Literature Cited

Brown, T. M. and A. W. A. Brown. 1974. Experimental induction of resistance to a juvenile hormone mimic. Jour. Econ. Ent. 67(6): 799–801.

Cerf, D. C. and G. D. Georghiou. 1972. Evidence of cross-resistance to a juvenile hormone analogue in some insecticide-resistant house flies. Nature 239(5372):401–402.

Cerf, D. C. and G. P. Georghiou. 1974. Cross-resistance to an inhibitor of chitin synthesis, TH-6040, in insecticide-resistance strains of the house fly. J. Agr. Food Chem. 22(6):1145–1146.

Henrick, C. A., G. B. Staal and J. B. Siddall. 1973. Alkyl 3, 7, 11-trimethyl-2, 4-dodecadienoates, a new class of potent insect growth regulators with juvenile hormone activity. J. Agr. Food Chem. 21(3):354–359.

Hsieh, M.-Y. G. and C. D. Steelman. 1974. Susceptibility of selected mosquito species to five chemicals which inhibit insect development. Mosq. News 34(3):278–282.

Jakob, W. L. 1972. Additional studies with juvenile hormone-type compounds against mosquito larvae. Mosq. News 32(4):592–595.

Rogers. A. J. and C. B. Rathburn, Jr. 1960. Airplane application of granular paris green mosquito larvicide. Mosq. News 20(2):105–110.

Schaefer, C. H. and Wilder, W. H. 1972. Insect development inhibitors: A practical evaluation as mosquito control agents. Jour. Econ. Ent. 65(4):1066–1071.

Schaefer, C. H. and W. H. Wilder. 1973. Insect development inhibitors, 2. Effects on target mosquito species. Jour. Econ. Ent. 66(4):913–916.

Thompson-Hayward Chemical Company. 1974. Technical information-TH-6040 insect growth regulator, 26 pgs.

Williams, C. M. 1956. The juvenile hormone of insects. Nature 178:212–312.

LABORATORY DOSAGE RESPONSE OF *AEDES TRISERIATUS* (SAY) TO ALTOSID® SR-10 AND 10-F[1]

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ABSTRACT. The dosage response of *Aedes triseriatus* (Say) was determined for 2 methoprene (Altosid®) formulations in laboratory tests. The LC_{50} and LC_{95} for the formulation, Altosid SR-10 was 0.135 ppb and 0.971 ppb, respectively, and for Altosid 10-F, 0.093 ppb and 0.363 ppb, respectively.

INTRODUCTION. In recent years many mosquito control programs have been dependent almost entirely upon the utilization of toxic chemical pesticides for larval and adult mosquito control. Development of widespread resistance to these chemicals, as well as the potential environmental hazards posed by their continued usage, has necessitated a vigorous search for new, effective, environmentally safe chemical control methods. The initial result of this search has been the development of insect developmental inhibitors or growth regulators, probably the best known of which is methoprene or Altosid® (Isopropyl 11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate). Two methoprene formulations were provided to this Agency for evaluation; a microencapsulated form registered as Altosid® SR-10 for use against floodwater mosquitoes, and Altosid® 10-F, an experimental formulation of finely ground charcoal impregnated with methoprene.

The dosage responses of several species of mosquitoes to various formulations of Altosid have been established (Hsieh and Steelman 1974, Jakob 1972, Schaefer and Wilder 1972); and, specifically, the dosage

[1] The opinions contained herein are those of the authors and should not be construed as official or reflecting the views of the Department of the Army. Mention of proprietary products is for the purpose of identification only and does not imply endorsement by the Department of the Army. The Altosid compounds were provided through the courtesy of Zoecon Corp., Palo Alto, CA. Address reprint requests to: Commander, US Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD 21010.

response of *Aedes triseriatus* (Say) to an emulsifiable concentrate formulation of Altosid was determined by Hsieh and Steelman (1974). No information, however, has been published regarding the dosage response of this mosquito species to either the microencapsulated formulation, Altosid SR-10, or the impregnated charcoal form (Altosid 10-F). Therefore, as a prelude to proposed field evaluations of these compounds, it was essential that dosage response values with *A. triseriatus* be established in the laboratory.

MATERIALS AND METHODS. The *A. triseriatus* mosquitoes utilized in this study have been colonized since 1974 at the US Army Environmental Hygiene Agency, Aberdeen Proving Ground, MD, and were obtained from a parent colony at Rutgers University, New Brunswick, NJ.

Each formulation of methoprene was serially diluted with distilled water so that 1 ml of the serial dilution when pipetted into 249 ml of distilled water resulted in the desired concentration. Untreated controls were maintained simultaneously with the treated mosquitoes.

Twenty, early 4th-instar larvae were placed in 437 ml capacity glass jars containing 250 ml of distilled water, the desired concentration of methoprene, and approximately 64 mg finely ground guinea pig food. No further food was added for the remainder of the test. Prior to use, the glass jars were soaked overnight in a potassium dichromate/sulfuric acid solution to eliminate contamination. Tests were conducted in an environmental chamber maintained at 26.7° C and a photoperiod of 16 hours light: 8 hours dark. All tests were replicated 3 times.

