

AUTOGENY IN *CULEX ANNULIROSTRIS* FROM AUSTRALIAB. H. KAY<sup>1</sup>, J. D. EDMAN<sup>2</sup> AND P. MOTTRAM<sup>1</sup>

**ABSTRACT.** In the laboratory, 8.5% and 5.1% of colonized *Culex annulirostris* from Brisbane and Mildura, Australia respectively were autogenous when reared and maintained on nutrient rich diets. Females reared and/or maintained on poor diets mainly had ovaries at Christophers' stage I and exhibited from 0 to 0.7% autogeny. All autogenous females had previously mated. Insemination rates in the Brisbane and Mildura colonies respectively, were 72.8% and 78.8%. No autogeny was detected in 997 females reared from 7 localities throughout Queensland but this may have been due to their poorer nutritional status (as indicated by wing size) or more likely to a low insemination rate of 0 to 16%. Our laboratory results, particularly with well-fed females, may have little relevance to the field situation, where adults are generally smaller and less well-nourished.

## INTRODUCTION

*Culex annulirostris* Skuse is considered the major vector of arboviruses such as Murray Valley encephalitis, Kunjin and Ross River virus in Australia (Doherty 1974). On biological and morphological characteristics, this species could be considered a counterpart of *Cx. tarsalis* (Coquillett) in North America and *Cx. tritaeniorhynchus* Giles in Asia (Reeves et al. 1954). As with *Cx. tritaeniorhynchus*, *Cx. annulirostris* has been recognized as being anaautogenous, in contrast to *Cx. tarsalis* which produces eggs both autogenously and anaautogenously (Kardos 1959, Spadoni et al. 1974).

During 1982, while dissecting *Cx. annulirostris* reared on different diets, JDE noted that 19 of 111 females reared on a rich diet (8.4 mg/larva) had well developed ova either at stage IV or V (Christophers 1911). As these adults had been held in a 0.7 m<sup>3</sup> cage on 25% sucrose solution for 12–14 days, this suggested (a) autogenous egg development or (b) unobserved blood feeding on an attendant during the process of food replenishment and cage cleaning. This paper reports findings relating to option (a).

## MATERIALS AND METHODS

To facilitate studies of vector bionomics, 2 colonies of *Cx. annulirostris* were established; one from Shepparton, Victoria (McDonald et al. 1977), which was later supplemented several times with specimens from Mildura, and one from Brisbane (Mottram et al. in press). Spermathecae of females from both colonies were examined to establish the insemination rate.

Batches of 250 newly hatched *Cx. annulirostris* were taken at random from colony oviposition trays containing 50–100 egg rafts and pipetted

into 420 × 310 mm rearing trays containing 500 ml tap water. One group was reared on a high diet, 7.5 mg/larva, using ground Harper's "Dog Chow" and yeast in a 3:1 ratio for all days except the first when 100 mg powdered yeast was provided. Pupae were harvested and placed in two 0.7 m<sup>3</sup> cages, one with 25% sucrose and the other with 5%. The second group of larvae were reared on a poor diet of 2.5 mg/larva and adults maintained on 5% sucrose.

In order to estimate differences in the rate of autogeny between sibling groups, egg rafts were reared individually on high diet and the adults maintained on 25% sucrose. After 12–14 days, all of these females were stored at -79°C for ovary dissection. The number of first instar larvae from any rafts produced were counted.

Late instar larvae and pupae were collected from nutrient-rich effluent ponds at 7 localities in Queensland (Dalby, Tara, Mitchell, Roma, Charleville, Townsville and Kowanyama) and the adults maintained for 7–15 days with 25% sucrose solution. Some nulliparous females were also collected from Kowanyama using a battery-powered aspirator (Kay 1983). The ovaries and spermathecae of the females were examined.

Females were judged to be autogenous if sugar fed individuals contained 1 (or more) mature (Christophers' stage IVb or V) ovum after the 12–14 day holding period. Nutritional status was determined by measuring wing length from the axillary incision to wing tip.

## RESULTS

Wing length of females reared on high and low diet respectively, ranged from 3.6 to 3.8 mm and 3.1 to 3.3 mm. Of those *Cx. annulirostris* fed high diets as larvae and 25% sucrose as adults, 163 of 1,915 (8.5%) and 66 of 1,292 (5.1%) of the Brisbane and Mildura colonies respectively, were autogenous (Table 1). For the Brisbane colony, expression was significantly greater in those with both high larval and adult nutrition (8.5%) compared to those fed a high larval but poor adult diet (0.7%) ( $\chi^2$ ,  $P < 0.01$ ),

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Table 1. Ovarian development of colonized *Culex annulirostris* after 12 to 14 days following sucrose feeding.

