FIELD TESTS OF INSECTICIDES AND INSECT GROWTH REGULATORS FOR THE CONTROL OF *CULEX QUINQUEFASCIATUS* IN ANAEROBIC ANIMAL WASTE LAGOONS[1]

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ABSTRACT. Four insecticides and 2 insect growth regulators (diflubenzuron and methoprene) were applied to anaerobic swine waste lagoons for the control of the mosquito *Culex quinquefasciatus* (Say). Chlorpyrifos at 0.4 lb/acre (448 g/hectare) or more gave excellent mosquito control for 3–5 weeks. Malathion at 1.0 lb/A (1.12 kg/ha.) did not give satisfactory control. Temephos at 0.5 lb/A (560 g/ha) gave control for only 3–4 days. Flit MLO at 7 gal/A (65 liters/ha) gave satisfactory control for 3–4 days and was not effective at lower rates.

Diflubenzuron at 0.1 lb/A (112g/ha) gave satisfactory control for 1–2 weeks. Methoprene did not give satisfactory control at 0.4 lb/A (448g/ha). A granular formulation of diflubenzuron was about equally effective as the wettable powder.

INTRODUCTION

Disposal of manure from farm animals (swine, poultry, cattle) confined in high densities is often accomplished by flushing the manure into a lagoon. These lagoons are made by constructing an excavated pit, dam, embankment, dike, or levee or a combination of these, to retain the water and wastes. Animal waste lagoons are often operated anaerobically because aerobic treatment requires too much water for proper dilution and too much land due to the large quantities of raw wastes (Loehr 1977). In practice these anaerobic lagoons are steep-sided and 3–5 m (8–15 ft) deep. These lagoons often provide a favorable environment for the production of those species of mosquitoes which prefer polluted waters. In the southern United States the principal species is *Culex quinquefasciatus* Say (Axtell *et al.* 1975). Production of this species in anaerobic animal waste lagoons is closely related to levels of organic pollution (manure loading rates) and abundance of shelter provided by emergent marginal vegetation and floating debris. An analysis of the factors affecting production of *Cx. quinquefasciatus* in anaerobic animal waste lagoons and a review of the relevant literature has recently been published by Rutz and Axtell (1978).

Although the problem of mosquito production from animal waste lagoons can often be eliminated, or nearly so, by proper design and maintenance (free of debris and shoreline vegetation) it is sometimes necessary to apply chemicals for mosquito control. With the high levels of organic pollution, questions arise regarding the effectiveness and longevity of insecticides and insect growth regulators under these conditions. Therefore, we evaluated 4 insecticides and 2 insect growth regulators in swine waste lagoons in eastern North Carolina.

MATERIALS AND METHODS

The insecticides tested were: Flit MLO®, a refined petroleum oil, Exxon Corp.; chlorpyrifos (Dursban®), 2 lb/gal active ingredient (AI) emulsifiable concentrate, Dow Chemical Co.; malathion (Cythion®), 4 lb/gal AI emulsifiable concentrate American Cyanamid Co.; temephos (Abate®), 2% AI granules, American Cyanamid Co. The insect

[1]Paper No. 6050 of the Journal Series of the North Carolina Agricultural Research Service, Raleigh, N.C. Use of trade names in this publication does not imply endorsement of the products named or criticism of similar ones not mentioned.

growth regulators (IGR'S) tested were: diflubenzuron (Dimilin®), 25% AI wettable powder and 1% AI granules, Thompson-Hayward Chemical Co.; methoprene (Altosid® SR–10), 0.86 lb/gal AI liquid, Zoecon Corp.

