The Effect of an Experimental Application of Dimilin® Upon Selected Forest Fauna

by

C.H. Buckner

B.B. McLeod

P.D. Kingsbury

Chemical Control Research Institute

Ottawa, Ontario

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ABSTRACT

The impact of the insecticide Dimilin® was studied on several components of the forest ecosystem in an experimental aerial application of 350 g/ha (5 oz/acre) in 1975. Emphasis was placed on the effects of this chemical upon populations of small songbirds, small mammals, honey bees and components of the aquatic ecosystem. Laboratory tests were initiated to study the toxicity of Dimilin® to different species of fish and its effect upon the moulting processes of crayfish. Small forest songbirds were not affected by this treatment and honey bee colonies remained normal in all aspects monitored. Small mammal populations were too low to assess for impact. There were indications of an impact upon amphipods and aquatic beetle larvae in a small stream within the treatment block. Laboratory studies revealed no toxic effects of Dimilin® on bullheads or sunfish with exposure to up to 125 ppm active ingredient for two weeks and no indications of toxicity or blockage of moulting in juvenile crayfish. Further field and laboratory testing of the effects of Dimilin® on non-target organisms is recommended.

RESUME

On a étudié, en 1975, les incidences de l'insecticide Dimilin® sur quelques organismes forestiers, après application aérienne à la dose de 350 g/ha (5 on/acre). On s'intéresse surtout aux effets exercés sur les petits oiseaux chanteurs, les petits mammifères, les abeilles et certains être aquatiques. Des essais en laboratoire ont été entrepris afin de déterminer la toxicité du produit sur différentes espèces de poisson et sur la mue de l'écrevisse. Les oiseaux de la forêt n'ont aucunement souffert du traitement, et le comportement des abeilles domestiques est demeuré normal, du moins dans les limites étudiées. Les petits mammifères étaient trop peu nombreux pour que l'effet ait pu être mesuré. Il semblerait y avoir un effet sur les amphopodes et les larves de coléoptères aquatiques d'un petit cours d'eau arrosant la parcelle à l'étude. Les essais en laboratoire n'ont fait ressortir aucun effet toxique de l'insecticide sur la barbotte ni sur le crapet-soleil, pour des expositions à 125 p.p.m. de substance active pendant deux semaines, ni chez l'écrevisse juvénile, pas plus qu'il n'a bloqué la mue chez cette dernière. On recommande de faire faire d'autres essais de terrain et de laboratoire pour observer les effets de la Dimilin® sur les organismes non visés.

INTRODUCTION

An experimental aerial application of 350 gm/ha (5 oz/acre) Dimilin® N[[4-Chlorophenyl) amino] carbonyl]-2,6-difluorobenzamide was carried out on a 242.8 hectare (600 acres) plot on Manitoulin Island, Ontario on June 9th and 10th 1975 to field test this chemical upon a spruce budworm Choristoneura fumiferana (Clem.) infestation. Manitoulin Island lies at the mouth of the St. Mary's River in Lake Huron and was selected because it is isolated from adjacent budworm infestations and it has a good representation of boreal forest fauna. Natural populations of small songbirds, small mammals and aquatic invertebrates were monitored for adverse side-effects. Colonies of domestic honey bees, Apis mellifera L. were placed in the forest on the treatment and untreated control plots to measure the impact of the application upon the field force, brood, queens and overall general health and honey production of the colonies. Aquatic invertebrates were monitored in a small stream flowing through the treatment area. Preliminary laboratory studies were initiated to determine the effects of Dimilin® upon the moulting processes of crayfish and its toxicity to bullheads and sunfish.

METHODS

Birds:- - Songbird populations were assessed daily on treated and untreated plots for 5 days prior to and continuing for 5 days after the application of Dimilin®. Breeding territories were mapped and all sighted birds recorded on 4 hectare (10 acre) plots using methods similar to those described by Kendeigh, 1944, and Buckner and Turnock, 1965. On the day of insecticide treatment, plot searches were conducted to recover any sick, dead or dying birds.

Small Mammals: - Small mammal populations were assessed using standard snap-back traps. A total of 150 traps were employed on each plot. A center line 140 m (150 yd) long was established and flagged at 9 m (10 yd) intervals. Ten traps were then placed at 1 m (1 yd) intervals across the center line at each flag location. The trapping took place over a period of 3 consecutive nights resulting in a total of 450 trap nights for each plot. All specimens trapped were preserved in a 10% solution of formalin and returned to the laboratory for identification, sexing and dissecting.

