EGG RETENTION IN MOSQUITOES (DIPTERA: CULICIDAE) AND ITS RELATION TO PHYSIOLOGICAL AGE

L. A. MAGNARELLI

Department of Entomology, Cornell University, Ithaca, N.Y. 14853 1

ABSTRACT. During the summer of 1973, 1,159 females were obtained from four dry ice-baited CDC Miniature Light Traps at sites 9.5 km. south and 6 km. east of Ithaca, N. Y. The incidence of egg retention and physiological age were determined for 1,093 females representing species of Aedes, Anopheles, Coquillettidia, Culex and Culiseta. The Aedes stimulans group, Aedes canadensis, and Coquillettidia perturbans females were most frequently encountered in collections.

Relict eggs can sometimes be seen in ovarioles of parous mosquitoes. It is not clear why some females retain mature eggs after oviposition, but egg retention has been frequently reported by investigators in conjunction with age-determination studies. Christophers (1911) suggested the utilization of egg retention information for estimating age in female mosquitoes. In her dissection of Anopheles maculipennis Meigen females that had recently oviposited, Detinova (1962) reported that ovarioles retaining mature eggs had a completely normal appearance. However, ovarioles with relict eggs were changed morphologically in those females that had obtained another blood meal; the follicle lying above the relict egg changed shape and then showed signs of degeneration. Detinova further observed that mature eggs retained in ovarioles may also degenerate. Bellamy and Corbet (1973, 1974) indicated that in nulliparous Culex tarsalis Coquillett numerous dilatations can result from resorbed follicles and that these dilatations may be misinterpreted as an indication of the parous condition (i.e., that the female had laid eggs). Similar accounts describing follicular degeneration and egg resorption are given

by Gillies (1958) and Clements (1963). Incidence of egg retention is significant since relict eggs may signify parity and since the presence of these eggs may denote follicular degeneration and "false dilatation" formation in certain ovarioles. The present study utilized mosquitoes collected near Ithaca, N. Y. and was conducted to determine the incidence of egg retention in parous specimens and the relationship of egg retention to physiological age.

MATERIALS AND METHODS. Female mosquitoes were collected from four CDC Miniature Light Traps (Sudia and Chamberlain, 1962). The traps were operated at least one night per week for 14 weeks from late May to early September 1973 at Brooktondale, N. Y. (9.5 km. south of Ithaca, N. Y.) and at Ellis, N. Y. (6 km. east of Ithaca). At each site, the traps were suspended from tree branches adjacent to woodland snow-melt pools in a beech-maple forest. The traps were at least 100 meters apart and were each baited with about 0.5 kg. of dry ice. The dry ice was placed in a plastic bag and hung about 30 cm. above each trap. The light traps operated from about 2000 hours until ogoo hours.

Since the female adults of Aedes stimulans (Walker), Ae. excrucians (Walker), and Ae. fitchii (Felt and Young) are taxonomically similar and the identification of older females is often difficult, these

Percentages of parous females containing relict eggs ranged from 8% in the Aedes stimulans group to 23% in Anopheles quadrimaculatus. Follicular degeneration and egg resorption were occasionally observed in uniparous Coquillettidia perturbans and Aedes canadensis. There was no apparent relationship between the number of retained eggs and physiological age of females; of the parous specimens with relict eggs, the majority had only one or two eggs.

¹ Present address: Connecticut Agricultural Experiment Station, Box 1106, New Haven, Conn. 06504.

species were grouped during this study. For similar reasons, Ae. abserratus (Felt and Young) and Ae. punctor (Kirby) were combined.

Mosquitoes were dissected on the day of capture or were frozen until dissections could be performed. Physiological age was determined by Polovodova's method (in Detinova, 1962), and the stage of follicular development was recorded for females following Christophers' (1911) classification as modified by Mer (1936). The dissections were performed in Ringer's physiological saline solution under a stereomicroscope at 30X magnification; ovarioles were teased apart, and dilatations were observed at 150X. The number of relict eggs per female was recorded.

RESULTS AND DISCUSSION. Results were compared for each species at the two sites. As there were no appreciable differences due to trap location, the data were pooled (Table 1). Twelve mosquito species, rep-

(Walker) females were most frequently encountered in light trap samples.

Sixty percent of the dissected specimens were parous, but the incidence of egg retention for each species was low. Figures ranged from a low of 8% egg retention in the Ae. stimulans group to 20% in An. punctipennis (Say), 22% in Ae. vexans (Meigen), and 23% in An. quadrimaculatus Say. The higher figures may be somewhat inflated for both Anopheles species and Ae. vexans since these values were based on relatively fewer parous individuals. Hitchcock (1968) reported an egg retention rate of 4.5% for 2,983 parous An. quadrimaculatus.

