PYRETHRIN TOSSITS AGAINST MOSQUITO LARVAE AND THEIR EFFECTS ON MOSQUITO FISH AND SELECTED NONTARGET ORGANISMS[1]

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ABSTRACT. Excellent control of larvae of *Culex tarsalis* in experimental ponds was obtained at the rate of 1 tossit/3 m^2 (44 g AI/ha). At the same rate, satisfactory control (92%) of *Cx. peus* larvae was obtained in dairy wastewater lagoons in Riverside County, Calif. At the effective mosquito larvicidal rate (44 g/ha), mosquito fish, *Gambusia affinis*, was affected when some tossits were placed with the fish in 0.9 m^3 screened cages. Five successive weekly treatments of *G. affinis* at the larvicidal rate and double that rate (44 and 88 g/ha) resulted in 37 and 67 percent reduction of fish yield harvested 42 days after the first treatment compared to the yield obtained from the untreated ponds (checks). No marked effects on some of the nontarget organisms such as dragonfly naiads, ostracoda, copepods, and *Dixa* midge larvae. Mayfly naiads were eliminated at all rates applied, but recovered within 2–3 weeks after treatment.

Utilization of briquettes and tossits in mosquito control programs is not a new concept, as these formulations were developed and used as early as 1949 (Raley and Davis 1949). They were developed to prolong the activity of mosquito larvicides, so that frequency of treatments in a given problem area could be reduced. In addition, tossits and briquettes are easily applied to small breeding sources where rough terrains hinder access and maneuverability of spray equipment. These formulations are also quite suitable for the treatment of domestic and periodomestic small breeding sources of mosquitoes such as catch basins, swimming pools, ornamental ponds, etc.

Raley and Davis (1949) used dieldrin impregnated briquettes, and obtained good larval control with one treatment for 5 years. Other organophosphorus larvicides and insect growth regulators formulated in briquettes of cement and plaster of paris, or in rubber mattrix were utilized successfully in mosquito control programs. These formulations include the use of chlorpyrifos, temephos and methoprene (Barnes and Webb 1968, Carestia et al. 1971, Evans and Fink 1960, McDonald and Dickens 1970, and Mulla et al. 1977).

Recently, a number of synthetic pyrethroids were reported to possess a high level of activity against larvae of several species of mosquitoes which are known to be highly resistant to chlorinated hydrocarbons and organophosphorus larvicides (Mulla et al. 1978a). At the larvicidal rates of 0.001–0.025 lb/A (1.1–28 g/ha) some of these pyrethroids displayed good margin of safety to some nontarget organisms and 4 species of freshwater fishes, including the mosquito fish *Gambusia affinis* (Baird and Girard), the desert pupfish *Cyprinodon macularius* (Baird and Girard), *Tilapia mossambica* (Peters), and rainbow trout *Salmo gairdneri* (Mulla et al. 1978b). These pyrethroids hold promise for use as tossit or briquette formulations.

The current studies were carried out to evaluate the activity and longevity of natural pyrethrin tossits against mosquito larvae, and to study their effects on the mosquito fish *G. affinis* and some nontarget organisms.

MATERIALS AND METHODS

Pyrenone® tossits containing 1% natural pyrethrins were provided by Fairfield American Corp. (3932 Salt Road,

[1] These studies were conducted in cooperation with the Coachella Valley and Northwest Mosquito Abatement Districts in Riverside County.

Medina, NY, 14103). Each tossit weighed 4.36 g, and contained 1% emulsifiable concentrate of pyrethrins. The emulsifiable concentrate material is encapsulated in a water soluble gelatin capsule, with a rubber band to create internal pressure and to enhance rupturing, causing a forceful discharge of the contents within a few minutes upon contact of the capsule with water. The tossits were evaluated against mosquito larvae in experimental ponds at the Aquatic Research Facilities located at the University of California at Riverside, and in the Coachella Valley of southern California. At both locations, mosquito larval populations consisted mostly of *Culex tarsalis* Coquillett. However, *Culiseta inornata* Williston larvae were present in small numbers in the Coachella Valley facility. These experimental facilities were described earlier by Mulla and Darwazeh (1971). The ponds at the Coachella Valley facility measure 30 m^2 in size, while those at Riverside are 27 or 33.7 m^2. Water level at both locations was maintained at 30 cm in depth by float valves.

