TOXICITY OF NEW ORGANIC INSECTICIDES TO MOSQUITO FISH AND SOME OTHER AQUATIC ORGANISMS [1]

MIR S. MULLA [2]

Department of Entomology, University of California, Riverside, California

Due to the development of resistance in mosquitoes to a variety of chemical larvicides, new and promising materials are explored and evaluated systematically for biological activity against mosquito larvae. During the course of this developmental research program, most of the materials evaluated against mosquito larvae are also subjected to field trials for an assessment of their biological activity against the mosquito fish *Gambusia affinis* (Baird and Girard) and other aquatic organisms which are encountered while the tests are in progress. This paper presents the results of these trials with several new organic insecticides.

METHODS AND MATERIALS. Field evaluation trials were conducted in the same manner as in earlier studies (Mulla 1963, Mulla and Isaak 1961, Mulla *et al.*, 1963). Field ponds 1/16 acre in size were filled with water to a depth of 10–12″. Emulsifiable concentrate formulations were mostly used for the preparation of dilute aqueous sprays. Wettable and soluble powder formulations of some materials were also used.

The sprays were applied at the rate of 8 gallons per acre by means of 1- or 3-gallon hand-sprayers. Fish (50 per pond, 25 per cage) and tadpoles of *Rana catesbeiana* (10 per pond) were confined in screen cages placed in the ponds immediately after treatment. Mortality was assessed 24 hours after exposure. If fish mortality in any cage was more than 4 percent, fresh fish were provided at the time of reading and the mortality was again assessed 24 hours later.

Observations on the mortality of organisms such as diving beetles and others present in the ponds were also made. These animals were not sampled for population density. Therefore, the notes presenting the results of these observations are not definitive in nature, but only provide preliminary data on the response of these organisms to the treatments.

Most of the materials evaluated for biological activity against *Gambusia affinis* have also been studied for potency against mosquito larvae (Mulla *et al.*, 1964, 1965). When the biological activity of the experimental compounds against mosquito larvae, fish, and other aquatic organisms is known, one is in a better position to select the most promising material for mosquito larval control.

Chemical descriptions of the experimental compounds field-tested against fish and other organisms are given in Table 1.

RESULTS AND DISCUSSION. The acute biological activity of nine materials against *Gambusia affinis* is shown in Table 2. AC-47921 indicated a high level of toxicity at the rates of 0.1 and 0.4 lb/acre. At a lower rate (0.05 lb/acre), this material showed a low order of fish toxicity. UC-8305 used at a single rate (0.5 lb/acre) was highly toxic. Bayer 73, at the two higher rates (1 and 2 lb/acre) was quite toxic, while at the lower rate (0.5 lb/acre) showed no appreciable level of toxicity.

Isolan and Ortho 5353, both carbamates, were toxic to the fish at least at the higher rates. N-2404 was quite toxic at the rates of 0.1 and 0.5 lb/acre. At the higher rate, this material produced appreciable kill of invertebrate non-target species. Ortho

[1] Paper from the University of California Citrus Research Center and Agricultural Experiment Station, Riverside, California.

These studies were conducted in cooperation with the Kern Mosquito Abatement District, Bakersfield, California.

[2] The able assistance of Gerrit Kats of the University of California Citrus Research Center and Agricultural Experiment Station, Riverside, during the course of these studies is acknowledged.

TABLE 1.—Chemical description of experimental compounds.

