BIOLOGICAL ACTIVITY OF ORGANOPHOSPHORUS COMPOUNDS AND SYNTHETIC PYRETHROIDS AGAINST IMMATURE MOSQUITOES

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ABSTRACT The synthetic pyrethroids NIA-18739 (5-benzyl-3-furyl) methyl(+)-trans-2,2-dimethyl-3-(2-methyl-1-propenyl) cyclopropane-1-carboxylate), NIA-24110 (RU-11679) (5-benzyl-3-furyl)methyl 1R-2-[(cyclopentylidene)methyl]-3,3-dimethyl-cyclopropanecarboxylate and NIA-26021 (*cis* component of SBP-1382) at the rate of 0.01 ppm produced complete mortality of 4th-stage larvae of *Aedes aegypti* Linnaeus, *Anopheles albimanus* Wiedemann and *Culex p. quinquefasciatus* Say in the laboratory within an hour after treatment. Organophosphorus larvicides which displayed high biological activity against 4th-stage larvae of these species at the rate of 0.05 ppm or less were Bay SRA-7660, N-2596 (*O*,ethyl-S-(p-chlorophenyl ethane phosphonodithioate), M-3196 (*O,O*-dimethyl *O*-3,5,6-trichloro-2-pyridylphosphorothioate, Cidial (ethyl mercaptophenyl-acetate, *O,O*-dimethyl phosphorodithioate) and Bay HOX-1619.

Larvae and pupae of *Culex tarsalis* Coquillett, *C. peus* Speiser and *Culiseta inornata* Williston in field breeding ponds were controlled by the pyrethroids applied at the rate of 0.25 lb/A AI. N-2596 and Bay ZUM-0677 gave complete control of larvae in these ponds at the rates of 0.01 and 0.025 lb/A respectively.

Mosquito resistance to chlorinated hydrocarbons and organophosphorus larvicides is widespread in California (Womeldorf *et al* 1972). In terms of mosquito resistance to larvicides, the central and southern San Joaquin Valley has experienced this problem for many years. In Merced, Fresno, Tulare, and Kern counties, conventional insecticides in some areas are no longer effective against *Aedes nigromaculis* (Ludlow) larvae in irrigated pastures. In these areas mosquitoes are being controlled with petroleum oils such as Flit MLO®, ARCO larvicidal oil or other oil formulations in the aquatic stages, or by the use of propoxur (Baygon®) as an adulticide (Darwazeh and Ramke 1972).

Mulla (1970) and Schaefer (1972) considered the mosquito problem rather critical and pointed out the need for continued research to develop alternate measures. Mulla (1970) emphasized the need for the development of new materials which have high biological activity against mosquito larvae and possess a different mode of action from that of the chlorinated hydrocarbons, organophosphates, and carbamates. Such materials include petroleum oils, aliphatic amines, insect growth regulators, and compounds derived from biological agents such as algal toxins. Schaefer (1972) also expressed the need for the development of new, safe, and highly effective materials, and suggested implementation of alternate measures to deal with the resistance problem in mosquitoes.

The current studies were initiated to find and evaluate new and safe materials with either conventional or different mode of action, but having high biological activity against immature mosquitoes.

METHODS AND MATERIALS. A number of new materials were evaluated in the laboratory against 4th-stage *Culex pipiens quinquefasciatus* Say. Those materials which showed considerable activity against larvae and pupae of this mosquito were evaluated further against 4th-stage *Anopheles albimanus* Wiedemann and *Aedes aegypti* Linnaeus.

Laboratory larval bioassay techniques are described elsewhere (Mulla *et al.* 1966). In brief, these techniques are as follows: One percent stock solutions (W/V) in acetone were prepared from technical grade materials, and serial dilutions were prepared as needed. Aliquots of 1 ml or less of the proper strength solution were added to 100 ml tap water in 4-oz waxed paper cups provided with 20 4th-stage larvae or pupae. Each concentration was run in duplicate, and each material was tested 2-3 times. After 24 hours of exposure, mortality readings were taken and LC_{50} and LC_{90} estimates in ppm were obtained from dosage response lines on a probit log paper based on mean mortality values versus concentrations. Larvae and pupae utilized were obtained from colonies maintained in the laboratory.

The most promising materials were then selected and evaluated under field conditions in experimental ponds in Oasis (Coachella Valley of southern California) and at Midgeville (Aquatic Research Facility, University of California, Riverside). Detailed descriptions of these facilities are given elsewhere (Mulla *et al.* 1970). Mosquito population in the Coachella Valley consisted of all aquatic stages of *Culex tarsalis* Coquillett and *Culiseta inornata* Williston. *Culex peus* Speiser larvae were predominant in ponds in Midgeville (Riverside).

