IMPACT OF EXPERIMENTAL INSECT GROWTH REGULATORS ON SOME NONTARGET AQUATIC INVERTEBRATES[1]

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ABSTRACT. The adverse effects of 3 IGRs, 25% WP of diflubenzuron, 25% WP and 0.5% G of Bay SIR-8514, and EC-4 of MV-678, when evaluated for chironomid control in a few experimental ponds, a sewage polishing pond, and a natural pond, were studied on the aquatic nontarget invertebrates.

The WP and the G of SIR-8514 at 56 and 112 g AI/ha adversely affected *Cyclops* spp., Collembola, *Chaoborus* larvae, nymphs of *Baetis* sp., notonectids and corixids, and larvae of Coleoptera in the experimental ponds. Diflubenzuron at 28 and 56 g AI/ha reduced *Cyclops* spp., Collembola, *Chaoborus* sp. and *Baetis* sp. in these ponds but *Cyclops* spp. and Coleoptera showed more sensitivity to SIR-8514. The EC of MV-678 at 56 and 112 g AI/ha proved the least harmful of the 3 IGRs to the various invertebrates.

In the sewage pond, SIR-8514 (WP) at 70 g AI/ha adversely affected *Cyclops* spp. and *Hyalella azteca* (Saussure). *Cypridopsis* sp. and Oligochaeta were not affected.

The EC of MV-678 at 0.22 kg AI/ha had no significant (P > 0.05) adverse effects on *Diaphanosoma brachyurum* (Liéven), *Bosmina coregoni* Baird, *Ceriodaphnia* sp., *Diaptomus* spp., Hydrachnellae, Hirudinea, and Oligochaeta in the natural pond.

INTRODUCTION

Chironomid midges emerge at nuisance levels from a variety of urban and suburban aquatic habitats, such as water percolation basins, flood control channels, sewage oxidation ponds, and manmade and natural lakes. In the United States, the midge problem has been reported from a number of states but during the past 2 decades, major investigations in midge control have been conducted in California and Florida.

To control chironomids, the periodic addition of insecticides to midge breeding sources is the most effective and widely used method. However, the chemical applications to the aquatic habitats often have undesirable effects on some nontarget aquatic invertebrates (Ali and Mulla 1978, Apperson et al. 1978, McAlonan et al. 1976, Muirhead-Thompson 1971). The adverse effects of biological and chemical mosquito control agents on nontarget biota in mosquito habitats were recently reviewed by Mulla et al. (1978).

The present study was made to elucidate the adverse effects of 3 experimental IGRs (evaluated for chironomid control) on the principal nontarget planktonic, nektonic, and benthic invertebrates in a few experimental ponds, a sewage polishing pond, and a natural pond in central Florida.

MATERIALS AND METHODS

STUDY AREAS. *Experimental Ponds.* The 18 earthen ponds at Sanford, FL, were described by Ali and Lord (1980). The water supply in these 45–50 cm deep ponds is from an underground artesian source.

Sewage Polishing Ponds. These 2 ponds (North Pond and South Pond) located in Sanford, FL, have also been previously

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[1] Florida Agricultural Experiment Stations Journal Series No. 2001.

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described (Ali and Lord 1980). The North Pond measured 0.8 ha at the surface and had an average water depth of 1.25 m, while the South Pond was 0.6 ha, containing 1.0 m deep water.

Natural Pond. This rectangle-shaped pond is located on the property of General Electric Company, Daytona Beach, FL. It had a surface area of 0.4 ha and an average water depth of 1.8 m.

IGR TREATMENTS. On April 27, 1979, a 25% wettable powder (WP) and a 0.5% granular (G) formulation of Bay SIR-8514 [1-(4-trifluoromethoxyphenyl)-3-(2-chlorobenzoyl)-urea] were applied to the experimental ponds in a completely randomized design. Three rates, 28, 56, and 112 g AI/ha of the WP were employed while the G formulation was applied at the last 2 rates only (Ali and Lord 1980). In this test, 3 replicates per treatment were used and 3 untreated ponds served as checks.

