POLYMER FORMULATIONS OF MOSQUITO LARVICIDES VII. LABORATORY AND FIELD EVALUATION OF ENCAPSULATED FORMULATIONS OF MALATHION AND ABATE®[1]

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ABSTRACT. Laboratory tests were conducted for 8 weeks to determine residue levels of malathion (diethyl mercaptosuccinate, *S*-ester with o,o-dimethyl phosphorodithioate) maintained in water treated with two encapsulated formulations, designated 3MCAP–M (2.0 percent malahtion) and AC–591 (75 percent malathion). The 3MCAP–M was dosed at 2.5 and 5.0 ppm and the AC–591 at 22.0 ppm. The two encapsulated malathion formulations, along with an encapsulated formulation designated 3MCAP–A and containing 1.1 percent Abate® (o,o-dimethyl phosphorothioate o,o-diester with 4,4-thiodiphenyl), were evaluated in artificial field pools at the following dosages: 3MCAP–M at 1.25, 2.5, and 5.0 ppm; AC–591 at 2.2, 22.0 ppm; and 3MCAP–A at 0.25, 0.5, and 1.0 ppm. In the laboratory tests, toxic malathion residues were maintained in the water for 8 weeks by the 3MCAP–M at 5.0 ppm and the AC–591 at 22.0 ppm. In the field tests, none of the encapsulated formulations were effective in providing extended control of mosquito larvae, although the 3MCAP–A gave about 7 weeks of >90 percent control at dosages of 0.5 and 1.0 ppm.

Earlier studies by Wilkinson *et al.* (1971), Roberts and Miller (1971), Miller *et al.* (1973), Roberts *et al.* (1973) and Nelson *et al.* (1973) demonstrated the feasibility of controlling larval mosquito populations through the use of various larvicides incorporated into solid polymers such as polyvinyl chloride, polyethylene, or chlorinated polyethylene. In a continuing effort at this Agency, other techniques have been explored for achieving the slow-release of non-persistent mosquito larvicides into water. This paper reports laboratory and field studies of the larvicidal effectiveness of encapsulated formulations of malathion (diethyl mercaptosuccinate, *S*-ester with o,o-dimethyl phosphorodithioate) and Abate® (o,o-dimethyl phosphorothioate o,o-diester with 4,4′-thiodiphenol).

MATERIALS AND METHODS

ENCAPSULATED FORMULATIONS. The encapsulated formulations evaluated in this study were: 3MCAP-M, containing 2.0 percent malathion; 3MCAP-A, containing 1.1 percent Abate; and AC-591, containing 75 percent malathion. The formulations were provided by the 3M Company, St. Paul, Minnesota (3MCAP-M, 3MCAP-A), and the National Cash Register Company, Dayton, Ohio (AC-591), without additional technical information concerning the chemical nature of the capsule walls. All formulations had a specific gravity >1.0.

LABORATORY TESTS. The 3MCAP-M and AC-591 formulations were evaluated in the laboratory to determine residues which would be maintained in water under static conditions. The 3MCAP-A formulation was not evaluated in the laboratory because the analytical capability available during the study would not have been sufficiently sensitive for the detection of Abate under the conditions of the test. Individual glass jars containing 3 liters of tap water were treated with 3MCAP-M at dosages of 2.5 ppm[4]

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[4] In both laboratory and field tests all references to dosage, whether stated as ppm or kg AI/hectare, are pounds AI/acre-foot equivalents.

and 5.0 ppm, and with AC-591 at a dosage of 22.0 ppm. Controls consisted of jars containing only tap water. Treatments and controls were replicated 3 times. Jars were sealed with screw-cap lids to minimize water loss through evaporation.

Water samples (10 ml) were taken weekly through 8 weeks after treatment. Each 10-ml water sample was extracted by partitioning with Nanograde® hexane. Samples were subjected to three successive partitionings of 10, 5, and 5 ml, respectively. Concentrations of malathion in the combined hexane fractions were determined by flame photometric gas-liquid chromatography without additional cleanup. The percent recovery obtained with the above extraction technique, based on the addition of a known quantity of malathion, was determined to be 100 percent.

A Tracor® Model MT-150 gas chromatograph fitted with a MELPAR flame photometric detector, equipped with 526 mμ interference filter (phosphorus) was used for analysis. The operating parameters were: Oven temperature 200° C, inlet temperature 230° C, outlet temperature 240° C, detector temperature 185° C, attenuation 10^8 x 126, carrier gas N_2, column flow rate 70 cc/min, hydrogen 180 cc/min, oxygen 20 cc/min, air 40 cc/min and chart speed 0.64 cm/min. The column utilized was 182.4 cm x 0.64 cm glass column packed with Chromosorb® W, 80–90 mesh, coated with 3 percent OV–1.