The chemicals were applied, by means of a hand-operated, compressed air sprayer, to the water surface of swine waste lagoons in which substantial numbers of mosquito larvae were present. The spray was applied to the margin of each lagoon in about a 10 ft (3m) band that could be easily reached from the shoreline. About 3–4 gal (1–15 liters) of spray was applied per lagoon with the chemical concentration adjusted to obtain the desired amount of active ingredient per acre of surface. Flit MLO was applied at the volume desired and granules of temephos were broadcast by hand. The lagoons were on swine farms located in eastern North Carolina (Columbus County). The sizes varied from 0.05 to 0.17 acres (0.02–0.07 ha.) with the shapes generally rectangular, the sides steeply sloped and the depths from 8 to 10 ft. (2.4–3.0 m). Repeated larval sampling and examination of adult male and female mosquitoes (emerged from larvae held in the laboratory) during the 3 years of testing verified that nearly all of the mosquitoes occurring in these lagoons were *Cx. quinquefasciatus.*

Evaluations of the effects of the chemical treatments were made by determining the number of mosquito larvae and pupae by standardized dipping with a 1-pt dipper at 10 approximately equal intervals around the perimeter of the lagoon and calculating the avg. no./dip. These counts were made immediately before treatment and at regular intervals (usually 3–4 days) after treatment. In the cases of treatments with insecticides, all larval instars as well as pupae were included in the counts and the % reduction was calculated by comparing the posttreatment counts to the pretreatment count for the same lagoon. The final % control for each treatment at each sample date was corrected for any reductions in the numbers of mosquitoes in the untreated lagoon by means of Abbott's formula.

In tests with the insect growth regulators diflubenzuron and methoprene it was necessary to use different procedures of evaluation due to the mode of action of these chemicals, i.e. they interfere with the normal molting process and most of the mortality is expressed in the pupal stage or in the failure of adults to emerge from the pupae. Therefore, from each lagoon pupae and/or 4th instar larvae (150) were collected and held in the laboratory in fresh water in screen-topped cups (25/cup) to determine the percentage yielding adults which were mobile and not deformed. Adult emergence from pupae and larvae collected from the untreated lagoon and held in the laboratory was routinely greater than 90% and usually 96–100%. In addition, counts were made by standardized dipping at 10 approximately equal intervals around the margin of each lagoon on each sample date to detect any changes in mosquito abundance which might possibly occur at the higher concentrations of the IGR's. Only the 3rd and 4th larval instars and the pupae were counted and used to calculate the avg. no./dip.

Diflubenzuron and methoprene were further evaluated in simulated swine waste lagoons which were metal drums (55 gal, 0.21 m^3) sunk into the ground with water and manure added. The weekly manure additions were equivalent to 160 and 320 ft^3 (4.6 and 9.2 m^3) lagoon volume per 100 lb (45 kg) hog or 29 and 14.5 lb COD (chemical oxygen demand) per 1000 ft^3 per wk (0.45 and 0.23 kg per m^3 per wk) in the first and second tests, respectively. In each test there were 3 replicates (drums) per treatment. Two formulations of diflubenzuron (wettable powder and granules) were also compared in these simulated lagoons using poultry manure added at 3 rates. The manure loading rates were equivalent to 17.6, 70.6 and 277.9 ft^3 (0.5, 2.0, 7.87 m^3) lagoon volume per 4 lb (1.8 kg) chicken or 15.9, 4.0 and 1.0 lbs COD per 1000 ft^3 per

week (0.25, 0.063 and 0.16 per kg per m^3 per wk).

RESULTS AND DISCUSSION

The results of the 4 tests of insecticides are shown in Table 1. Chlorpyrifos applied at the lowest rates of 0.01 lb/A (11.2 g/ha) and 0.05 lb/A (5.6 g/ha) gave very little control of *Cx. quinquefasciatus* (75.3 and 28.5%, respectively at 3–4 days posttreatment). At 0.1 lb/A (112 g/ha.) and 0.2 lb/A (224 g/ha) control was satisfactory (90% or more) for up to 7 days posttreatment. At rates of 0.4 lb/A (448 g/ha) and more, chlorpyrifos gave satisfactory mosquito control for 21 days or more. Malathion at 1.0 lb/A (1.12 kg/ha) gave unsatisfactory control. Temephos at 0.3 lb/A (336 g/ha) gave unsatisfactory control and at 0.5 lb/A (560 g/ha) gave satisfactory control (98.2%) only for 3–4 days. Flit MLO treatments yielded variable results at 5 gal/A (46 liters/ha) and effective control (98.6%) for 3–4 days at 7 gal /A (65 liters/ha). Lower rates of application of Flit MLO were previously tested in simulated lagoons (drums) and were not effective in controlling *Cx. quinquefasciatus* (unpublished data).