Materials evaluated in the experimental ponds were either wettable powders or emulsifiable concentrates. The required amount of the EC or WP were mixed with 80 ml of water and applied with an all purpose 1-qt household sprayer (Smart and Final Iris Co., Los Angeles, Calif. 90058). For assessment, five dips were taken per pond prior to and 48 hrs after treatment. Each concentration was replicated twice and in every test two ponds were left untreated as check. Chemical description of the materials tested is as follows:

NIA-26021: (NRDC 119 is the *cis* component of NIA-17370 or resmethrin or SBP-1382).

NIA-18739: (NRDC 107 or bioresmethrin)-(5-benzyl-3-furyl)methyl(+)-trans-2,2-dimethyl-3-(2-methyl-1-propenyl)cyclopropane-1-carboxylate.

NIA-24110(RU-11679): (-5-benzyl-3-furyl)methyl 1R,2R-2-[(cyclopentyl-idene) methyl]-3,3-dimethyl-cyclopropanecarboxylate.

Me-Dursban (M-3196) *O,O*-dimethyl *O*-3,5,6-trichloro-2-pyridyl phosphorothioate

Cidial: ethyl mercaptophenylacetate, *O,O*-dimethyl phosphorodithioate

N-2596: *O*,ethyl-*S*-(*p*-chlorophenyl ethane phosphonodithioate)

Dasanit: *O,O*-diethyl *O*[*p*-(methylsulfinyl)phenyl] phosphorodithioate

Bux: *m*-(1-ethylpropyl)phenyl methylcarbamate and *m* (1-methylbutyl)phenyl methylcarbamate

Dyfonate: *O*-ethyl-*S*-phenylethyl-phosphonodithioate

RESULTS AND DISCUSSION. The synthetic pyrethroids NIA-26021, NIA-18739, and NIA-24110 (RU-11679) were highly active in the laboratory against 4th stage larvae. In general, *C. p. quinquefasciatus* was more susceptible than *Ae. albimanus* (Table 1). These compounds were quick acting against all 3 species. At the LC_{90} level, larval mortality occurred within an hour after treatment.

Other fast acting and equally effective chemicals at the rate of 0.05 ppm or less included Bay SRA-7660, Bay HOX-1619, and Cidial. Me-Dursban and N-2596 were also highly effective against all species, but larval mortality occurred slowly, reaching maximum within 24 hours after treatment. Preliminary observations showed that several of these compounds manifested activity against pupae. This feature is an additional bonus to larvicidal activity and, therefore, some of these were evaluated against pupae.

NIA-26021 showed very high activity against pupae, producing complete mortality of pupae of *C. p. quinquefasciatus* at the rate of 0.003 ppm (Table 2). Other fast acting and effective materials against pupae at the rate of 0.05 ppm or less include NIA-18739, NIA-24110, Bay KUE-2302, and Bay ZUM-0676 (Table 2).

In field evaluation the synthetic pyrethroids NIA-24110, NIA-26021, NIA-18739, and RU-11679 suppressed larvae and pupae of *C. tarsalis, C. peus,* and *Culiseta inornata* at the rate of 0.25-0.50 lb/acre applied to experimental ponds (Table 3).

Field efficacy of these pyrethroids is not commensurate with the activity found in

TABLE 1. Activity of various mosquito larvicides (in ppm) against 4th-stage larvae of mosquitoes.

Material	AN. ALBIMANUS		AE. AEGYPTI		C.P. QUINQUE-FASCIATUS	
	LC_{50}	LC_{90}	LC_{50}	LC_{90}	LC_{50}	LC_{90}
NIA-26021	0.003	0.006	0.002	0.004	0.0007	0.001
NIA-18739	0.003	0.006	0.003	0.009	0.002	0.003
NIA-24110[a]	0.003	0.009	0.002	0.004	0.003	0.005
Me-Dursban	0.003	0.006	0.003	0.005	0.001	0.002
Cidial	0.003	0.007	0.012	0.025	0.004	0.008
N-2596	0.005	0.012	0.006	0.01	0.003	0.007
Bay SRA-7660	0.003	0.01	0.006	0.012	0.003	0.005
Bay KUE-2302	0.006	0.014	0.025	0.065	0.005	0.009
Bay HOX-1619	0.007	0.017	0.025	0.050	0.005	0.008

[a]Same compound as RU-11679.

TABLE 2. Laboratory evaluation of mosquito larvicides against pupae of *Culex p. quinquefasciatus*.

Material	PPM	
	LC_{50}	LC_{90}
NIA-26021	0.0012	0.0025
NIA-18739	0.0120	0.025
NIA-24110	0.0140	0.030
BAY KUE-2302	0.0140	0.030
BAY ZUM-0676	0.0140	0.030
BAY ZUM-0677	0.075	0.100
BAY KUE-2327	>0.100	—
BAY SRA-7660	>0.100	—
BAY ZUM-0673	>0.100	—
BAY MEB-6046	>0.100	—
BAY HOX-1619	>0.100	—

laboratory. Based on experience and information on other larvicides, the pyrethroids were expected to yield field control at rates of 0.025-0.1 lb/acre. In view of the very high activity in laboratory but no or little activity in the field, it appears that degradation of the compounds occurs at a rapid rate. Development of stable formulations might improve the field activity spectrum of these novel compounds.