FIELD TESTS. Thirty-two artificial pools, similar to those described by Nelson, *et al.* (1973), were set up in an open field on Aberdeen Proving Ground, Maryland. The 3MCAP-M, AC-591, and 3MCAP-A formulations were applied to 3 replicate pools at each of the 3 dosages shown in Table 1 in a random block design. A 57 percent emulsifiable concentrate of malathion was applied as a water emulsion (WE) to three replicate pools at a dosage of 0.18 ppm. Two untreated pools served as controls. A WE formulation of Abate® was not tested since earlier studies (Roberts and Miller, 1971) had shown this type of formulation to be effective for about 3 weeks. The encapsulated formulations were applied by hand broadcasting pre-measured amounts of the capsules onto the pool surfaces. The WE of malathion was dispersed over the pool surfaces using a pipette.

TABLE 1.—Dosages for encapsulated formulations applied to artificial field pools.

Insecticide dosage (ppm)	Equivalent Kg formulation per hectare	Equivalent Kg insecticide per hectare
	3MCAP–M	
1.25	188.1	3.8
2.5	377.4	7.6
5.0	756.0	14.1
	AC–591	
2.2	8.8	6.6
22.0	88.7	66.5
220.0	887.0	665.2
	3MCAP–A	
0.25	70.5	0.7
0.5	142.2	1.5
1.0	284.4	3.1

In-pool bioassays were conducted weekly with 4th instar laboratory-reared larvae of *Culex pipiens quinquefasciatus* Say using the techniques reported by Miller *et al.* (1973).

Water volumes were calculated and recorded weekly. Water temperatures were measured each week approximately 10 cm below the surface at the center of the pool. Water samples (10-ml) for residue analysis were collected weekly from pools treated with 3MCAP-M, AC-591, malathion WE, and untreated control pools. Collections were taken approximately 10 cm below the surface at the center of the pool and immediately returned for laboratory extraction and analysis as previously outlined. Samples were not collected from pools treated with 3MCAP-A for the reasons discussed earlier.

Although populations of naturally-occurring mosquitoes were not monitored during the entire study, natural larval populations were sampled with an enamel dipper one week before and one week after treatment to determine the immediate effects of the treatments.

RESULTS

LABORATORY TESTS. Malathion residues were maintained during the 8-week test period in laboratory jars treated with the 3MCAP-M and AC-591 formulations. Average 8-week malathion residues were 0.409 ppm and 0.945 ppm for the 3MCAP-M formulation at dosages of 2.5 and 5.0 ppm, respectively. The average 8-week malathion residue for the AC-591 formulation was 1.270 ppm. Malthion residues ranged from 0.291 to 0.548 ppm for 3MCAP-M dosed at 2.5 ppm, from 0.715 to 0.158 ppm for 3MCAP-M at 5.0 ppm, and 0.888 ppm to 1.952 ppm for AC-591. The maximum malathion residues were observed at 4 weeks after treatment for 3MCAP-M at both dosages, and at 2 weeks after treatment for AC-951.

FIELD TESTS. Pretreatment sampling of pools scheduled for treatment with malathion WE showed an average of >25 naturally-occurring *Culex restuans* Theobald larvae per dip. The natural larval population was eliminated by the treatment, and no *C. restuans* larvae were observed through week 1 posttreatment. Pools treated with malathion WE were bioassayed at the time of treatment and 100 percent mortality was observed 24 hours later. Thereafter, no mortality was observed in bioassays of these pools on weeks 1 through 4 posttreatment. Bioassays were terminated after the 4th week. No residue was detected during analysis of water samples collected from the pools treated with malathion WE. Residue analysis was terminated after the 4th week.

Pretreatment sampling of pools scheduled for treatment with 3MCAP-M showed an average of >25 *C. restuans* larvae per dip. The 3MCAP-M, at dosages of 1.25, 2.5, or 5.0 ppm eliminated the natural mosquito larvae population through posttreatment week 1. No significant larval mortality was observed in bioassays of the treated pools during posttreatment weeks 1 through 4 (Table 2). Gas chromatograph analysis showed that residues were below the LC_{90} level during all posttreatment weeks. During the posttreatment period, water temperatures (25.5–27.0 C) remained relatively constant, while water volumes showed a general increase.

Pretreatment sampling of pools scheduled for treatment with AC-591 showed an average of >25 *C. restuans* larvae per dip. The AC-591, at dosages of 2.2, 22.0 or 220.0 ppm, eliminated the natural mosquito larval population through posttreatment week 1. No significant larval mortality was observed in bioassays of the pools treated at 2.2 or 22.0 ppm dosages during posttreatment weeks 1 through 8 (Table 2). In pools dosed at 220.0 ppm larval mortality was <90 percent during all posttreatment weeks, except weeks 1 and 4. Residue levels were below the LC_{90} level during all posttreatment weeks, except weeks 1, 7, and 8 for the 220.0 ppm dosage.

Pretreatment sampling of pools scheduled for treatment with 3MCAP-A showed an average of >25 *C. restuans* larvae per dip. The 3MCAP-A, at dosages of 0.25, 0.5 or 1.0 ppm, eliminated the natural larval population through posttreatment week 1. Bioassays of pools treated with 3MCAP-A at the 0.25 ppm dosage (Table 2) resulted in mortalities <90 percent of all posttreatment weeks, except week 2. In pools treated with 3MCAP-A at a dosage of 0.5 ppm bioassay mortality averaged >90 percent from 1 through 7 weeks after treatment. Thereafter, mortality dropped off and was zero by the 12th week. The 3MCAP-A treatment at 1.0 ppm produced bioassay mortalities >90 percent during posttreatment weeks 1 through 7, and again at week 10. Otherwise mortality was <90 percent and had dropped to zero by week 12.