Of the 4 insecticides tested, chlorpyrifos was the most effective and practical since frequent retreatment which would be required by the other insecticides is expensive and inconvenient. The rates of chlorpyrifos required in our tests for effective control of *Cx. quinquefasciatus* in animal waste lagoons were higher than the rates generally recommended for waters with considerably less organic pollution. Based on our results, an application of chlorpyrifos at 0.5 lb/A (560 g/ha) should generally give satisfactory mosquito control in animal waste lagoons for 3–5 weeks (see footnote, Table 1).

Table 2 presents the results of the field tests with the insect growth regulators diflubenzuron and methoprene. At the rates of 0.4 lb/A (448 g/ha) and 0.2 lb/A (224 g/ha.) methoprene gave only partial and unsatisfactory mosquito control in the waste lagoons and the results were highly variable. At 4 days posttreatment the lowest adult emergence obtained was 18.7% and at 7 days posttreatment the lowest was 30.7%.

Diflubenzuron gave no mosquito control at the rate of 0.03 lb/A (33.6 g/ha) and at 0.06 (672 g/ha) partially inhibited adult emergence. At 0.08 lb/A (89.6 g/ha) mosquito control was improved with only 1.4% and 10% adult emergence at 7 days posttreatment in 2 tests. Some initial reduction in the numbers of larvae and pupae were evident at this rate. At 0.1 lb/A (112 g/ha) diflubenzuron was consistently effective for at least 7 days in drastically reducing the numbers of larvae and pupae in the lagoons to the extent that the dip counts were at or near zero and extensive searching was required to find enough larvae or pupae to hold in the laboratory. No adults emerged from these specimens. At 14, 21 and 28 days posttreatment there were some reductions in adult emergence but these were variable.

Based on these field tests, diflubenzuron at 0.1 lb/A (112 g/ha) can be expected to give satisfactory control of *Cx. quinquefasciatus* in animal waste lagoons for 1 to 2 weeks. This was supported by additional tests in simulated lagoons (Table 3). Methoprene at 0.4 lb/A (448 g/ha) or less did not give satisfactory control; higher rates may be effective but were not tested. Additional tests with methoprene at 0.1 lb/A (112 g/ha.) in simulated lagoons (Table 3) resulted in unsatisfactory control with 37.3 and 31.4% adult emergence 3 days posttreatment. These data suggest that the high levels of organic pollution in animal waste lagoons may interfere with the action of methoprene.

The wettable powder formulation of diflubenzuron was used in all of these tests. In a comparison of the granular and the wettable powder formulations applied at the same amount of A.I. (0.1 lb/A, 112 g/ha) both gave essentially complete mosquito control (i.e., little or no adult emergence) for 14 days posttreatment

Table 1. Duration of effectiveness of treatments with 4 insecticides for the control of *Culex quinquefasciatus* in anerobic swine waste lagoons.

