HYBRIDIZATION OF *AEDES TAENIORHYNCHUS* (WIEDEMANN) WITH *AEDES NIGROMACULIS* (LUDLOW) OR *AEDES SOLLICITANS* (WALKER) BY INDUCED COPULATION [1, 2]

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ABSTRACT. *Aedes taeniorhynchus* (Wiedemann) was hybridized reciprocally with *Aedes nigromaculis* (Ludlow) or *Aedes sollicitans* (Walker) by induced copulation. The viability of the F_1 eggs of reciprocal *A. taeniorhynchus*—*A. sollicitans* crosses was greater than that of the F_1 eggs of reciprocal *A. taeniorhynchus*—*A. nigromaculis* crosses. Hatched larvae of the crosses developed normally and produced healthy adults except that the F_1 male (*A. nigromaculis* x *A. taeniorhynchus*) and the F_1 male (*A. sollicitans* x *A. taeniorhynchus*) lacked vigor and did not fly. The posterior spiculation of the larval anal saddle and the absence of the adult abdominal dorsal median longitudinal stripe characteristic of *A. taeniorhynchus* and the dark and light scaling of the wings characteristic of *A. nigromaculis* and *A. sollicitans* adults were consistently present in all hybrids. However, in general, the traits of *A. taeniorhynchus* were dominant. Both sexes of the adult hybrids were sterile, as would be expected from interspecific crosses. Although *A. taeniorhynchus* and *A. sollicitans* often breed in the same habitat, behavioral and genetic inconsistencies should prevent hybridization.

The possibility of using genetic incompatibility to control mosquito populations has stimulated new interest in studies of hybridization. However, in some instances, hybridization between species (especially eurgyamic populations) can only be accomplished by induced copulation.

Previous attempts at interspecific hybridization by induced copulation have produced varied results. McClelland (1962) obtained negative results in crosses of the *Aedes* in the subgenus *Stegomyia*. Ellis and Brust (1973) reported that the few resulting F_1 larvae from crossing *A. churchillensis* Ellis and Brust males with *A. communis* (DeGeer) females died during the moult to the second instar. Horsfall and Brust (1962) obtained normally developing hybrids from reciprocal crosses of *Psorophora ferox* (Humboldt) and *P. horrida* (Dyar and Knab) but all were sterile. Also, we (Fukuda and Woodard 1974) recently successfully hybridized the two closely related species *A. nigromaculis* (Ludow) and *A. sollicitans* (Walker) and demonstrated complete fertility in the hybrids and backcross combinations. Additional hybridization studies are reported here.

MATERIALS AND METHODS. The colonies of *A. sollicitans* and *A. taeniorhynchus* maintained in our laboratory were started from field collections made in southwestern Louisiana. The colony of *A. nigromaculis* was started from eggs obtained from the University of California Mosquito Control Research Laboratory, Fresno, California. The *A. taeniorhynchus* colony is maintained by natural mating in cages, and the *A. nigromaculis* and *A. sollicitans* colonies are maintained by induced copulation.

All cross-matings were accomplished by induced copulation by using modifications of the method described by Miura (1967). The procedures used to achieve egg deposition and hatching, the method of rearing larvae, and the preparation of adult hybrids for mating and backcrossing were described by Fukuda and Woodard (1974). Since *A. taeniorhynchus* mate naturally in

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[2] Mention of a proprietary product in this paper does not constitute a recommendation or an endorsement of this product by the USDA.

TABLE 1. Results of force mating *Aedes taeniorhynchus* (T) with *A. nigromaculis* (N) or *A. sollicitans* (S).

Crosses		No. of females force-mated	Percent of females ovipositing	Percent viable egg batches
Female	Male			
N	T	40	60	50
T	N	60	68	59
S	T	20	90	67
T	S	20	75	67

cages, male and female *A. taeniorhynchus* and male and female hybrids were separated as pupae and allowed to emerge in separate cages.

Aedes taeniorhynchus was mated in both directions with *A. nigromaculis* and *A. sollicitans*. A number of larvae and adults of the parent types and hybrids were preserved and mounted for observation as described by Fukuda and Woodard (1974). An attempt was made to backcross and interbreed the remaining adults.

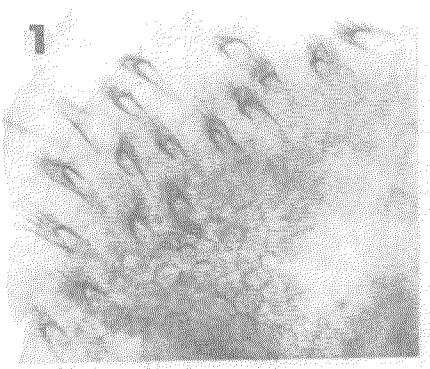
Egg embryonation was determined by placing the eggs in a Clorox® solution; collapse of the chorion was taken to indicate lack of embryonation. Fertility of hybrid males was determined by dissection and microscopic examination of the reproductive organs for the presence of sperm. Sterility of female hybrids was assumed when negative results were observed from backcrossing.