On July 3, 1979, 25% WP of diflubenzuron [1-(4-chlorophenyl)-3-(2,6-difluorobenzoyl)-urea], 25% WP of SIR-8514, and emulsifiable concentrate (EC-4) of Stauffer MV-678 [2methoxy-9-(4-isopropylphenyl)-2,6-dimethylnonane] were evaluated. Diflubenzuron and SIR-8514 were applied at 28 and 56 g AI/ha and MV-678 was applied at 56 and 112 g AI/ha, while 3 untreated ponds served as checks (Ali and Lord 1980).

The 25% WP of SIR-8514 was also applied at 70 g AI/ha to the North Pond on July 10, 1979, while the South Pond was left untreated. The natural pond was treated with the EC of MV-678 at 0.22 kg AI/ha on August 2, 1979.

SAMPLING. Immediately prior to and at intervals after treatments, the planktonic, nektonic, and benthic invertebrates were sampled. In the experimental ponds, a 50 cm long nylon net (20 cm diameter mouth and 125 μm pore size) mounted on a metal sled was used (Hurlbert et al. 1970). To obtain a sample, the sled was released from one end of the pond (lengthwise) and was gently pulled by rope through the water column from the other end of the pond. In this manner, 2 plankton samples were taken from either side along the length of the pond. The material collected in the net was completely transferred into suitably-labeled bottles and preserved in formaldehyde.

The nektonic organisms in the experimental ponds were sampled by a 400 ml dipper. Five dip samples were taken from each pond (Fanara and Mulla 1974) and were composited and filtered through a 125 μm Nitex® screen. The residue on the screen was transferred into suitably-labeled bottles to which formaldehyde was added. For benthic sampling, 2 mud samples were randomly taken from each pond by using a 15 x 15 cm scoop sampler and the invertebrates were separated from the mud (Mulla et al. 1971).

The zooplankton and nektonic invertebrates in the 2 sewage ponds and the natural pond were sampled by towing the net behind a motor boat. On each visit, 4 zooplankton samples (also containing nektonic invertebrates) were obtained from 4 randomly predetermined locations (2 along the length and 2 across the width) in each pond. The 2 parallel sampling lines chosen along the length as well as across the width in each pond were at least 5 m away from the shoreline and 10–20 m apart from each other. The net remaining submerged within the top ½ m column of water was towed for the same distance along each sampling line on each occasion. The collected organisms were transferred into suitably-labeled bottles and preserved. The benthic samples from the 2 sewage ponds and the natural pond were obtained with a 15 × 15 cm Ekman dredge. Eight dredge samples were randomly collected, and the invertebrates were washed and segregated (Mulla et al. 1971).

In the laboratory, the zooplankton and nektonic samples were processed (Ali and Mulla 1978). The zooplankton and other invertebrates were identified (Edmondson 1963) and counted, and the mud samples were also analyzed for midge larvae.

In the natural pond (treated with MV-

678), it was not possible to maintain a separate check area within the same pond; therefore, the pre-, and posttreatment data were compared. Also, the qualitative and quantitative compositions of the invertebrate fauna in the 2 sewage polishing ponds were not comparable; thus, the pre-, and posttreatment invertebrate populations in the North Pond only were compared with each other to elucidate the effects of SIR-8514. The posttreatment reductions of the nontarget invertebrates in the experimental ponds were calculated (Ali and Lord 1980); comparing posttreatment population levels with their corresponding pretreatment counts as well as with the population changes in the check ponds during the same sampling periods.