Chemical & rate of application (lb or gal/acre)	Lagoon Code No.	Pretreatment Avg. no. larvae & pupae/dip[a]	Avg. no. larvae & pupae/dip (N) and percent (%) at days posttreatment[a, b]							
			3–4 da		7 da		14 da		21 da	
			N	%	N	%	N	%	N	%
Test no. 1—Treated Sept. 13, 1973										
Malathion, 1.0	3	82.0	17.8	78.3	96.1	0	161.4	0		
Flit MLO, 5 gal	1	216.3	16.3	92.5	21.8	89.9	286.1	0		
" , 5 gal	2	1043.9	1439.3	0	876.2	16.1	968.6	7.2		
untreated	7	862.7	2870.4		2088.4		1740.8			
Test No. 2—Treated Sept. 26, 1973										
chlorpyrifos, 1.0	2	968.6	0	100	0	100	0	100	0	100
" , 0.5	4	138.3	0	100	0	100	0	100	15.3	88.9
malathion, 1.0	3	161.4	2.6	98.4	50.8	53.6	205.6	0	246.9	0
Flit MLO, 7 gal	1	286.1	4.0	98.6	31.0	84.1	306.9	0	456.1	0
untreated	7	1740.8	2805.5		1182.0		1436.6		2277.1	
Test no. 3—Treated July 4, 1974										
chlorpyrifos, 0.1	3	253.2	0.8	99.6	0.5	99.8	84.3	0		
" , 0.05	4	139.2	81.2	28.5	179.0	0	282.3	0		
" , 0.01	12	671.2	135.9	75.3	589.1	9.4	527.1	0		
temephos, 0.3	9	159.4	34.2	73.7	153.0	.9	436.5	0		
untreated	7	1170.2	956.5		1134.0		268.3			
Test no. 4—Treated August 22, 1974										
chlorpyrifos, 0.4	3	1641.1	0	100	0	100	0	100	0[c]	100
" , 0.2	4	323.1	3.9	98.8	2.8	98.7	177.9	0	1685.1	0
" , 0.1	12	1671.4	8.3	99.5	120.3	89.7	366.9	59.6	637.6	47.4
temephos, 0.5	9	134.6	2.4	98.2	73.7	22.0	51.9	29.2	269.9	0
untreated	7	616.3	834.7		433.1		335.9		247.1	

[a] Based on 10 dips per lagoon at each sampling time.

[b] % mortality in the treated as compared to the pretreatment counts and corrected for mortality in the untreated lagoon by Abbott's formula.

[c] Larvae reappeared in this lagoon in the 36 da posttreatment counts.

Table 2. Duration of effectiveness of the insect growth regulators diflubenzuron (Dimilin®) and methoprene (Altosid SR-10®) for the control of *Culex quinquefasciatus* in anaerobic swine waste lagoons.

Chemicals & rate of application (lb AI/Acre)	Lagoon Code No.	Pretreatment Avg. no. 3 & 4th instar larvae & pupae/dip[a]	Avg. no. 3rd & 4th instar larvae & pupae/dip (N) and percent adult emergence (%) at days posttreatment[a].									
			4 da		7 da		11 da		14 da		21 da	
			N	%	N	%	N	%	N	%	N	%
Test no. 1—Treated July 4, 1974												
diflubenzuron, 0.1	2	49.6	0	1.4	0	(0)			2.4	—	69.1	74.0[b]
" , 0.6	5	34.6	89.8	2.0	83.7	36.7			23.1	—	11.1	—
" , 0.03	10	188.1	143.3	70.7	197.6	94.0			127.3	—	138.8	—
methoprene, 0.2	11	158.0	195.5	72.7	48.9	—			166.0	74.7	296.7	95.4
untreated	7	318.2	409.0	100	340.5	100			70.3	100	98.8	100
Test no. 2—Treated Aug. 22, 1974												
diflubenzuron, 0.1	2	134.8	0.5	0	0.1	0			147.9	10.0	133.7	38.0[c]
" , 0.06	10	247.7	44.7	11.4	155.6	15.4			554.1	25.4	189.6	62.7[d]
methoprene, 0.4	11	402.8	274.8	18.7	162.8	30.7			228.0	65.4	297.4	96.0
untreated	7	255.5	288.2	100	115.1	94.0			197.0	98.0	116.6	100
Test no. 3—Treated June 19, 1975												
diflubenzuron, 0.1	2	58.1	0	0	0	0	73.2	44.7	89.0	75.3		
" , 0.08	10	564.3	212.5	0.7	363.9	10.0	655.4	87.4	333.8	91.4		
" , 0.06	12	68.7	115.1	2.6	102.3	32.0	206.2	75.3	200.5	91.7		
untreated	7	98.7	159.0	98.0	47.1	100	59.9	96.0	25.1	92.0		
Test no. 4—Treated July 31, 1975												
diflubenzuron, 0.08	2	380.9	4.3	9.7	96.0	1.4	205.6	56.0	196.0	86.7		
methoprene, 0.04	12	576.3	344.3	26.7	516.9	50.7	303.5	68.7	246.2	92.0		
" , 0.02	10	1105.7	661.6	38.0	811.8	94.0	445.1	89.4	—	—		
untreated	7	124.9	92.9	96.0	247.7	98.0	245.9	94.0	261.4	96.0		

[a] Avg. no. larvae & pupae/dip based on 10 dips/lagoon; % adult emergence based on 150 larvae (4th instar) and/or pupae held in fresh water in the laboratory.