Compound	Chemical description
AC-38023 (Famophos®)	*O,O*-dimethyl *O-p*-(dimethylsulfamoyl) phenylphosphorothioate
AC-43064	2-(diethoxyphosphinothioylimino)-1,3-dithiolane
AC-47031	2-(diethoxyphosphinylimino)-1,3-dithiolane
AC-52160 (Abate)	*O,O,O'*-tetramethyl *O,O'*-thiodi-*p*-phenylene phosphorothioate
Bayer 73	2-aminoethanol salt of 2',5-dichloro-4'-nitrosalicylanilide
Bayer 44646	3-methyl-4-dimethylaminophenyl N-methylcarbamate
Bayer 46676	*O*-ethyl *O*-2-ethylthio-4-methyl-6-pyrimidyl ethylphosphonothioate
Bayer 47940	*O,O*-dimethyl O-(3-chloro-4-cyanophenyl)-thionophosphate
Bayer 52957	*O,O*-diethyl O-(5-chlorobenzisoxazolyl-(3)-phosphorothioate
Bomyl® (GC-3707)	Dimethyl 1,3-(dicarbomethoxy)-1-propen-2-ylphosphate
Cela S-1942	*O-O*-dimethyl O-(2,5-dichloro-4-bromophenyl) thionophosphate
Cela S-2225	*O-O*-diethyl *O*-(2,5-dichloro-4-bromophenyl) thionophosphate
Dimetilan	2-dimethylcarbamyl-3-methylpyrazolyl-(5)-dimethyl-carbamate
GC-9160	Delta-(5-hydroxy-1,2,3,4,6,7,8,9,10,10-decachloro-pentacyclo [$5.3.0^{2,6}.0^{3,9}.0^{4,8}.$]decyl) ethyl levulinate
GC-9879	Alpha (diethoxyphosphinothioylthio) gamma-butyrolactone
GS-12968	*O,O*-dimethyl-S-[5-ethoxy-1,3,4-thiadiazol-2-(3H)-on-3-yl-methyl]-dithiophosphate
GS-13005	*O,O*-dimethyl-S-[5-methoxy-1,3,4-thiadiazol-2(3H)-on-3-yl-methyl]-dithiophosphate
Isolan	Dimethyl 5-(1-isopropyl-3-methylpyrazolyl) carbamate
N-2404	*O*-isopropyl *O*-(2 chloro-4-nitrophenyl)-ethylphosphonothioate
N-2788	*O*-ethyl *S-p*-tolyl-ethylphosphonodithioate
N-2790	*O*-ethyl-*S*-phenyl-ethylphosphonodithioate
Ortho 5305	3-*sec*-butylphenyl-N-methyl carbamate
Ortho 5353	3-*sec*-amylphenyl N-methylcarbamate
Ortho 5655	3-*sec*-butyl 6-chlorophenyl N-methylcarbamate
Pyramat	2-*n*-propyl-4-methylpyrimidyl (6) dimethylcarbamate
SD-7727	Methanesulfonic acid,2,4-dichlorophenyl ester
SD-8211	Phosphoric acid,2-chloro-1-(2,5-dichlorophenyl) vinyl dimethyl ester
SD-8447	Phosphoric acid,2-chloro-1-(2,4,5-trichlorophenyl) vinyl dimethyl ester
SD-8530	Carbamic acid,methyl-3,4,5-trimethyl phenyl ester
SD-8803	Phosphorothioic acid,*O*-(2-chloro-1-(2,4-dichlorophenyl) vinyl)*O,O*-diethyl ester
SD-9020	Phosphorothioic acid,*O*-(2-chloro-1-(2,4-dichlorophenyl) vinyl)*O,O*-dimethyl ester
SD-9129	3-(dimethoxyphosphinyloxy)-N-methylcrotonamide
UC-8305	P-chloro-2,4-dioxa-5-methyl-P-thiono-3-phosphabicyclo (4.4.0) decane
UC-19786	2-*sec.* butyl 4,6-dinitrophenyl isopropylcarbonate
UC-20047	3-chloro-6-cyano-2 norbornanone *O*(-methyl-carbamoyl) oxime
UC-21149	2-methyl-2-(methylthio) propionaldehyde *O*-methylcarbamoyl) oxime

5305, another carbamate material, was also quite toxic to the fish. GC-9160, an organochlorine compound, showed a moderate degree of toxicity to the mosquito fish. This material, however, showing appreciable activity against mosquito larvae in the field would control mosquitoes at a rate of about 0.25 lb/acre.