RESULTS AND DISCUSSION

EXPERIMENTAL PONDS. The pre-, and posttreatment nontarget invertebrate data pertaining to the April 27 treatments of the WP and G formulations of SIR-8514 are presented in Table 1. The WP at all 3 treatment rates (28, 56, and 112 g AI/ha) reduced *Cyclops* spp. up to 99% during the 5 wk after treatment. The lowest rate of the WP had equally deleterious effects on *Cyclops* spp. as the highest rate. *Chaoborus* sp. larvae and the semi-aquatic collembolans were also reduced by the IGR but they recovered within 1–3 weeks. *Baetis* sp. was affected for 1–2 wk after treatment. The G formulation reduced *Baetis* sp. slightly more than WP applied at comparable rates. Nymphs of corixids and notonectids were severely affected, the former for ca. 1 wk and the latter for 2–3 wk after treatment. Coleopterous larvae also declined during the 1–2 wk posttreatment but adult beetles were the least affected by either formulation of SIR-8514 even at 112 g AI/ha rate of treatment. Rotifers were not affected by this IGR, their populations rather increased during the posttreatment observations. Overall, both formulations of SIR-8514 applied at comparable rates had somewhat similar adverse effects on the nontarget invertebrates in the experimental ponds.

Diflubenzuron at 28 and 56 g AI/ha reduced 96–100% of *Cyclops* spp. populations for at least 9 days after treatment (Table 2). Recovery of *Cyclops* spp. in ponds treated at the lower rate of diflubenzuron was sooner than in those receiving its higher rate. SIR-8514 at comparable rates of application to diflubenzuron affected *Cyclops* spp. more severely and for a longer period after treatment. Collembola were affected by SIR-8514 at both rates, as well as by diflubenzuron at the higher rate but the effects of each compound lasted for <1 week. Larval *Chaoborus* sp. was temporarily suppressed by both IGRs. Immature *Baetis* sp. declined 32–85% at 28 g AI/ha and 37–85% with double the rate of diflubenzuron. Similar reductions of *Baetis* sp. were caused by SIR-8514. The beetle larvae showed slightly more sensitivity to SIR-8514 than to diflubenzuron, while adult beetles, not affected by the former compound, were reduced up to 56% at 56 g AI/ha of SIR-8514 during the 1–2 wk of posttreatment.

The EC of MV-678 applied at a rate comparable to diflubenzuron or SIR-8514 had fewer deleterious effects on invertebrates in the ponds than the latter 2 IGRs. MV-678 at 56 g AI/ha had no effect on *Cyclops* spp. but at 112 g AI/ha it reduced 82% of *Cyclops* spp. 9 days after treatment; the copepod recovered soon after (Table 2). Collembola were affected for a short while by this IGR as well as by the other 2 growth inhibitors. Larvae of *Chaoborus* sp. were affected by MV-678. Immature *Baetis* sp. and coleopterous larvae were also reduced by MV-678 but the magnitude of their reduction was not as great as induced by diflubenzuron or SIR-8514. Adult Coleoptera remained unaffected by MV-678. In general, diflubenzuron had slightly less longer-lasting adverse effects on the pond fauna (except for *Baetis* sp.) than SIR-8514, and MV-678 was the least harmful of the 3 IGRs to the various invertebrates inhabiting the experimental ponds.

SEWAGE POLISHING POND. The ostracod, *Cypridopsis* sp., the amphipod, *Hyalella azteca*, *Cyclops* spp., and the oligochaete worms were the predominant invertebrates in the sewage pond at the time of SIR-8514 application. *Cyclops* spp. and *H. azteca* showed considerable sensitivity to SIR-8514 applied at 70 g AI/ha. *Cyclops* spp., as in the experimental ponds was severely affected declining to zero within 9 days posttreatment, but it recovered rapidly (Table 3). A reduction of 98% of *H. azteca* occurred after 16 days of treatment but recovering thereafter. *Cypridopsis* sp. and Oligochaeta were not affected by the treatment.

Table 1. Effects of a wettable powder (WP) and a granular (G) formulation of the IGR, Bay SIR-8514, (applied at different rates) on some planktonic and nektonic invertebrates in experimental ponds at the aquatic research facility, University of Florida, Agricultural Research and Education Center, Sanford, FL (April–June 1979).

