

present writing (December 1975), 4 daughter generations have been reared, and the yield has been increased by a factor of 3.6. The colony is now regarded as being firmly established in our laboratory.

Currently, our efforts are directed toward bringing the colony to strength and the development of methods for mass-production of the species. The immature stages are presently reared in 6 x 5 cm ID plastic tubes with floors and inner coatings of plaster of paris. The rearing medium is commercial organic garden compost supplemented with liver powder. The adults are maintained in cylindrical 1-pint ice cream carton cages on 10 percent sucrose in demineralized water. The adult females are fed on hamsters. It is anticipated that present methods will be modified substantially as rearing experiments currently in progress proceed. At present the major problem is that of excessive mortality among the gravid females, as reported by Killick-Kendrick (1973).

The species *L. longipalpis* is a well-known vector of leishmaniasis in South America. The present colony was established for use in ongoing research on this disease and in studies directed toward the development of improved insect repellents for the protection of military personnel from vector-borne diseases. Interested investigators are invited to keep abreast of the progress of the colony as a source of starter material.

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## EFFECTS OF ABATE 2G® AND ABATE 4E® MOSQUITO LARVICIDES ON SELECTED NON-TARGET ORGANISMS COEXISTING WITH MOSQUITO LARVAE IN WOODLAND DEPRESSIONS

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As a follow-up of the laboratory work done by Dida et al. (1975), a study of the effects of Abate 2 G (o, o, o', o'-tetramethyl o, o' thioldi-phenylene phosphorothioate) and Abate 4E on

non-target organisms was carried out in the field. Dida et al. (1975) found that the organisms most susceptible to the insecticide were members of the Order Cladocera, (*Simocephalus* sp. and *Ceriodaphnia* sp.), and members of the Order Diptera (F. Chironomidae and *Culex pipiens* L.). The populations of the remaining non-target organisms sustained little or no mortality. It was determined that Abate 2G at a concentration of 2.5 lbs. per acre killed all cladocerans within 1 day. This mortality rate occurred for 2 consecutive days, when new populations were introduced. At higher application rates of 5.0 lbs. per acre, a total mortality was observed each day for 5 consecutive days. After the 5th day a total mortality occurred within 2 days, and after the 7th day, a mortality rate of approximately 30% occurred within 3 days. Similar mortality rates were found with chironomid and *Culex pipiens* larvae.

The non-target organisms used in this study and found in woodland depressions throughout the Bremen Township area of Illinois were the cladocerans, *Simocephalus* sp., and *Ceriodaphnia* sp., the copepods *Cyclops* sp., *Ectocyclops* sp., and *Eucyclops* sp., the ostracods (1 species), and 1 species of damselfly (*Lestes* sp.). The target organisms were the mosquito species *Aedes fitchii* (Felt & Young), *A. canadensis* (Theobald), *A. stimulans* (Walker) and *A. vexans* (Meigen).

Several woodland depressions serving as natural breeding sites for the above species of mosquitoes were selected within the boundaries of the South Cook County Mosquito Abatement District. These breeding sites were observed daily at approximately the same time. Rainfall and temperatures as well as rate of development and mortality of both target and non-target organisms were recorded. All the organisms studied were collected from each source by means of an enamel dipper (350 cc in volume). Ten dips of water were taken from each mosquito-breeding source and poured through a plankton net into a vial (50 cc in volume). This was done in order to concentrate the organisms and facilitate their transportation and storage. The population estimates were determined according to the stratified random sampling procedure for cladocerans, copepods and ostracods (Snedecor and Cochran 1971) and by actual counts for the damselfly nymphs and mosquito larvae. Observations on the fluctuations of the population of each organism were recorded for a period of 5 days before treatment until 10 days after the treatment.

To record the natural development and fluctuations of untreated organisms in the field, a control was set up which consisted of 2 enamel pans at each treated source. The enamel pans were filled with water and debris taken from the source and pretreatment population counts of all organisms collected from 10 dips were placed inside the pans. Each of these enamel pans was then placed inside a cut-off metal drum to protect it from being disturbed by animals or other unexpected forces.

The insecticide was applied to each source when most of the mosquito larvae were in the 2nd and 3rd instars. Abate 4E was introduced to the breeding site by using a 3-gallon B & G hand sprayer at an application rate of 1 oz of active ingredient per acre. A Buffalo turbine machine was used to apply the granular Abate 2G at a rate of 5 lbs. per acre.