Locality	Larval diet	% sucrose	Ovarian stage*								Total
			I	IIa	IIb	IIIa	IIIB	IVa	IVb	V	
Brisbane	high	25	32.7	35.4	20.9	1.3	0.5	0.7	2.2	6.3	1,915
	high	5	63.2	30.1	3.9	0.7	1.3			0.7	152
	low	5	100								189
Mildura	high	25	42.8	24.8	24.6	2.0	0.2	0.5	0.5	4.6	1,292

\* After Christophers (1911).

or those fed poorly throughout (0%). Ovarioles of this latter group showed the least development of the 3 groups examined.

The insemination rate of 7-12 day old females from the Brisbane colony was 72.8% (250 dissected) whereas 78.8% of 231 females of similar age from the Mildura colony contained sperm. All spermathecae of 51 females judged to be autogenous (Brisbane 32, Mildura 19) were positive.

There was considerable variation in the autogeny rates of rafts reared individually. For Brisbane rafts (n = 16), the mean ( $\pm$  1 standard deviation) number of female progeny dissected was  $77.8 \pm 26.1$  with an average autogeny rate of  $9.3 \pm 9.8\%$  (range 0-30.9%); for Mildura (n = 7),  $70.5 \pm 33.9$  females/raft with an autogeny rate of  $4.8 \pm 6.9\%$  (range 0-19.3%). The correlation between egg raft size (as number of females dissected) and the autogeny rate was not significant (R = -0.086).

Of those females judged to be autogenous, the mean number of mature ova from the Brisbane and Mildura colonies respectively, was 32.7 (n = 84) and 39.6 (n = 45). Rafts laid by autogenous females from the Brisbane colony contained a mean of 47.6 eggs (n = 55). Attempts to establish an autogenous colony from this stock failed.

The ovaries of 997 females reared from 7 field sites throughout Queensland were at Christophers' stage I, IIa or in one case, IIb. However, the insemination rate after 7-15 days in 0.7 m<sup>3</sup> cages was low, from 0 to 16% (Table

2). Female wing length ranged from 3.2 to 3.5 mm.

## DISCUSSION

This is the first report of autogeny in *Cx. annulirostris* and although the rates were generally low, they offer an additional means of survival for this species. From the data on individual raft rearings, it would seem that this trait is variably distributed throughout siblings of rafts of all sizes and may reach 31% (or even higher). However, fecundity resulting from sugar fed autogenous females in the Brisbane and Mildura colonies was much lower (32-48 eggs/raft) than that from blood-fed anautogenous development; quoted as 37-355 eggs/raft for the precursor of the Mildura colony (McDonald et al. 1977).

Previous studies of *Cx. tarsalis* have linked high nutritional status to the expression of autogeny (Kardos 1959, Spadoni et al. 1974), and this also seems applicable to *Cx. annulirostris*. *Culex annulirostris* reared on high diet and maintained on high percentage sucrose showed greater ovarian development than those fed inferior diets. The failure to find autogeny in the field populations tested may be due to two factors, (1) their nutritional status was inferior to that of high diet mosquitoes produced in the laboratory, as indicated by wing length, but more importantly, (2) most were uninseminated.

For most mosquitoes, mating does not di-

Table 2. Ovarian development in field collected *Culex annulirostris*, 7 to 15 days after feeding with 25% sucrose.

Locality	% positive spermathecae	Larval diet	Ovarian stage after 7-15 d			Total
			I	IIa	IIb	
Dalby	5	sewage				
Roma	7	nutrients,	98.8	1.2		81
Mitchell	16	followed by high	72.6	27.4		102
Tara	6	laboratory diet	87.6	12.4		129
Charleville	0		53.6	44.9	1.5	69
Townsville	0		97.4	2.6		311
Kowanyama	0		100			305

rectly affect the development of the initial egg batch (O'Meara 1979). However, blood-feeding studies of virgin *Cx. annulirostris* would indicate that mating is an important but not an essential factor influencing ovarian development (Kay 1978). From this and because all autogenous females were mated, we suggest that autogeny in *Cx. annulirostris* is possibly the male-induced form that has been described by O'Meara (1979) for one type of *Ae. taeniorhynchus* (Wiedemann). Consequently, future studies of autogeny in field populations of *Cx. annulirostris* should be made either with inseminated females or by inoculation of male accessory gland fluid into virgin females.

To date, autogeny has been associated with few Australian mosquitoes: the malaria vector, *Anopheles hilli* Woodhill and Lee (Sweeney and Russell 1973), Ross River virus vector *Ae. vigilax* (Skuse) (Sinclair 1976), *Cx. pipiens molestus* Förskal (= *Cx. pipiens* Linn.) and *Ae. australis* (Erichson) (Dobrotworsky 1965). With respect to the vector potential of *Cx. annulirostris*, because nutrient levels under field conditions are usually inferior to those in the laboratory (especially high diet level), we suspect that expression of autogeny would rarely occur in nature.

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