The tossits were also evaluated at Vandemeer Dairy wash lagoon, located on Archibald Blvd. and Schliesman Ave., 6 miles south of Pomona Freeway (Highway 60) in Riverside County. *Culex peus* Speiser larvae were heavily concentrated in dense floating vegetation (moves from side to side according to wind direction), and along the vegetated sides of the lagoon. The mosquito breeding area measured 122 m in length and 18 m in width (0.55 acres). Wash water in the lagoon is pumped out regularly, and the water is used for irrigation; therefore, water depth in the basin is constantly changing, and fluctuates from 1.2–3.5 m, depending on irrigation and milking cycles. To this lagoon, 226 tossits were applied, each 3 m apart, covering the entire breeding area at the rate of 44 g/ha AI (1 tossit/100 ft^2). Larval assessment was conducted by taking 20 dips in the vegetation, along the side, prior to, and 2 and 7 days after treatment.

In the experimental ponds, the required number of tossits for each application rate were placed in the pond, each at equal distance from the other to insure good coverage. In each test, 3–4 replicates were used per application rate and the check. Procedures utilized to determine the efficacy of the tossits against mosquito larvae and nontarget organisms were published earlier by Mulla et al. (1978a). Five dips per pond were taken prior to, 2 days and weekly after treatment, until the mosquito larval populations reestablished and reached the late stages of development (3rd–4th stages). The 5 dips on each sampling date were concentrated into one composite sample, and preserved by the addition of 95% ethyl alcohol. Organisms found in the sample were counted and identified under a dissecting microscope in the laboratory.

Fish studies were conducted in the ponds at the Aquatic Research Facility at Riverside. In the first test, a 0.9 m^3 window screen cage was placed in the middle of each pond, and 20 fish (3 cm) in size were added to each cage. After 2 days of fish acclimation, the tossits were applied, utilizing 4 replicates per application rate and the check. In 2 replicates, all the tossits were placed outside the fish cage, while one of the tossits was placed inside the cage in the remaining 2 replicates. Fish mortality readings were taken 1, 2, 4 and 8 days after treatment.

In the second test at the Aquatic Research Facility at Riverside, 25 fish of *G. affinis* (avg. 3 cm) were placed in each pond 10 days after flooding, when mosquito larval population and other aquatic organisms were established. The pond system consists of 12 ponds, constructed in 3 rows, 4 ponds/row, and each measuring 4.5 × 7.5 m (33.7 m^2). Water level in the pond system was maintained constant (30 cm) by the use of float valves. The ponds are provided with a drainage system through (10 cm) pipe placed in the bottom of each pond, leading into 1.2 m^3 concrete weir structure. Each pond could be drained separately within 15–20 min by removing the screw cap at the end of

the drain pipe, and the fish from each pond could be captured and retrieved with a plankton net.

After 2 days of acclimation period, 4 or 8 Pyrenone tossits were placed/pond, leaving equal distance between tossits, at an application rate of 44 and 88 g/ha active ingredients. Four replicates were used per application rate and the check. Five consecutive weekly applications were made and 7 days after the last application, the ponds were drained and fish present were retrieved and counted.

Prior to each treatment and 2 and 7 days after treatment, 5 dips per pond were taken and the number of mosquito larvae present were counted. All non-target organisms recovered by dipping were also monitored and recorded on each sampling date.

During the course of these studies (May–June 1980), water temperature was monitored with a mini-max recording thermometer (Markson Science Inc., Del Mar, CA 92014), and water pH in the ponds was in the range of 8.0–8.2.

RESULTS AND DISCUSSION

At the recommended rate of 1 tossit/3 m² (44 g/ha), excellent control (96%) of *Cx. tarsalis* and *Cs. inornata* was obtained 2 days posttreatment. In the test conducted at Riverside, the population recovered rapidly and was reestablished within one week after treatment. Increase in the application rate (2–4 times the recommended rate) did not increase the longevity of the toxicant in water but marked reduction was obtained at 9 times the recommended rate one week posttreatment (Table 1).

In the Coachella Valley, however, complete control of 3–4 stage larvae was possible for a week or so at 1.25–1.85 tossits/3 m² (44–66 g/ha) (Table 2). No significant reduction in the population was attained at the lower rate of 11 g/ha, but 90% reduction occurred for one week at 22 g/ha. At all rates tested, the larval population recovered 2 weeks after treatment (Table 2).

In a dairy lagoon, at the recommended rate (1 tossit/3 m² or 44 g/ha), 92% reduc-

Table 1. Evaluation of Pyrenone® tossits (1% pyrethrins) against *Cx. tarsalis* in experimental ponds.[a] (UCR Aquatic Res. Facility, July 1979).