In the 94 specimens with relict eggs, most females retained only one or two eggs. However, an An. quadrimaculatus female retained 17 stage V eggs, and one An. punctipennis female had 14 eggs. In each of these Anopheles specimens, the ovarioles without relict eggs were in the

Table 1. Incidence of egg retention and physiological age of female mosquitoes collected in CDC Miniature Light Traps at Brooktondale and Ellis, N. Y., 1973.

Species	Total caught	Total dissected	No. of pars	% of Pars re- taining eggs	Relict eggs per female		No. of pars with eggs and age class		
					*	range	1-P**	2-P	3-P
Ae. abserratus-punctor	106	92	64	16	1.5	1-2	7	3	О
Ae. canadensis	185	171	89	14	1.5	1-2	8	3	1
Ae. stimulans group	231	230	161	8	1.6	1-7	10	2	1
Ae, vexans	45	45	27	22	2.5	1-6	5	1	0
An, punctipennis	91	81	49	20	3.5	1-14	8	1	0
An. quadrimaculatus	108	95	57	23	3.4	1-17	12	I	0
Coq. perturbans	203	194	126	15	1.7	1-4	14	3	2
Culex restuans	119	115	62	13	1.3	1-3	6	2	O
Culiseta morsitans dyari	71	70	20	15	1.6	1-3	2	I	0
TOTALS	1,159	1,093	655				72	17	5

* \bar{x} excludes pars that did not retain eggs.

** 1-P=Uniparous, 2-P=Biparous, 3-P=Triparous; figures represent minimum number of completed gonotrophic cycles.

resenting five genera, were present in light trap collections. A total of 1,159 females was collected, of which 1,093 were dissected to determine physiological age and the incidence of egg retention. The Ae. stimulans group, Ae. canadensis (Theobald), and Coquillettidia perturbans

sac-stage (i.e., posterior portions of follicular tubes were distended). For most sac-stage uniparous *Coq. perturbans* and *Anopheles* females, each ovariole that had a relict egg also had nurse cells differentiated from the oocyte in the secondary follicle, but no yolk had been deposited

(stage I). In those ovarioles without relict eggs, small amounts of yolk (stage I-II) had already accumulated in secondary follicles. These observations suggest that relict eggs in ovarioles may be affecting yolk deposition and follicular development in secondary follicles. Although these findings suggest delay in yolk deposition of certain follicles, there was no evidence to indicate follicular degeneration or egg resorption in any sac-stage ovarioles. However, in six uniparous Ae. canadensis and eight Coq. perturbans females with dilatations (contracted follicular tubes), relict eggs were partially decomposed, and each secondary follicle adjacent to relict eggs was irregular in shape and without nurse cells or oocyte.

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In studies conducted in Maryland, Hitchcock (1968) reported that the incidence of retained eggs increased with physiological age in An. quadrimaculatus, but that the number of eggs retained had no apparent relationship to physiological age. He also found that there was a greater incidence of egg retention in the spring and fall. The results of the present study agree with his finding that the number of eggs retained has no apparent relationship to physiological age, but since small numbers of parous specimens were recorded, no reliable comparisons can be made with his other results.

Although the presence of a few relict eggs may signify a completed gonotrophic cycle, egg retention information by itself is of little value in the determination of oviposition activity on a seasonal basis. Knowledge of the incidence of egg retention may, however, be very important in the interpretation of dilatations and physiological age. During ovariole examination of Ae. canadensis, the Ae. stimulans group, Coq. perturbans, and Anopheles specimens, it was not uncommon to observe a few ovarioles with two dilatations while the remaining ovarioles of the same ovary had only one dilatation. Detinova (1962) discussed abortive oogenesis in relationship to the presence of relict eggs, and she hypothesized that

relict eggs in some ovarioles could be responsible for degeneration of secondary follicles. Based on observations of the present study, it is possible that after degeneration of the secondary follicle, a small dilatation forms anterior to the relict egg; then, following resorption, the second dilatation of these ovarioles forms at the site of the relict egg. In light of this possibility, it would be erroneous to assume that two dilatations indicated two completed gonotrophic cycles. An alternative explanation for the variability in dilatation number is that the female may produce significantly fewer eggs during the second gonotrophic cycle.

It is apparent that there is a need for extensive laboratory studies (comparable to Detinova's work with *An. maculipennis*) on blood-feeding patterns, oocyte development, oviposition, and dilatation formation in each species to determine whether or not the number of dilatations per ovariole represents the minimum or maximum number of completed gonotrophic cycles. Also, it is important to emphasize that not only can dilatations result from resorbed follicles in nullipars (Bellamy and Corbet, 1974), but similar processes do occur in parous individuals as well.

Acknowledgments. I wish to thank Dr. E. W. Cupp of the Department of Entomology at Cornell University for his criticism of this manuscript.

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