	Mean no. planktonic inverts./250 liters or nektonic inverts./5 dips pre-, and posttreatment (days)											
Invertebrates	Pre-treat	3	7	14	21	35	Pre-treat	3	7	14	21	35
	28 g AI/ha or 6.0 ppb (25% WP)						Check					
†Rotifera	0	15	34	>500	—	—	0	312	150	74	181	59
†*Cyclops* spp.	3702	506	20	5	2	110	56	43	193	411	849	328
*Collembola	174	15	15	21	45	9	267	108	102	9	18	3
†*Chaoborus* sp. L	1	0	1	14	27	4	22	101	61	8	15	8
**Baetis* sp. N	48	36	15	27	6	81	87	108	12	27	6	102
*Corixids[a] N	5	3	33	87	115	20	3	15	19	2	4	2
*Notonectids[a] N	2	2	6	5	28	70	1	5	16	18	39	52
*Coleoptera[b] L	20	22	5	8	3	1	32	40	20	3	2	1
*Coleoptera[b] A	23	21	26	6	9	24	23	24	22	7	2	14
	56 g AI/ha or 12.0 ppb (25% WP)						56 AI/ha or 12.0 ppb (0.5% G)					
†Rotifera	0	194	140	>500	—	—	0	34	412	>500	—	—
†*Cyclops* spp.	23734	146	91	45	5	226	398	10	2	2	1	117
*Collembola	72	6	21	18	15	3	435	48	39	27	6	3
†*Chaoborus* sp. L	26	3	0	3	27	25	85	24	1	3	57	20
**Baetis* sp. N	78	21	18	9	18	96	132	15	21	15	12	180
*Corixids[a] N	3	4	4	36	40	11	6	1	4	33	176	16
*Notonectids[a] N	2	3	7	4	28	65	3	3	6	5	76	95
*Coleoptera[b] L	12	10	1	2	6	1	23	49	6	2	16	3
*Coleoptera[b] A	10	14	22	2	3	16	29	34	37	5	6	16
	112 g AI/ha or 24.0 ppb (25% WP)						112 g AI/ha or 24.0 ppb (0.5% G)					
†Rotifera	0	133	340	>500	—	—	0	3	2	>500	—	—
†*Cyclops* spp.	2308	15	3	5	1	500	2	1	1	1	1	130
*Collembola	564	27	12	9	3	6	—	—	—	—	—	—
†*Chaoborus* sp. L	30	4	0	4	29	42	11	1	0	1	5	4
**Baetis* sp. N	111	36	18	33	6	174	252	9	12	66	12	42
*Corixids[a] N	2	1	0	8	67	38	7	0	3	9	55	11
*Notonectids[a] N	1	1	3	2	11	31	2	3	6	4	13	42
*Coleoptera[b] L	42	34	4	2	5	1	35	22	1	6	4	1
*Coleoptera[b] A	16	25	51	13	4	10	21	22	28	4	8	17

† = planktonic * = nektonic

N = nymphs L = larvae A = adults

[a] Mean no./1000 liters.

[b] Mostly Hydroporinae and Hydrophilidae.

NATURAL POND. In this pond, the cladocerans, *Diaphanosoma brachyurum* (Liéven), *Bosmina coregoni* Baird, and *Ceriodaphnia* sp., and the copepod, *Diaptomus* spp., were the abundant zooplankton prior to treatment.

The EC of MV-678 at 0.22 kg AI/ha had statistically insignificant ($P > 0.05$) effects on the predominant zooplankton (Table 4). Hydrachnellae also remained unaffected by MV-678. Similarly, no reduction in populations of leeches (Hirudinea) or oligochaetes was detected.

The results obtained during this study can be compared with a number of earlier reports on the beneficial nontarget aqua-

Table 2. Effects of three IGRs (various formulations) on some planktonic and nektonic invertebrates in experimental ponds at the aquatic research facility, University of Florida, Agricultural Research and Education Center, Sanford, FL (July–August 1979).