Rate			Average no. larvae and pupae/5 dips pre- and posttreatment (days)							
			Pretreatment		2			7		
lb/A	g/ha	Tossit/pond	Larvae	Pupae	Larvae	Pupae	(%R)	Larvae	Pupae	(%R)
					Test A					
0.0145	12	1	81	25	190	17	0	267	14	0
0.0290	24	2	100	19	49	21	51	92	0	8
0.0435	36	3	55	22	2	21	96	39	1	29
Check	—	—	66	16	280	80	—	330	43	—
					Test B					
0.0435	36	3	93	8	3	4	97	54	8	42
0.0870	72	6	67	6	0	3	100	42	1	37
Check	—	—	34	0	33	0	—	28	1	—
					Test C					
0.1885	150	13	134	0	0	0	100	72	0	46
0.3770	300	26	165	5	0	0	100	18	0	89
Check	—	—	262	0	187	0	—	240	0	—

[a] Pond size 12 x 24 = 288 ft² (27 m²).

(%R) = Percent reduction is based on number of larval count in posttreatment vs. pretreatment.

Table 2. Evaluation of 1% Pyrenone® tossits (1% pyrethrins) against mixed populations of *Cx. tarsalis* and *Cs. inornata* larvae in experimental ponds (Coachella Valley Aquatic Research Facility, CA, Nov. 1979).

Rate			Average no. larvae and pupae/5 dips pre- and posttreatment (days)										
			Pretreatment			2				7			
lb/A	g/ha	Tossit/ pond[a]	1–2	3–4	Pupae	1–2	3–4	Pupae	(%R)	1–2	3–4	Pupae	(%R)
0.01	11	1	46	48	4	14	44	4	8	16	18	5	63
0.02	22	2	37	51	9	10	5	5	90	11	4	2	92
0.04	44	4	38	46	6	3	0	1	100	22	0	0	100
0.06	66	6	83	72	10	5	0	0	100	7	0	0	100
Check	—	—	11	8	21	4	5	21	—	11	4	15	—

[a] Pond size 18 x 18 = 324 ft² (30 m²).
(%R) = Percent reduction is based on 3–4 larval stages in posttreatment vs. pretreatment.

tion in the larval population of *Cx. peus* was obtained 2 days after treatment, but larvae reestablished within a week (Table 3).

Differences in the results obtained in the Riverside and Coachella Valley facilities could be attributed largely to the rate of water flow into the ponds which exceeded 15 liters/min/pond in the Riverside facility, compared to 1 liter/min/pond in the Coachella Valley. Also, the Coachella Valley ponds have natural vegetation, while the Riverside facility is kept clean of plant growth.

As shown in Tables 1 and 2, pupae were not greatly affected at the larvicidal rate of 1 tossit/3 m² 2 days after treatment, but were greatly reduced 1 week posttreatment. This reduction is attributed to the excellent larval control obtained rather than to the effectiveness of the material against pupae. Higher rates, however, (2–9 times the recommended larvicidal rates) were toxic to pupae which were controlled for more than one week.

At the larvicidal rate of 1 tossit/3 m² (44 g/ha), nontarget organisms such as mayflies were completely eliminated for 2 weeks. At the higher rate of 66 g/ha, dragonfly naiads were not adversely affected. Ostracoda were also not affected at all rates applied (Table 4).

Studies on caged mosquito fish *G. affinis* produced interesting results. They were markedly affected at both rates applied (44 and 88 g/ha) when one tossit was placed inside the fish cage. The higher rate produced complete mortality, while the low rate caused 55% mortality 4 days after treatment. When all tossits

Table 3. Evaluation of Pyrenone® tossits (1%) against *Cx. peus* in dairy waste water lagoons[a] (Riverside County, California, Oct. 1979).

Location	Rate	Average no. larvae and pupae/dip pre- and posttreatment (days)							
		Pre-treatment		1			7		
	g/ha	Larvae	Pupae	Larvae	Pupae	(%R)[b]	Larvae	Pupae	(%R)[b]
Vandemeer Dairy	44	87	7	7	2	92	17	1	80
Hettinga Dairy	Check	30	2	38	5	0	32	4	0

[a] Prior to treatment, water depth was 1.8 m but was increased to 2.7 m 24 h after treatment.
[b] Percent reduction calculation is based on number of all larval stages in posttreatment vs. pretreatment.