Data on the biological activity of another group of six insecticides are presented in Table 3. All the compounds listed in this table showed moderate to high level of toxicity to the mosquito fish at the rates tested. Among the compounds listed, GS-13005 has been recently evaluated against mosquito larvae, and it shows a high level of activity (Mulla *et al.*, 1966). At mosquito larvicidal rate (0.1 lb/acre), this compound manifested a high level of toxicity to the mosquito fish.

The toxicity of several materials to the mosquito fish and some invertebrate nontarget species is presented in Table 4. With the exception of SD-9129, all materials listed in this table were found to be toxic to the fish at the higher rates. Both

SD-8803 and SD-9020, although highly effective against mosquito larvae (Mulla *et al.*, 1966), showed little toxicity at the larvicidal rate (0.1 lb./acre) to the fish. They both were relatively harmless to some of the invertebrate non-target species at the larvicidal rates.

The following compounds used at the maximum rates (in parentheses in lb/acre following each compound), were found to be relatively harmless to the mosquito fish under the test conditions:

Bayer 44646 (0.4), Bayer 47940 (0.4), GC-9879 (2.0). Small tadpoles of *Bufo boreas* and tadpole shrimps survived these treatments.

AC-43064 (2.0), Cela S-1942 (1.0), Cela S-2225 (1.0), and Ortho 5655 (1.0), Pyramat (2.0), Dimetilan (2.0), SD-7727 (2.0), AC-38023 (0.3) and UC-20047 (1.0).

Bayer 52957 (0.4), SD-8211 (0.4), SD-8447 (0.4), SD-8530 (0.4) and UC-21149 (1.0). Also, these materials did not show toxicity to tadpoles of *Bufo boreas,* tadpole shrimps, and diving beetle larvae and adults.

Abate or AC-52160 (0.4), CL-43913 (0.4) and UC-19786 (1.0). These three materials, in addition to being non-toxic to the fish, were also harmless to the tadpoles of *Rana catesbeiana* which were ex-

TABLE 2.—Toxicity of new insecticides to the mosquito fish *Gambusia affinis* in field ponds.

Toxicant & formulation	Dosage lb./acre	Av. 24-hr. percent mortality days after treatment			
		1	2	3	4
AC-47921 EC4	0.05	18	14	10	0
	0.1	100	70	32	0
	0.4	100	100	100	100[1]
UC-8305 EC4	0.5	100	100	100	18
Bomyl EC4 (GC-3707)	0.5	4	0	0	0
	2.0	66	8	0	0
Bayer 73 WP 71 percent	0.5	12	6	0	2
	1.0	100	20	0	10
	2.0	100	100	100	12
Control	..	6	4	2	0
Isolan EC2	0.55	10	2	0	0
	2.00	100	100	100	42
Ortho 5353 EC2	0.2	62	0	0	0
	1.0	100	0	0	0
N-2404 EC4	0.05	26	4	0	..
	0.1	100	0	2	0
	0.5[2]	96	6	0	0
Control	..	20	2	0	0
GC-9160 EC2	0.5[3]	30	4	0	2
	2.0[3]	62	10	0	6
Ortho 5305 EC2	0.1	10	5	..	2
	0.2	88	100	30	14
	0.8	100	100	100	100[4]
Control	..	4	2	0	0

[1] 20 percent mortality 5 days after treatment.

[2] Appreciable kill of diving beetle larvae and adults, chironomid larvae and dragonfly naiads; mortality of dragonfly naiads spectacular.

[3] Tadpole shrimps survived these treatments.

[4] Average 24-hr mortality was 100 percent and 34 percent, 8 days and 9 days after treatment respectively.

TABLE 3.—Toxicity of new insecticides to the mosquito fish *Gambusia affinis* and to the tadpoles of *Rana catesbeiana* in field ponds.