	Mean no. planktonic inverts./250 liters or nektonic inverts./ 5 dips pre-, and posttreatment (days)											
Invertebrates	Pre-treat	3	9	16	24	38	Pre-treat	3	9	16	24	38
	Diflubenzuron (25% WP)											
	28 g AI/ha (6.0 ppb)						56 g AI/ha (12.0 ppb)					
†*Cyclops* spp.	10	0	0	41	11	—	421	1	2	145	107	—
*Collembola	11	46	10	10	26	—	35	63	22	62	90	—
†*Chaoborus* sp. L	10	1	23	7	5	—	41	1	4	1	1	—
**Baetis* sp. N	54	16	105	24	68	—	46	14	77	13	54	—
*Coleoptera[a] L	23	6	3	2	0	—	20	5	5	3	0	—
*Coleoptera[a] A	5	6	9	1	11	—	5	3	5	1	6	—
	Bay SIR-8514 (25% WP)											
	28 g AI/ha (6.0 ppb)						56 g AI/ha (12.0 ppb)					
†*Cyclops* spp.	47	1	0	0	4	11	64	1	0	0	0	16
*Collembola	13	10	27	45	54	—	33	20	22	38	49	—
†*Chaoborus* sp. L	33	1	0	47	7	—	46	0	5	1	4	—
**Baetis* sp. N	50	23	193	18	88	—	81	14	58	33	68	—
*Coleoptera[a] L	54	2	3	3	0	—	27	1	1	2	0	—
*Coleoptera[a] A	3	5	2	3	7	—	8	4	2	4	6	—
	Stauffer MV-678 (EC 4)											
	56 g AI/ha (12.0 ppb)						112 g AI/ha (24.0 ppb)					
†*Cyclops* spp.	47	5	47	461	3040	—	200	9	6	591	1002	—
*Collembola	38	67	13	34	70	—	52	81	28	28	45	—
†*Chaoborus* sp. L	25	8	30	3	15	—	492	6	6	62	15	—
**Baetis* sp. N	29	28	47	15	104	—	39	19	80	35	85	—
*Coleoptera[a] L	20	7	1	2	2	—	28	6	2	2	0	—
*Coleoptera[a] A	2	4	2	2	4	—	1	2	6	2	7	—
	Check											
†*Cyclops* spp.	1341	81	223	1283	2437	1259						
*Collembola	36	147	14	26	38	—						
†*Chaoborus* sp. L	52	86	171	56	28	—						
**Baetis* sp. N	48	95	145	60	89	—						
*Coleoptera[a] L	25	10	1	3	0	—						
*Coleoptera[a] A	7	4	4	1	5	—						

† = planktonic * = nektonic
N = nymphs L = larvae A = adults
[a] Mostly Hydroporinae and Hydrophilidae.

Table 3. Effects of the IGR, Bay SIR-8514 (25% WP), at 70 g AI/ha (6.0 ppb) on planktonic, nektonic, and benthic invertebrates in a sewage polishing pond, Utility Department, City of Sanford, Seminole County, FL (July-August 1979).

Invertebrates	Mean[b] no. planktonic or nektonic inverts./500 liters or benethic inverts./900 cm² mud pre-, and posttreatment (days)				
	Pretreatment	7	14	21	31
			Planktonic		
Cyclops spp.	24 b	0 a	1 a	559 c	753 c
			Nektonic		
Cypridopsis sp.	48 a	55 a	49 a	1343 b	1667 b
Hyalella azteca	1051 c	128 ab	20 a	134 ab	247 b
			Benthic		
Oligochaeta[a]	412 a	964 c	696 b	1172 c	—

[a] Naididae and Tubificidae.