Honey bees: - Newly purchased 1.4 kgm (3 lbs) packages of honey bees were set up at the headquarters apiary prior to their transfer to the Manitoulin Island treatment sites. When the colonies became well established with healthy queens and brood production well underway they were moved to treatment areas and located in openings in the forest. Five colonies were placed on each of the treatment and untreated control plots. When the bees became adjusted to the new sites, queens and brood were checked and impact monitoring equipment attached to each hive. Monitoring equipment employed consisted of a dead bee trap, a pollen trap, and an electronic counter placed at the hive entrance to measure the activity of bees entering or leaving the hive. Hives were weighed every second day. Prior to the application of the insecticide, small metal rings were embedded into comb containing eggs or newly hatched larvae (brood). Each ring contained approximately 250 undamaged cells and covered an area of 68 sq. cm (10.5 sq. in.) Two rings were placed in comb in the "treatment hives" and 1 ring in the "control hives". These marked areas were monitored until the eggs had hatched into brood, developed to the pupal stage and

emerged as adults (approximately 20 days). Eight days following treatment, the impact monitoring equipment was dismantled and the colonies transferred back to the headquarters apiary to prevent predation by black bears observed in the area. Queen and brood checks continued for several days after the transfer to assess any delayed effects resulting from the treatments.

Aquatic fauna: - Aquatic organisms were sampled from a stream located within the boundaries of the treatment plot and from an untreated stream approximately 8 km (5 mi) distant from the nearest treatment area. A series of five samples of bottom dwelling fauna were taken from the same section of stream before and after treatment using a Surber sampler (Surber, 1936). Sampling commenced at the bottom of the area and was repeated approximately every 5 m (16 ft) until all five samples had been collected. Pre-spray samples were taken just prior to treatment and post spray samples were collected three days after the insecticide had been applied. Samples were preserved in a 10% formalin solution and returned to the laboratory for sorting and identification.

Laboratory studies were conducted to study the effects of various concentrations of Dimilin® wettable powder (25% active ingredient) on juvenile crayfish and fish. Juvenile crayfish of the species Orconectes virilis (Hagen) were collected from Gore Bay, Manitoulin Island by capturing females carrying young. Ten juvenile crayfish 10 mm (3/8 in) in length were placed in 4 litres (0.9 gal) of control, 1 ppm, 10 ppm, 100 ppm and 1000 ppm aqueous solutions of Dimilin® wettable powder Observations on mortality and moulting success were made at frequent intervals. Juvenile brown bullheads Ictalurus nebulosus (Lesueur) 3 to 4

cm (1 to 1½ in) in length and small sunfish Lepomis gibbosus (Linnaeus) 8 to 10 cm (3 to 4 in) in length were collected from small ponds in Larose forest near Ottawa. Ten bullheads and four sunfish were placed in separate four litre test vessels of control, 10 ppm, 100 ppm and 500 ppm solutions of Dimilin® wettable powder. Mortality was recorded at frequent intervals up to two weeks exposure time.

RESULTS

Birds: - A total of 37 species of birds representing 13 families were recorded on the treatment plot (Table I) and 36 species representing 13 families recorded on the untreated control plot (Table II). The family Parulidae (wood warblers) was recorded most often on both plots with 15 species on the control and 12 species on the treatment. Turdidae (thrushes) were next with 4 and 5 species respectively and the family Fringillidae (finches) with 5 and 3 species respectively. Daily population fluctuations were relatively small and the post spray daily average populations show a slight decline on both plots probably as a result of having to carry out population censuses on days in inclement weather.

Territories of 5 species of the wood warbler, family (Parulidae), the black and white warbler, Mniotila varia (Linnaeus) (Fig. 1); the Nashville warbler, Vermivora ruficapilla (Wilson) (Fig. 2); the black-throated green warbler, Dendroica virens (Gamelin) (Fig. 3.); the black-burnian warbler, Dendroica fusca (Muller) (Fig. 4.); and the ovenbird, Seiurus aurocapillus (Linnaeus) (Fig. 5) and the territory of the veery Hylocichla fuscescens (Stephens) (Fig. 6) (family Turdidae) illustrate the location of pre and post treatment territories on each experimental plot. It is concluded that the treatment had no observable effect on the avian population.