Toxicant & formulation	Dosage lb./acre	Av. 24-hr. percent mortality of *G. affinis* days after treatment						
		1	2	3	7	8	9	10
N-2790 [2] EC4	0.1	28	22	..	6	..	..	..
	0.2	64	62	82	62	50	8	..
	1.0	100	100	100	100	100	100	100[3]
GS-13005 [2] EC4	0.1	60	64	88	92	28	12	22
	0.4	100	100	100	100	100	100	100
AC-47921 [2] EC4	0.1	30	0	22	8	..	..	..
	0.4	96	94	100	48	34	28	..
UC-21427 [2] EC2	0.5	100	82	0	0	..	..	..
	1.0	100	0	0	..	..	..	..
Bayer 46676 EC2	0.2	100	100	100	..	..	48	..
	1.0	100	100	100	..	..	52	..
AC-47031 EC4	0.5	100	100	80	..	..	10	..
	2.5	100	100	52	..	..	8	..
Control	..	0	0	6	8	6	15	..

[1] Small tadpoles of *Bufo boreas* survived all these treatments.

[2] No mortality in tadpoles of *Rana catesbeiana* during an exposure period of one week. Tadpoles were not exposed in ponds treated with Bayer 46676 and AC-47031.

[3] This treatment gave 80 percent and 30 percent mortality 15 and 16 days after treatment. No kill was obtained in fish placed 20 days after treatment. About 10 percent tadpole kill was also observed in this treatment.

TABLE 4.—Toxicity of new insecticides to the mosquito fish *Gambusia affinis* in field ponds.

Toxicant & formulation	Dosage lb./acre	Av. 24-hr. percent mortality days after treatment			Other organisms surviving [3]
		1	2	6	
N-2790 EC4	0.1	28	22	6	T, TP alive
	0.2	100	92	18[1]	T, TP alive DBL dead
N-2788 EC4	0.1	100	14	0	T, TP alive
	0.2	100	100	2	T, TP alive
	1.0	100	100	100[2]	..
GS-12968 EC4	0.1	0	14	0	T, TP alive
	0.4	72	56	0	T, TP alive
SD-8803 EC2	0.1	18	8	0	T, TP
	0.4	100	36	0	DBA alive TP, DBA dead
SD-9020 EC2	0.1	20	6	0	T, DBA alive
	0.4	56	18	4	DBA dead
SD-9129 EC3	0.2	14	10	4	TPS alive
	0.8	16	14	4	..
Control	..	8	4	8	..

[1] Percent mortality after 7 and 8 days was 18 percent and 4 percent respectively.

[2] Mortality was 100 percent and 85 percent, 10 days and 14 days after treatment respectively.

[3] DBA=diving beetle adults; DBL=diving beetle larvae; DFL=dragonfly naiads; MFL=mayfly naiads; T=juvenile toads (*Bufo boreas*); TP=tadpoles (*Bufo boreas*) and TPS=tadpole shrimp.

posed to these chemicals for one week. Abate and CL-43913 are outstanding larvicides (Mulla *et al.*, 1964), which showed no toxicity to non-target aquatic organisms. Most of the materials listed above showed no marked acute toxicity against some selected invertebrate non-target organisms. However, these organisms were not present in noticeable numbers in all of the treatments. Lack of positive evidence, therefore, does not imply that the materials for which no observations have been recorded are either toxic or safe to these non-target organisms.

References Cited

MULLA, M. S. 1963. Toxicity of organochlorine insecticides to the mosquito fish *Gambusia affinis* and the bullfrog *Rana catesbeiana.* Mosquito News. 23(4):299–303.

———, and ISAAK, L. W. 1961. Field studies on the toxicity of insecticides to the mosquito fish *Gambusia affinis.* Jour. Econ. Ent. 54(6):1237–42.

———, ———, and AXELROD, H. 1963. Field studies on the effects of insecticides on some aquatic wildlife species. Jour. Econ. Ent. 56(2):184–8.

———, METCALF, R. L., and KATS, G. 1964. Evaluation of new mosquito larvicides, with notes on resistant strains. Mosquito News 24(3):312–9.