[b] 98.0% adult emergence at 28 days posttreatment.

[c] 50.7% adult emergence at 28 days and 84.7% at 35 days posttreatment.

[d] 94.0% adult emergence at 28 days posttreatment.

Table 3. Duration of effectiveness of the insect growth regulators diflubenzuron (Dimilin®) and methoprene (Altosid SR-10®) for the control of *Culex quinquefasciatus* in simulated (55-gal drums) anaerobic swine waste lagoons.

Chemical & rate or application (lb A.I./acre)	Pretreatment Avg. no. 3rd & th instar larvae & pupae/dip[a]	Avg. no. 3rd & 4th instar larvae & pupae/dip (N) and percent adult emergence (%) at days posttreatment[a].									
		3 da		8 da		11 da		15 da		18 da	
		N	%	N	%	N	%	N	%	N	%
Test no. 1—Treated June 10, 1974											
diflubenzuron, 0.1	157.9	131.8	0	4.2	0	83.2	2.6	329.6	68.0	323.2	94.0
" ,0.03	150.9	129.4	38.6	51.4	68.0	187.4	97.4				
methoprene, 0.1	120.6	111.3	37.3	199.8	32.0	144.2	98.0				
untreated	378.6	160.2	98.0	345.2	100	165.1	100	2869.	98.0	126.5	99.4
Test no. 2—Treated Sept 9, 1974											
diflubenzuron, 0.1	123.5	2.2	0	0	[b]	.1	[b]	6.3	[b]	28.6	57.0[c]
" , 0.03	109.6	26.8	18.7	24.5	38.7	22.0		51.7	62.7	68.9	66.7[d]
methoprene, 0.1	140.2	54.7	31.4	72.4	70.0	106.3	73.4	60.0	81.4	84.6	94.0
untreated	116.9	105.0	99.4	155.7	99.4	137.6	99.4	70.1	94.0	99.0	94.0

[a] Avg. no. larvae & pupae/dip based on 3 dips/drum, 3 drums/treatment; % adult emergence based on up to 150 larvae and/or pupae held in fresh water in the laboratory.

[b] No larvae or pupae were present to hold for adult emergence.

[c] Adult emergence was 77.0% at 25 days posttreatment and 94.7% at 29 days posttreatment.

[d] Adult emergence was 94.0% at 25 days posttreatment.

Table 4. Comparison of the duration of effectiveness of wettable power (25% A.I.) and granular (1% A.I.) formulations of diflubenzuron (Dimilin®) applied at 0.1 lb A.I./A (112 g/ha.) for the control of *Culex quinquefasciatus* in simulated (drums) poultry waste lagoons with 3 manure loading rates.

Loading rate (ft^3 lagoon volume/4 lb chicken)	Avg. % adult emergence at days posttreatment[a]					
	2 da	4 da	7 da	10 da	14 da	21 da
			Wettable powder			
17.6	0	0	0	0	3.2	10.3
70.6	1.0	0	0	0	2.9	88.2
277.9	0	9.5	21.1	54.6	16.7	57.3
			Granules			
17.6	2.3	0	0	1.7	0	2.5
70.6	1.9	0	0	0	0	5.0
277.9	3.3	0	5.3	3.0	24.1	68.5

[a] Avg. % adult emergence based on 45 larvae or pupae collected from each drum (3 drums/treatment) and held in the lagoon water in the laboratory. % emergence corrected for mortalities in samples from untreated (control) drums with the same loading rates by means of Abbott's formula.

when applied to simulated lagoons (drums) with poultry manure added at 3 rates (Table 4).

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