Table I

Forest bird population census
Dimilin® treatment plot
Manitoulin Island, Ontario
June 4 to June 18, 1975

Family	Species	Pre-spray						Post-spray					
		June 4	June 6	June 7	June 8	June 9	Daily	June 11	June 12	June 14	June 16	June 18	Daily
		-6	-4	-3	-2	-1	ave	+1	+2	+4	+6	+8	ave
Picidae	Yellow-shafted Flicker	0	0	0	0	1	0.2	0	0	0	0	0	0.0
	Piliated Woodpecker	1	0	0	0	0	0.2	0	0	0	0	0	0.0
	Hairy Woodpecker	0	0	2	0	0	0.4	0	0	0	0	0	0.0
	Downy Woodpecker	2	0	0	0	0	0.4	0	0	0	0	0	0.0
Tyrannidae	Great-crested Flycatcher	2	2	0	0	2	1.2	0	0	0	2	0	0.4
	Least Flycatcher	0	0	0	0	0	0.0	0	0	0	2	0	0.4
	Eastern Wood Pewee	0	0	0	0	0	0.0	0	0	0	2	0	0.4
Corvidae	Blue Jay	2	1	0	0	1	0.8	3	0	5	0	0	1.6
	Common Raven	0	0	0	0	2	0.4	0	0	0	0	0	0.0
	Common Crow	0	0	1	0	0	0.2	0	0	0	0	0	0.0
Paridae	Black-capped Chickadee	0	0	0	0	2	0.4	2	0	0	2	2	1.2
Sittidae	Red-breasted Nuthatch	0	0	0	2	0	0.4	0	0	0	0	0	0.0
Turdidae	American Robin	2	3	5	3	2	3.0	1	3	4	2	3	2.6
	Wood Thrush	0	0	0	0	0	0.0	0	0	0	2	2	0.8
	Hermit Thrush	2	2	2	2	2	2.0	0	2	2	0	0	0.8
	Swainson's Thrush	4	0	0	0	0	0.8	0	0	0	0	0	0.0

Table I (Cont'd)

Family	Species	Pre-spray June 4	June 6	June 7	June 8	June 9	Daily	Post-spray June 11	June 12	June 14	June 16	June 18	Daily
		-6	-4	-3	-2	-1	ave	+1	+2	+4	+6	+8	ave
Turdidae (cont'd)	Veery	4	3	5	1	6	3.8	5	2	5	4	4	4.0
Sylviidae	Golden-crowned Kinglet	6	0	2	2	10	4.0	4	2	6	8	6	5.2
Bombycillidae	Cedar Waxwing	0	0	0	0	1	0.2	0	0	0	0	0	0.0
Vireonidae	Red-eyed Vireo	0	0	2	0	0	0.4	2	0	0	2	0	0.8
Parulidae	Black and White Warbler	4	6	4	2	6	4.4	10	6	6	8	8	7.6
	Tennessee Warbler	0	2	0	0	0	0.4	0	0	0	0	0	0.0
	Nashville Warbler	12	8	8	10	16	10.8	10	6	8	8	6	7.6
	Magnolia Warbler	6	4	4	6	4	4.8	0	0	2	4	0	1.2
	Cape May Warbler	0	0	0	0	2	0.4	8	6	4	4	8	6.0
	Myrtle Warbler	0	0	0	2	0	0.4	0	0	0	2	0	0.4
	Black-throated Green Warbler	6	4	6	6	10	6.4	8	6	8	4	6	6.4
	Blackburnian Warbler	2	4	6	4	2	3.6	0	0	4	4	0	1.6
	Chestnut-sided Warbler	0	0	0	0	4	0.8	0	0	0	0	0	0.0
	Bay-breasted Warbler	2	2	0	0	0	0.0	0	0	0	2	0	0.4
	Ovenbird	12	12	12	12	18	13.6	8	8	10	6	12	8.8

Table I (Cont'd)

Family	Species	Pre-spray June 4	June 6	June 7	June 8	June 9	Daily	Post-spray June 11	June 12	June 14	June 16	June 18	Daily
		-6	-4	-3	-2	-1	ave	+1	+2	+4	+6	+8	ave
Parulidae (cont'd)	Canada Warbler	0	0	2	2	4	1.6	2	0	2	0	2	1.2
	American Redstart	0	0	0	0	0	0.0	2	0	0	0	0	0.4
Icteridae	Brown-headed Cowbird	2	6	8	3	6	5.0	2	0	2	0	0	0.8
Fringillidae	Rose-breasted Grosbeak	2	2	2	0	2	1.6	0	0	0	4	0	0.8
	Purple Finch	2	0	2	0	1	1.0	0	0	0	2	0	0.4
	White-throated Sparrow	0	2	2	2	2	1.6	0	2	0	0	4	1.2
Unidentified	Species	0	0	0	0	0	0.0	0	1	0	1	0	0.4
	Totals	75	63	75	59	106	75.6	67	44	68	75	63	63.4

Table II

Forest bird population census
Dimilin® untreated control plot
Manitoulin Island Ontario
June 4 - June 18 1975

