiment, there was great variation in the time required by females to find hosts and obtain bloodmeals. Uniparous individuals recaptured while seeking a bloodmeal ranged from 9–31 days of age, and at least 12% of Release Group #6 were nulliparous and non-bloodfed at 9 days of age when uniparous individuals began to form part of the bloodseeking fraction of the population.

The 9-day-old bloodseeking females

Table 1. Age and parity condition of marked Aedes triseriatus at time of recapture.

Age (days)	Release group no.1						01 - £
	$\frac{1}{\mathbf{U}}$	$\frac{2}{U}$	3	4	5	6	% of total parous
			NU	\overline{N} U	NU	NU	
9						8 3	27
11					1 0	4 2	29
13	•			2 0	3 2	0 2	44
15			0 1	1 5	0 5	0 0	92
17		2	1 2	0 3	0 0	0 5	92
19	0	ō	$\overline{0}$ $\overline{1}$	0 2	0 3	0 1	100
21	ŏ	ĺ	0 1	0 2	0 2	0 3	100
23	ŏ	Õ	0 1	0 0	0 2	0 0	100
25	ŏ	2	0 1	0 1	0 0	0 1	100
27	0	1	0 1	0 0	0 0	0 0	100
29	0*	1 **	0 0	0 0	0 0*	0 0	100
31	0	0	0 0	0 1	0 0	0 0	100
%	-						
recovered	01	09	09	17	19	29	

¹ N=nulliparous; U=uniparous; each asterisk represents one biparous ♀.

that were uniparous apparently succeeded in attaining mates, hosts and oviposition sites in near minimum time. We have laboratory data (unpublished) showing that, at 22°C, the minimum intervals from eclosion to acceptance of a proffered bloodmeal is 2 days; from bloodmeal to completion of oviposition is 5 days: and from completion of oviposition to acceptance of a proffered host is 4 hours or less. Foster and Lea (1975) reported that, at 27°C, approximately 20%, 50%, and 80% of female Ae. triseriatus were receptive to insemination on posteclosion days 3, 4 and 5, respectively. These laboratory data indicate, then, that some females are physiologically capable of re-entering the biting population as uniparous individuals at 8-9 days of age if minimum time is used in seeking a host, mate, and oviposition site.

The mean age of uniparous bloodseeking females at time of recapture in our experiment can be calculated only for females in Release Group #6 as none of the other groups, apparently, were sampled from the beginning of the period when uniparous mosquitoes might have entered the bloodseeking fraction of the population. Although it can be considered as indicative only, because of the small number of included females, the mean age of the 17 uniparous females from Group #6 at time of recapture was 15.6 days. This is 7.6 days longer than the minimum required but it is not possible, from currently available data, to estimate the average proportion of the interval spent in searching for hosts as opposed to other variables such as search time for mates and oviposition sites, or above minimum time required by some individuals for ovarian development. Mates were apparently found more readily than hosts, however, as all but one of the nulliparous females had been inseminated before recapture. Also, oviposition sites were presumably readily available, as 9 ovitraps were installed in the woodlot at the beginning of the releases.

Only 5% of recovered mosquitoes were biparous and all of these were 29 days of

age when recovered attempting a 3rd bloodmeal. This compares with 8% of feeding females found to be biparous in a natural population (unpublished results), and indicates that only a small percentage of females survive to the 3rd bloodmeal in nature.