RESULTS AND DISCUSSION. The results of cross-mating *A. taeniorhynchus* with *A. nigromaculis* or *A. sollicitans* are summarized in Table 1. The mating efficiency of reciprocal *A. taeniorhynchus* and *A. sollicitans* crosses approached the 70–80 percent efficiency of normal mating in the colonies. Reciprocal *A. taeniorhynchus*—*A. nigromaculis* crosses were less efficient and also produced a greater number of partially hatched egg batches. The unhatched eggs showed no embryonation. The hybrid larvae from all crosses developed and pupated normally, but the adult F_1 male from *A. nigromaculis* x *A. taeniorhynchus* and from *A. sollicitans* x *A. taeniorhynchus* lacked vigor and did not fly. The adults of the other crosses were vigorous and flew normally.

When the hybrid adults were forcemated, copulation occurred, but we found no sign of sperm transfer; that is, there was no strand of seminal fluid between the genital pore of the male and the atrium of the female when they were separated. Only the F_1 males (*A. nigromaculis* x *A. taeniorhynchus*) had sperm in the reproductive organs, but these males did not copulate when mating was attempted. Since the hybrid males were sterile or unable to copulate, only hybrid females were backcrossed to parent males. However, of the 78 backcrossed females, only 12 produced eggs, and none of these eggs possessed developing embryos.

The major differences between larvae of *A. taeniorhynchus, A. nigromaculis,* and *A. sollicitans* are the number, shape, and arrangement of pecten teeth, and the number and shape of comb scales. In the case of the pecten teeth, *A. taeniorhynchus, A. nigromaculis,* and *A. sollicitans* larvae range from 11 to 17, from 16 to 23, and from 15–27, respectively. Also, *A. taeniorhynchus* and *A. sollicitans* have evenly spaced, barbed pecten teeth; *A. nigromaculis* has pecten teeth with the distal 1–3 teeth detached and smooth. In the case of comb scales, *A. taeniorhynchus, A. nigromaculis,* and *A. sollicitans* larvae range from 9 to 20, from 6 to 12, and from 11 to 21, respectively. Furthermore, *A. nigromaculis* and *A. sollicitans* have comb scales with a long apical spine (Fig.

FIG. 1. *Aedes taeniorhynchus* larva comb scales. FIG. 2. *Aedes taeniorhynchus* adult abdomen. FIG. 3. *Aedes nigromaculis* larva comb scales. FIG. 4. *Aedes nigromaculis* adult abdomen. FIG. 5. (*Aedes taeniorhynchus* x *Aedes nigromaculis*) F_1 larva comb scales. FIG. 6. (*Aedes taeniorhynchus* x *Aedes nigromaculis*) F_1 adult abdomen.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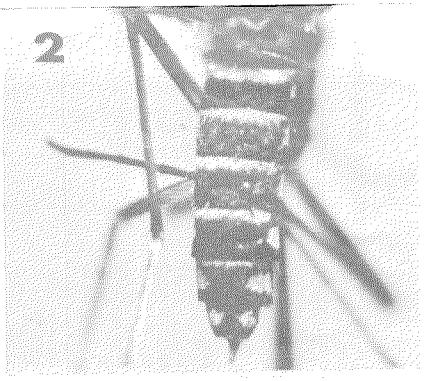
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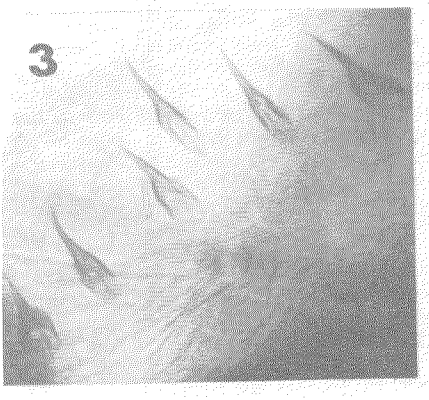
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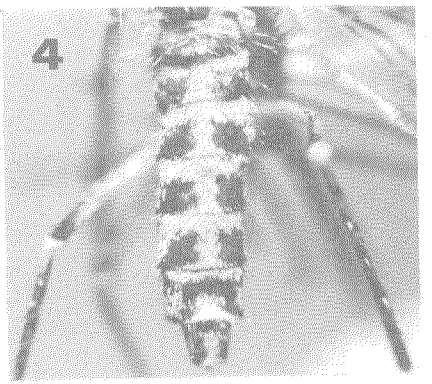
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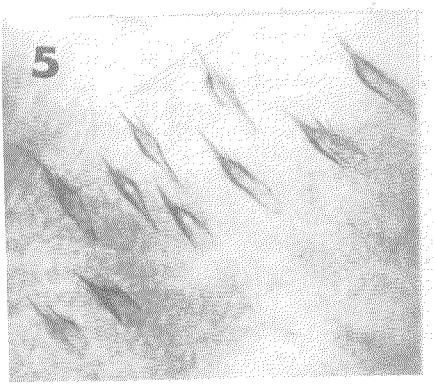
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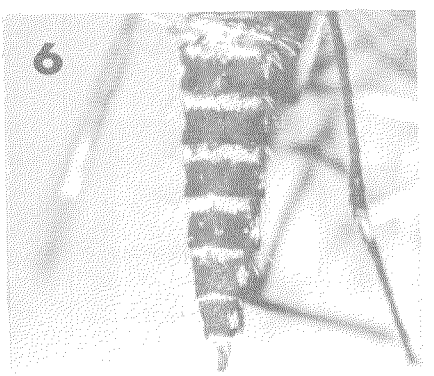
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TABLE 2. Mean and standard deviation (s.d.) of pecten tooth and comb scale counts, distal pecten teeth displacements, and siphon lengths of *Aedes taeniorhynchus* (T), *A. nigromaculis* (N), *A. sollicitans* (S), and the hybrids.