[b] Means in a row followed by the same letter are not significantly different ($P>0.05$) from each other when subjected to analysis of variance and Duncan's Multiple Range Test; data were transformed to log (n+1) for the analysis.

tic invertebrates as affected by IGRs in the laboratory, and in a variety of semi-field and field freshwater habitats (Apperson et al. 1978, Mulla and Darwazeh 1979, Mulla et al. 1975, Miura and Takahashi 1974, 1975, Miura et al. 1975, Schaefer et al. 1976, 1978, Steelman et al. 1975). Most of this work concerns diflubenzuron which, at application rates comparable to the ones employed here, suppressed copepods, mayfly nymphs, and coleopterous larvae, but they recovered within a few days or weeks after treatment.

The other IGR, SIR-8514, applied for mosquito control at 112 g AI/ha to 0.02 ha field plots simultaneously reduced cladocerans and mayfly nymphs, but

Table 4. Effects of the IGR, Stauffer MV-678 (EC 4), at 0.22 kg AI/ha (12.2 ppb) on planktonic, nektonic, and benthic invertebrates in a natural pond, General Electric premises, Daytona Beach, Volusia County, FL (August 1979).

Invertebrates	Mean[b] no. planktonic or nektonic inverts./100 liters or benthic inverts./900 cm² mud pre-, and posttreatment (days)			
	Pretreatment	5	11	17
		Planktonic		
Diaphanosoma brachyurum	812 a	605 a	1023 a	1781 a
Bosmina coregoni	2414 a	1877 a	3148 a	2255 a
Ceriodaphnia sp.	2169 a	819 a	1707 a	1401 a
Diaptomus spp.	9263 a	8173 a	2231 a	3114 a
		Nektonic		
Hydrachnellae	10 a	16 a	8 a	14 a
		Benthic		
Hirudinea	4 a	7 a	6 a	9 a
Oligochaeta[a]	142 a	129 a	186 a	119 a

[a] Mostly Tubificidae.

[b] Means in a row followed by the same letter are not significantly different ($P>0.05$) from each other when subjected to analysis of variance and Duncan's Multiple Range Test; data were transformed to log (n+1) for the analysis.

copepods, ostracods, and rotifers remained unaffected (Schaefer et al. 1978). Mulla and Darwazeh (1979) also reported a short-term reduction of *Baetis* sp. caused by SIR-8514 at 110 g AI/ha in experimental ponds. The results obtained with SIR-8514 in the present study are in general agreement with those of Schaefer et al. (1978) and Mulla and Darwazeh (1979). However, the observations of Schaefer et al. (1978) on copepods do not agree with the present findings where *Cyclops* spp. was severely affected by SIR-8514 even at a rate as low at 28 g AI/ha in the experimental ponds and at 70 g AI/ha in the sewage pond.

Of the studied IGRs, MV-678 applied at the same rates as diflubenzuron or SIR-8514, induced the minimum adverse effects on the nontarget organisms. MV-678 at 0.22 kg AI/ha in 1.8 m deep natural pond had no adverse effects on the cladocerans, *D. brachyurum, B. coregoni,* and *Ceriodaphnia* sp., the copepod, *Diaptomus* spp., the water mites, Hydrachnellae, and the Oligochaete worms, but in the experimental ponds at 56 and 112 g AI/ha, it caused some reductions of *Cyclops* spp., *Baetis* sp., and beetle larvae. Previously, Schaefer et al. (1976) had observed no adverse effects of MV-678 on Cladocera, mayflies, copepods, and immature Hemiptera and Coleoptera at rates of 0.04–0.2 kg AI/ha. Similarly, Mulla and Darwazeh (1979) reported no reductions of mayfly, dragonfly, and beetle larvae in 30–40 cm deep experimental ponds treated with MV-678 at 56 and 112 g AI/ha.

From these studies, it is evident that the 3 IGRs adversely affected some nontarget invertebrates in the habitats studied, but these effects lasted only for a few days or weeks. Moreover, these materials are effective against mosquitoes and midges at rates lower than those affecting many nontarget organisms (Ali and Lord 1980, Mulla and Darwazeh 1979); therefore, these IGRs offer a good potential for future use against aquatic pest and vector insects.

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