Table 4. Effect of Pyrenone® tossits (1% pyrethrins) against nontarget organisms in experimental ponds (Coachella Valley Aquatic Research Facility, CA, Nov. 1979).

Rate			Average no. of larvae and pupae/5 dips pre- and posttreatment (days)														
			Mayfly naiads					Dragonfly naiads					Ostracoda				
lb/A	g/ha	Tossit/ pond	Pre-	2	7	14	21	Pre-	2	7	14	21	Pre-	2	7	14	21
0.01	11	1	17	25	9	9	9	1	4	19	11	12	23	30	59	41	81
0.02	22	2	32	0	1	9	7	0	1	8	18	26	5	12	56	73	161
0.04	44	4	22	0	0	4	13	2	4	8	13	23	9	48	27	42	34
0.06	66	6	33	0	0	1	2	1	0	0	10	33	12	16	64	128	235
Check	—	—	3	5	5	3	3	0	0	0	0	1	6	3	23	33	73

were placed outside the fish cage, no fish kill occurred at the low rate while 43% mortality occurred at the higher rate, which is 2 times the recommended larvicidal rate (Table 5).

In another test using the same rates, 5 successive weekly treatments did not affect an increase in the number of free-swimming *G. affinis* in the ponds. However, both rates caused some reduction in the number of fish harvested at the end of the experiment. Compared to fish yield harvested from the check ponds, 37 and 67% reduction occurred in the fish from ponds treated at the rates of 44 and 88 g/ha, respectively. In addition, all mosquito larval stages were eliminated in all treated ponds 1 day after treatment, but 1st stage larvae began to appear 3 days after treatment and all stages were present one week after treatment. These findings indicate rapid degradations of pyrethroids in water when applied weekly. Furthermore, joint action of pyrethrins and mosquito fish *G. affinis* (25/pond) did not eliminate the mosquito larval population completely, which persisted for 5 weeks (Table 6). Nontarget organisms such as *Dixa* midge larvae and copepods were observed in all treated ponds during the duration of these studies.

Table 5. Effects of Pyrenone® tossits (1% pyrethrin) against the mosquito fish *Gambusia affinis* in experimental ponds (UCR, Aquatic Res. Facility, Nov. 1979).

Rate			Average (%) cumulative fish mortality after treatment (days)			
lb/A	g/ha	Tossit/pond	1	2	4	7
			1 tossit inside fish cage			
0.04	36	3	0	12	55	55
0.08	72	6	0[b]	18[b]	100	100
—	—	Check	0	0	1	1
			All tossits outside fish cage			
0.04	36	3	0	0	0	0
0.08	72	6	0[b]	8[b]	43	43
—	—	Check	0	0	1	1

[a] Pond size 12 x 24 ft. = 288 ft² (27 m²).
[b] Fish in distress.

From the data presented, pyrethrin tossits could be used effectively for mosquito larval control at the rate of 1 tossit/3 m² in situations where free-swimming and surface-frequenting fish such as *G. affinis* are present. In these situations it is likely that Pyrenone tossits will have little effect on bottom feeding fish. Other nontarget insects which were eliminated at larvicidal or higher rates recovered rapidly and reestablished within 2–3 weeks after treatment. Higher rates in excess of 44 g/ha should be avoided where surface-frequenting fish are present.

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Table 6. Effect of Pyrenone® tossits (1%) against mosquito larvae of *Cx. tarsalis* and mosquito fish *G. affinis* in experimental ponds[a] (Aquatic Research and Vector Control Facility, University of California, Riverside, May–June 1980).

	Average no. mosquito larvae/5 dips pre- and posttreatment (days)								
	44 g/ha			88 g/ha			Check		
Weekly Treatment No.	Pre-	3[b]	7	Pre-	3[b]	7	Pre-	3	7
1	56	0	10	117	0	38	56	78	46
2	10	17	9	38	29	49	46	29	43
3	9	1	1	49	11	70	43	2	1
4	1	5	0	70	34	59	1	1	2
5	0	0	—	59	2	—	2	2	—
Average no. fish harvested/pond[c]	748 ab			395 a			1190 b		
(%) decline	37			67			—		

[a] Water temperature 17.7°C mean min. and 29°C mean max.

[b] Only newly hatched 1st stage larvae present.

[c] Twenty-five fish/pond were introduced prior to the first treatment. Means followed by the same letter(s) in a row are not significantly different from one another, using Duncan's multiple range test (p = 0.05).

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