In Oasis ponds, N-2596 gave 100% control of the two species *C. tarsalis* and *Culiseta inornata* for more than 5 days at the rate of 0.01 lb/acre (Table 4). Bay ZUM-0677 and Bay MEB-6046 gave complete control of larvae at the rates of 0.01 and 0.05 lb/acre respectively, the former material being more effective and long lasting. Good results (87-95 percent control) were obtained with Bay KUE-2302 and Bay SRA-7660 at the rate of 0.05 lb/acre. At the same rate, poor control was obtained with Bay ZUM-0673.

In field ponds at Midgeville, excellent larval control was obtained with N-2596, Bay ZUM-0676, and Dasanit at the rate of 0.025, 0.05, and 0.5 lb/acre respectively (Table 5). The former material was slightly less effective here than in the Oasis ponds. Dyfonate EC-4 produced complete reduction in the larval population at the rate of 0.3 lb/acre, while Dyfonate flowable (F4) failed to control mosquito larvae adequately at the same rate. At 0.5 lb/acre, only 70 percent mortality occurred with Bux.From these studies, it is apparent that some of the compounds evaluated may be suitable for use as mosquito larvicides and pupicides. Further studies on formulation and efficacy will likely improve the efficiency of some of these compounds.

TABLE 3. Field evaluation of synthetic pyrethroids against *Culex tarsalis* and *C. peus* in experimental ponds (March 1973).

Chemical and formulation	Dosage lb/acre	No. of larvae and pupae/5 dips[a] Pretreatment L	Pretreatment P	Post-treatment L	Post-treatment P	(%) reduction[c]
NIA-18379 EC 1	0.05	94	0	23	0	76
	0.10	19	19	6	1	82
	0.25	54	4	0	2	97
	0.05	34	4	0	0	100
NIA-24110 EC 1	0.05	20	9	67	13	0
	0.10	65	10	23	6	61
	0.25	45	6	0	0	100
	0.50	70	26	3	7	90
NIA-26021 EC 1	0.05	18	8	13	11	8
	0.10	81	0	6	0	93
	0.25	43	6	0	2	96
	0.5	28	7	0	0	100
Check	—	6	8	8	7	0
RU-11679 EC 1[b]	0.05	41	0	21	1	46
	0.10	93	0	14	2	83
	0.25	69	0	0	0	100
Check	—	26	0	33	2	0

[a]L = 3rd- and 4th-stage larvae.
P = pupae.
[b]Same as NIA-24110, except from a different supplier. Evaluated in ponds at the Aquatic Research Facility. All others evaluated in Oasis ponds.
[c](%) reduction based on larvae and pupae count.

TABLE 4. Field evaluation of mosquito larvicides against *Culex tarsalis* and *Culiseta inornata* in field experimental ponds (Oasis, January 1973).

Material and formulation	Dosage lb/acre	Pretreat (days)	Avg. no. larvae/dip aft. treat. (days) No.	% R[a]	No.	% R[a]
N-2596 EC 4	0.01	7	0	100	0	100
	0.05	11	0	100	0	100
Bay KUE-2302 EC 4	0.01	6	5	17	7	0
	0.05	12	1	95	2	83
Bay SRA-7660 EC 2	0.01	12	6	50	14	0
	0.05	15	2	87	2	87
Check	—	13	15	0	18	0
Bay ZUM-0677 EC 4	0.025	10	0.6	94	0	100
	0.05	8	0	100	0	100
Bay MEB-6046 WP 75	0.025	15	12	20	13	13
	0.050	6	0	100	5	17
Bay ZUM-0673 WP 75	0.025	7	5	30	3	57
	0.050	9	12	0	7	22
Check	—	4	6	0	4	0

[a]% R is based on 3rd- and 4th-stage larval count.

TABLE 5. Field evaluation of various mosquito larvicides against larvae and pupae of *Culex tarsalis* in experimental ponds (Midgeville, June-July 1973).[a]

Chemical and formulation	Dosage lb/acre	No. of larvae and pupae/4 dips Pretreatment L	P	Post-treatment L	P	(%) reduction[b]
Bay ZUM-0676 EC 4	0.01	72	3	33	7	54
	0.025	53	17	8	6	85
	0.05	74	5	0	0	100
N-2596 EC 4	0.01	8	3	1	0	87
	0.025	10	3	0	5	100
Dasanit EC 6	0.25	35	4	4	3	89
	0.50	15	8	0	3	100
Bux EC 2	0.10	108	10	155	23	0
	0.25	29	2	13	0	55
	0.50	50	7	15	11	70
Dyfonate EC 4	0.20	22	6	10	0	55
	0.30	65	4	0	0	100
Dyfonate F 4	0.20	12	14	11	2	8
	0.30	42	5	20	5	53
Check	–	16	0	37	2	0

[a] Very few *C. peus* were found during this experiment.
[b] % reduction is based on 3rd- and 4th-stage larval counts.

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