TABLE 2.—Larvicidal effectiveness observed in artificial field pools treated with encapsulated formulations.[a]

Weeks after treatment	24-hour percent mortality	Insecticide residue (ppm)[b]	Volume of water (liters)	24-hour percent mortality	Insecticide residue (ppm)[b]	Volume of water (liters)	24-hour percent mortality	Insecticide residue (ppm)[b]	Volume of water (liters)	Water temperature (° C)
	AC–591 at 2.2 ppm			AC–591 at 22.0 ppm			AC–591 at 220.0 ppm			
1	0	<0.025	189	3	0.044	189	100	4.818	189	27.6
2	7	<0.025	173	0	<0.025	173	77	0.036	173	26.0
3	0	<0.025	173	3	<0.025	173	67	0.028	173	26.0
4	0	<0.025	158	0	<0.025	158	97	0.046	142	25.5
5	0	<0.025	268	3	<0.025	252	33	<0.025	252	26.0
6	3	<0.025	268	0	<0.025	268	33	<0.025	268	27.0
7	0	<0.025	252	0	<0.025	268	33	0.127	252	24.5
8	0	<0.025	236	0	<0.025	236	40	0.201	236	25.0
	3MCAP–M at 1.25 ppm			3MCAP–M at 2.5 ppm			3MCAP–M at 5.0 ppm			
1	37	<0.025	173	0	<0.025	158	33	<0.025	173	26.0
2	3	<0.025	158	0	<0.025	142	30	<0.025	158	25.5
3	3	<0.025	252	0	<0.025	252	30	<0.025	268	26.0
4	0	<0.025	268	7	<0.025	268	33	<0.025	268	27.0
	3MCAP–A at 0.25 ppm			3MCAP–A at 0.5 ppm			3MCAP–A at 1.0 ppm			
1	57	.....	158	97	.....	142	100	.....	158	26.0
2	100	.....	142	100	.....	126	100	.....	142	25.5
3	73	.....	236	97	.....	236	100	.....	252	26.0
4	50	.....	252	100	.....	252	100	.....	268	27.0
5	80	.....	236	100	.....	236	100	.....	268	24.5
6	73	.....	221	100	.....	221	100	.....	236	25.0
7	37	.....	299	100	.....	299	100	.....	315	24.0
8	13	.....	268	67	.....	284	60	.....	284	25.1
9	17	.....	315	23	.....	299	67	.....	315	22.5
10	0	.....	315	23	.....	315	90	.....	331	27.6
11	0	.....	299	0	.....	299	27	.....	315	17.5
12	3	.....	299	0	.....	299	0	.....	315	19.8

[a] Values for percent mortality, insecticide residue, and water volume represent the average of 3 replicates. Values for water temperature represent the average temperature of six randomly selected pools.

[b] Insecticide residue values represent ppm malathion, residue levels for Abate were not determined.

DISCUSSION AND CONCLUSIONS

The reported LC_{95} level (Anonymous, 1971) for malathion is 0.5 ppm for *Aedes aegypti* (Linnaeus) and *Anopheles quadrimaculatus* Say, and 0.1 ppm for *C. p. quinquefasciatus*. Tests have shown the LC_{90} for this Agency's laboratory colony of *C. p. quinquefasciatus* to be 0.1 ppm malathion. On this basis, the laboratory tests with 3MCAP-M and AC-591 indicated that each formulation was capable of maintaining toxic malathion residues in water for at least 8 weeks, although the 3MCAP-M formulation would be only marginally effective at the lower dosage (2.5 ppm) against *A. aegypti* and *A. quadrimaculatus*.

In the field tests with the 3MCAP-M and AC-591 formulations, all dosages were initially effective in eliminating the natural *C. restuans* population. However, with the exception of the AC-591 which showed some residual effect through week 4 when dosed at 220.0 ppm, all of the formulations and dosages were ineffective in providing control of mosquito larvae beyond that which was achieved with malathion WE. Additionally, the kg of 3MCAP-M or AC-591 required on a hectare basis (Table 1) may make them impractical for use at the dosages tested in this study, or at higher dosages.

In the field tests with 3MCAP-A, the natural *C. restuans* population was initially eliminated at all dosages. At the 0.25 ppm dosage, the 3MCAP-A showed no residual activity at all, while at dosages of 0.5 or 1.0 ppm control was observed for approximately 7 weeks. This did not actually represent significant extended control, since it is possible to achieve 3 weeks of control by treatment with a WE formulation of Abate alone (Roberts and Miller, 1971). Also, from the standpoint of the kg required on a hectare basis, the 3MCAP-A may not be practical for use at the dosages evaluated in this study, or at higher dosages.

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