On the 3rd day of each test, a plastic sandwich bag (22 cm L x 17 cm W) was placed over the mouth of each jar and secured with a rubber band so as to contain emerging adults.

All successfully emerged adults, dead larvae, pupae and adults were removed daily and the data recorded. Adults were not categorized as live unless they were found totally emerged and for purposes of this study, adults were considered "normal" only if they were completely free of the exuviae. No attempts were made to determine morphological or physiological anomalies of adult mosquitoes which may have resulted from exposure to methoprene while in the immature stages; however, an evaluation of the mode of action of the developmental inhibitors required examination of the dead pupal forms for characters of pupal developmental stages. Control mortality was divided into groups correlating with these characters, and a percentage of each group was determined using total control mortality as 100 percent. The 6 groups which corresponded chronologically to normal development from pupa formation to emerged adult were unmelanized, newly formed pupa; melanized pupa; adult abdomen separated and retracted from the pupal case; cephalothoracic split present; partially emerged adult, ranging from only the mesonotum protruding through the cephalothoracic split, to the entire body exposed except for the legs and tip of the abdomen which remained within the exuviae; and unsuccessfully emerged adult, ranging from the entire body exposed with one or more legs remaining in the exuviae to completely emerged adults which died on the water surface. The percent mortality for each developmental stage of the mosquitoes subjected to methoprene treatment was compared with those percentages determined for the controls.

RESULTS AND DISCUSSION. The dosage response of *A. triseriatus* to methoprene as determined in laboratory bioassays and using the Bliss method of probit analysis is presented in Table 1. The encapsulated formulation (Altosid® SR-10) required 2.7 times as much active ingredient as the impregnated charcoal formulation to effect 95% mortality.

Hsieh and Steelman (1974) found 90% mortality at 0.6 ppm when treating 3rd instar *A. triseriatus* larvae with a methoprene emulsifiable concentrate. This formulation's rapid degradation (Schaefer et al. 1974), coupled with the time period neces-

Table 1. Dosage response of *Aedes triseriatus* (Say) to Altosid® SR-10 and Altosid® 10-F

Dosage Response	Formulation	
	Altosid SR-10	Altosid 10-F
LC_{50} (ppb)	0.135	0.093
95% confidence limits	0.116–0.157	0.083–0.105
LC_{95} (ppb)	0.971	0.363
95% confidence limits	0.777–1.210	0.239–0.471
Slope	1.9202	2.7863

sary for 3rd instar larvae to reach the susceptible 4th instar stage, necessitates the use of more material at the time of treatment (Schaefer and Wilder, 1972). Formulation and larval instar differences at the time of treatment probably contributed to the dissimilarity of the dosage response values observed between the present study and those reported by Hsieh and Steelman (1974). At the concentration tested, larval mortality in the methoprene-treated jars was less than 1 percent (Table 2), reflecting methoprene's mode of action which is to effect mortality in the pupal stage.

Control mortality was less than 5% in the pupal developmental stage wherein the cephalothoracic split occurred, while the methoprene-treated mosquitoes had over 12% mortality at the same stage of development. A Chi square test indicated these 2 figures were significantly different at the .01 level of probability.

Results of these laboratory tests suggest methoprene-impregnated charcoal would be an effective mosquito control agent. However, due to the rapid degradation of methoprene, this impregnated formulation requires longevity evaluations before its efficacy against natural populations of mosquitoes can be determined.

Literature Cited

Hsieh, M. Y. G. and C. D. Steelman. 1974. Susceptibility of selected mosquito species to five chemicals which inhibit insect development. Mosq. News 34(3):278–282.

Jakob, W. L. 1972. Additional studies with juvenile hormone-type compounds against mosquito larvae. Mosq. News 32(4):592–595.

Schaefer, C. H. and W. H. Wilder. 1972. Insect developmental inhibitors: A practical evaluation as mosquito control agents. J. Econ. Entomol. 65(4):1066–1071.

Schaefer, C. H., W. H. Wilder, F. S. Mulligan, III and E. F. Dupras, Jr. 1974. Insect developmental inhibitors: Effects of Altosid®, TH6040 and H24108 against mosquitoes (Diptera: Culicidae). Calif. Mosquito Cont. Assoc. Proc. 42: 137–139.

Table 2. Effect of Altosid® on *A. triseriatus* exposed as early 4th instar larvae.

Compound Concentration (ppb)	Percent Mortality				
	Larval	Pupal	Pupal-Adult Intermediate	Adult	Total*
Altosid SR-10					
0.10	0	18.15	15.80	14.04	39.78
0.25	0.35	23.70	26.89	24.40	71.40
0.50	0.33	39.33	24.36	22.28	84.14
Altosid 10-F					
0.05	0	15.79	6.89	8.04	19.64
0.10	0.56	29.61	20.74	12.22	56.58
0.25	0.55	47.68	24.04	17.23	87.18

* Adjusted using Abbott's formula (control mortality 13.8%).

Data accumulated from these tests pertaining to the distribution of percent mortality per pupal developmental stage of *A. triseriatus* are beyond the scope of this paper; however, with the exception of one variation, the distribution was very similar.