Crosses		No. specimens	No. of pecten teeth		Comb scales		Distance between distal two pecten teeth (microns)		Length of siphon in mm	
Female	x Male		Mean	s.d.	Mean	s.d.	Mean	s.d.	Mean	s.d.
T	T	20	15.6	1.7	18.5	3.5	31	6	0.689	0.025
N	N	13	16.6	2.3	8.2	2.7	143	38	0.887	0.064
S	S	15	20.1	1.7	23.1	4.1	40	7	0.899	0.047
N	T	20	16.5	1.9	10.0	2.3	70	18	0.776	0.035
T	N	20	16.6	1.8	12.6	4.6	60	20	0.764	0.038
S	T	18	18.8	2.0	19.0	3.3	34	3	0.757	0.032
T	S	20	18.7	2.6	18.3	2.8	30	4	0.723	0.038

3); *A. taeniorhynchus* (Fig. 1) has comb scales that are rounded apically. Also, *A. taeniorhynchus* has a shorter siphon than the other two species. The posterior spiculation of the anal saddle is a characteristic only of *A. taeniorhynchus* (Carpenter and LaCasse, 1955).

Table 2 summarizes the variations in the cited characteristics of larval *A. taeniorhynchus, A. nigromaculis, A. sollicitans,* and the hybrids observed in the present study. *Aedes taeniorhynchus* and *A. nigromaculis* were dissimilar in most characteristics except in the number of pecten teeth. Reciprocal hybrids of these two species had barbed pecten teeth with counts near those of the parents; pecten teeth displacement and siphon length of the hybrids were between the parent types but near to the values of *A. taeniorhynchus.* Comb scale counts of these hybrids were nearer to those of *A. nigromaculis.* Hybrids of *A. taeniorhynchus* and *A. sollicitans* reciprocal crosses had pecten tooth counts between those of the parent types but nearer *A. sollicitans;* in contrast, pecten teeth displacement, comb scale count, and siphon length were nearer the values of *A. taeniorhynchus.*

The posterior spiculation of the anal saddle characteristic of *A. taeniorhynchus* was carried to all hybrids. Also, the prominent apical spine of the comb scales (Fig. 5) of parent *A. nigromaculis* and *A. sollicitans* was carried to all hybrids. However, in general, the hybrid larvae tended to have characteristics near the *A. taeniorhynchus* parent.

Although adult *A. nigromaculis* and *A. sollicitans* are difficult to distinguish, adult *A. taeniorhynchus* has characteristics that separate it from the other two species, namely, wings with entirely dark scaling and an abdomen lacking the dorsal median longitudinal stripe (Fig. 2); in contrast, adult *A. nigromaculis* and *A. sollicitans* have wings with dark and light scaling and abdomens with a yellowish dorsal median longitudinal stripe (Fig. 4) (Carpenter and LaCasse 1955). The hybrid adults all possessed similar characteristics regardless of which species was

crossed with *A. taeniorhynchus* or in which direction the cross was made. The dark and light scaling of the wings, characteristic of *A. nigromaculis* and *A. sollicitans* was carried on to the hybrids; likewise the lack of the dorsal median longitudinal stripe of the abdomen characteristic of *A. taeniorhynchus* was common in all hybrids (Fig. 6). In general, the adult hybrids appeared more like *A. taeniorhynchus* than *A. nigromaculis* or *A. sollicitans*.

The difference in the success of the reciprocal *A. taeniorhynchus* crosses to *A. sollicitans* or to *A. nigromaculis* is probably the result of mating technique rather than an inherent characteristic of the species. Many more crosses would have to be attempted before any conclusive statement can be made.

The results of crossing *A. taeniorhynchus* with *A. nigromaculis* or *A. sollicitans* were similar to those obtained by Horsfall and Brust (1962) when they crossed *P. ferox* with *P. horrida* and are what would be expected from interspecific crosses. The crosses also differ greatly from the complete fertility of hybrids and backcrosses of the *A. nigromaculis—A. sollicitans* cross reported by Fukuda and Woodard (1974), but this is not surprising in view of the taxonomic relationship of these species.

Although *A. taeniorhynchus* and *A. sollicitans* are often found breeding in the same habitats, hybridization seems unlikely because of apparent genetic incompatibility and behavior differences.

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