Sinsko and Craig (1979) found no interchange of Ae. triseriatus between 2 woodlots in Indiana that were separated by 300 m of open terrain. Prior to our 1st releases, a 6-week period of egg monitoring with ovitraps indicated that neither resident nor immigrant Ae. triseriatus were present in our test woodlot. It became apparent however, as our experiment progressed that there was egress, and, probably, ingress by Ae. triseriatus despite our effort to choose a site that was completely isolated. The most conclusive proof was the finding of a marked 19day-old female in a woodlot 425 m to the southwest of the test plot, despite the fact that prevailing winds were from the southwest, and only 9 man-hours were spent searching there for marked Ae. triseriatus. The frequent occurrence of Ae. triseriatus eggs in an ovitrap attached to a small isolated tree 70 m from the western edge of the test plot provided additional evidence of movement into and out of the test woodlot. Finally, the number of unmarked Ae. triseriatus captured in the test area increased from a mean of 2.3 per collection day in late July to 3.8 in August and to 6.3 in early September. Most of these unmarked individuals appeared very fresh, however, and it is possible that some of them originated within the test woodlot from arboreal treeholes possibly present but undetected during the earlier egg monitoring because they were dry, but which became activated by unusually heavy rainfall during July and August.

References Cited

Detinova, T. S. 1962. Age-grouping methods in Diptera of medical importance with special reference to some vectors of malaria. Monog. Ser. Wld. Hlth. Org. 47:1–216.

Foster, W. A. and A. O. Lea. 1975. Sexual behavior maturation in male Aedes triseriatus

(Diptera: Culicidae): a re-examination. J. Med. Ent. 12:459-63.

Polovodova, V. P. 1949. The determination of the physiological age of female *Anopheles* by the number of gonotrophic cycles completed. Medskaya Parazit. 18:352–55 (In Russian).

Scholl, P. J. and G. R. DeFoliart. 1978. The

influences of seasonal sex ratio on the number of annual generations of *Aedes triseriatus*. Ann. Entomol. Soc. Am. 71:677–70

Sinsko, M. J. and G. B. Craig, Jr. 1979. Dynamics of an isolated population of *Aedes triseriatus* (Diptera: Culicidae). I. Population size. J. Med. Entomol. 15(2):89–98.

WHITE-EYE, A MUTANT IN THE MOSQUITO ERETMAPODITES QUINQUEVITTATUS THEOBALD¹

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ABSTRACT. A recessive mutant is described in *Eretmapodites quinquevittatus*. The mutant, white-eye (w), exhibits complete penetrance and uniform expressivity. It can be

INTRODUCTION

White eye appears to be a common eye color mutant in many of the species of mosquitoes where genetic studies have been carried out. It has been found in Anopheles pharoensis, Anopheles gambiae and Anopheles quadrimaculatus (Kitzmiller and Mason 1967), Anopheles stephensi (Aslamkhan 1973 and Sharma et al. 1977), Culex pipiens (Gilchrist and Haldane 1947 and Laven 1967), Culex tarsalis (Barr and Myers 1966), Culex tritaeniorhynchus (Baker 1969), Aedes aegypti (Bhalla 1968) and Aedes cooki (Wade 1977). White eve is sex-linked for all these species except Cx. tarsalis where it appears to be autosomal and An. stephensi where the mutant colorlesseye (a white eye) described by Sharma et al. (1977) is autosomal. Thus the white eye mutant reported for Eretmapodites quinquevittatus in the present study is at least the third white-eyed mutant that segredetermined in the larvae, pupae, and adults of both males and females. Experimental cross data indicate that w segregates independently of sex (m), and that w is epistatic to red-eye (re).

gates independently of sex to be described in mosquitoes.

MATERIALS AND METHODS

The mutant white-eye (w) was first isolated in 1976 by the second author. Several white-eved male and female pupae were isolated from the EQ-MIXED strain of Er. quinquevittatus. The resulting adults were crossed and their progeny inbred to establish a pure-breeding white-eye strain which was designated EQ-W. Two other strains of Er. quinquevittatus, EQ-PURE with normal eye phenotype and EQ-RE/ GL homozygous for the red-eye mutant (re) (Hartberg and Johnston 1977), were also used in this investigation. All of the strains used were selected from the colonies maintained at the Mosquito Genetics Laboratory, Georgia Southern College.

Rearing methods used were generally similar to those described by Hartberg and Gerberg (1971) for rearing Er. quinquevittatus. Rearing was in an insectary

¹ This investigation was supported in part by a grant from the Georgia Southern College Faculty Research Fund to the senior author.