Results of these field experiments showed that within a period of 24 hr after treatment a total mortality was observed for all 4 species of flood-water mosquitoes used in this study.

The non-target organisms varied in their mortality rates; some were more readily affected by the insecticide than others. All of them, however, returned to their normal population levels after 48 hours. The organisms most susceptible to the insecticide were the cladocerans. A maximum reduction of 40% of the population of cladocerans was achieved within 48 hours after the treatment. On the 3rd and 4th day after application of the chemical, an increase in the number of cladocerans was observed and the population had completely recovered by the 7th day.

The copepods, ostracods, and damselfly nymphs were only slightly affected. A decrease of 10 percent of the population of copepods and ostracods was observed after 24 hours, and the population of these groups of organisms returned to normal levels within 3 days after the treatment. Although damselfly nymphs were found somewhat sluggish in their movements no mortality was observed.

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#### LABORATORY EVALUATION OF THE ACTIVITY OF INSECT GROWTH REGULATORS AGAINST *CULICOIDES VARIIPENNIS* (DIPTERA: CERATOPOGONIDAE)

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Insect Growth Regulators (IGRs) have been shown to prevent the emergence of nuisance chironomid midges (Mulla et al. 1974a, 1975) and mosquitoes (Jacob 1973, Mulla et al. 1974b,

Sacher 1971). IGR compounds are especially promising as control agents since they are active against some Diptera resistant to conventional larvicides (Schaefer and Wilder 1972, 1973, Schaefer et al. 1974).

In a previous investigation (Apperson 1975) the biological activities of organochlorine and organophosphorus insecticides were determined against *Culicoides variipennis* Coquillett larvae. The population was found to be operationally resistant to these chemicals. As a consequence, and in our search for effective compounds, a variety of IGRs were tested in the laboratory. This report presents methods of evaluation and research findings.

**METHODS AND MATERIALS.** Field-collected larvae were tested after a 24 hr equilibration period. Twenty late 4th instars were transferred to glass petri dishes containing 100 ml of filtered water (pH 7.5-8.0) from Clear Lake. *Culicoides* require a substrate for pupation and adult emergence. Therefore 3 balls of cotton (Johnson and Johnson®) were placed in each bioassay container. Greater rates of emergence in controls resulted when the cotton fibers were loosened with forceps and spread throughout each petri dish.

The compounds tested were of technical grade. Solutions were prepared on a wt./vol. basis in acetone and diluted to give desired serial concentrations. From 1/2 to 1 ml was added to each bioassay dish, prior to addition of the cotton. Larvae were held in the same containers throughout the tests at 26-28° C under a 12:12 hr light: dark photoperiod. Water in each container was adjusted to pretreatment levels on a daily basis. Pupal exuviae were removed and counted each day until all adults had emerged or all larvae and pupae had died. Duration of the tests ranged from 6 to 9 days. Tests were set in duplicate and replicated at least 3 times on different days. Percent inhibition of emergence was adjusted relative to controls, which varied from 35 to 11%. Methods for testing IGRs against Ceratopogonidae have not previously been published.

The following compounds were evaluated: Dimilin® (TH-6040) (1-(4-chlorophenyl)-3-(2,6-difluorobenzoyl)urea) Thompson-Hayward Co.; Methoprene (Altosid®) (Isopropyl 11-methoxy-3,7,11-trimethyldeca-2,4-dienoate) Zeecon Corp.; R-20458 (4-ethylphenyl-6,7-epoxy geranyl ether) Stauffer Chem. Co.; Mon-858 (2,6, di-tert-butyl-4-( $\alpha, \alpha$ -dimethylbenzyl) phenol) Monsanto Chem. Co.; HE-24108 (3-butyl-2-yl N-(p-chlorophenyl) carbamate) Hercules Powder Co.; RO-203600 (6,7-epoxy-3-methyl-7-ethyl-1-[3,4-(methylenedioxy)phenoxy]=2- cis/trans-octene) Hoffman La Roche Co.

**RESULTS AND DISCUSSION.** Table 1 presents the effectiveness of IGRs against *C. variipennis* larvae. A broad spectrum of activity was manifested. Dimilin and methoprene were the most active compounds, achieving at least 90% inhibition of emergence at 0.5 ppm and 1.0 ppm, respectively. RO-203600 and HE-24108 were the least active.