Family	Species	Pre-spray						Post-spray					
		June 4	June 6	June 7	June 8	June 9	Daily	June 11	June 12	June 14	June 16	June 18	Daily
		-6	-4	-3	-2	-1	ave	+1	+2	+4	+6	+8	ave
Trochilidae	Ruby-throated Hummingbird	0	0	0	0	2	0.4	0	0	0	0	0	0.0
Picidae	Yellow-shafted Flicker	2	0	0	0	0	0.4	0	2	0	2	0	0.8
	Hairy Woodpecker	0	0	0	0	0	0.0	0	0	0	0	2	0.4
Tyrannidae	Great-creasted Flycatcher	2	2	2	0	0	1.2	0	2	2	2	0	1.2
Corvidae	Blue Jay	0	0	0	2	0	0.4	0	2	0	0	0	0.4
	Common Crow	0	0	0	0	0	0.0	0	0	0	3	0	0.4
Paridae	Black-capped Chickadee	2	0	2	0	2	1.2	0	2	0	2	2	1.2
	Boreal Chickadee	0	0	0	0	0	0.0	0	0	0	0	2	0.4
Troglodytidae	Winter Wren	0	2	0	0	2	0.8	2	0	0	2	2	1.2
Mimidae	Catbird	0	0	0	0	2	0.4	0	2	2	2	2	1.6
Turdidae	American Robin	2	3	2	1	1		2	1	0	2	1	1.2
	Wood Thrush	2	0	0	0	0	0.4	0	0	0	0	0	0.0
	Hermit Thrush	0	0	0	2	0	0.4	2	2	0	2	0	1.2
	Veery	0	0	0	0	0	0.0	0	0	0	2	2	0.8
Sylviidae	Golden-crowned Kinglet	0	6	2	4	4	3.2	2	0	4	0	2	1.6

Table II (Cont'd)

Family	Species	Pre-spray June 4	June 6	June 7	June 8	June 9	Daily	Post-spray June 11	June 12	June 14	June 16	June 18	Daily
		-6	-4	-3	-2	-1	ave	+1	+2	+4	+6	+8	ave
Vireonidae	Red-eyed Vireo	4	2	0	0	2	1.6	4	2	4	2	4	3.2
Parulidae	Black and White Warbler	10	2	2	4	6	4.8	0	4	8	6	2	4.0
	Tennessee Warbler	0	0	2	2	4	1.6	0	2	2	2	0	1.2
	Nashville Warbler	0	2	2	0	2	1.2	0	2	2	2	2	1.6
	Magnolia Warbler	2	0	0	0	0	0.4	2	0	0	0	0	0.4
	Cape May Warbler	2	2	0	2	2	1.6	0	0	0	0	0	0.0
	Myrtle Warbler	0	2	0	0	0	0.4	2	0	0	0	0	0.4
	Black-throated Green Warbler	4	4	4	0	4	3.2	4	2	4	4	6	4.0
	Blackburnian Warbler	2	6	2	4	2	3.2	0	4	2	2	2	2.0
	Chestnut-sided Warbler	4	2	0	2	0	1.6	2	2	2	4	2	2.4
	Bay-breasted Warbler	0	0	2	0	0	0.4	0	0	0	0	0	0.0
	Ovenbird	6	6	2	8	4	5.2	4	6	4	9	2	5.0
	Northern Waterthrush	2	2	2	2	2	2.0	0	2	2	2	2	1.6
	Connecticut Warbler	0	0	0	2	2	0.8	0	0	0	0	0	0.0

Table II (Cont'd)

Family	Species	Pre-spray June 4	June 6	June 7	June 8	June 9	Daily	Post-spray June 11	June 12	June 14	June 16	June 18	Daily
		-6	-4	-3	-2	-1	ave	+1	+2	+4	+6	+8	ave
Parulidae (cont'd)	Mourning Warbler	0	0	0	2	0	0.4	0	0	0	2	0	0.4
	Canada Warbler	4	6	2	2	2	3.2	0	2	2	4	0	1.6
Icteridae	Brown-headed Cowbird	2	4	0	2	0	1.6	0	0	0	0	0	0.0
Fringillidae	Rose-breasted Grosbeak	0	2	2	0	2	1.2	0	2	0	2	0	0.8
	Idigo Bunting	0	0	0	0	0	0.0	2	0	0	2	0	0.8
	Purple Finch	0	0	4	0	2	1.2	0	2	0	2	2	1.2
	White-throated Sparrow	0	2	0	0	0	0.4	2	0	0	2	0	0.8
Totals		52	59	34	41	49	47.0	30	46	40	66	39	44.2

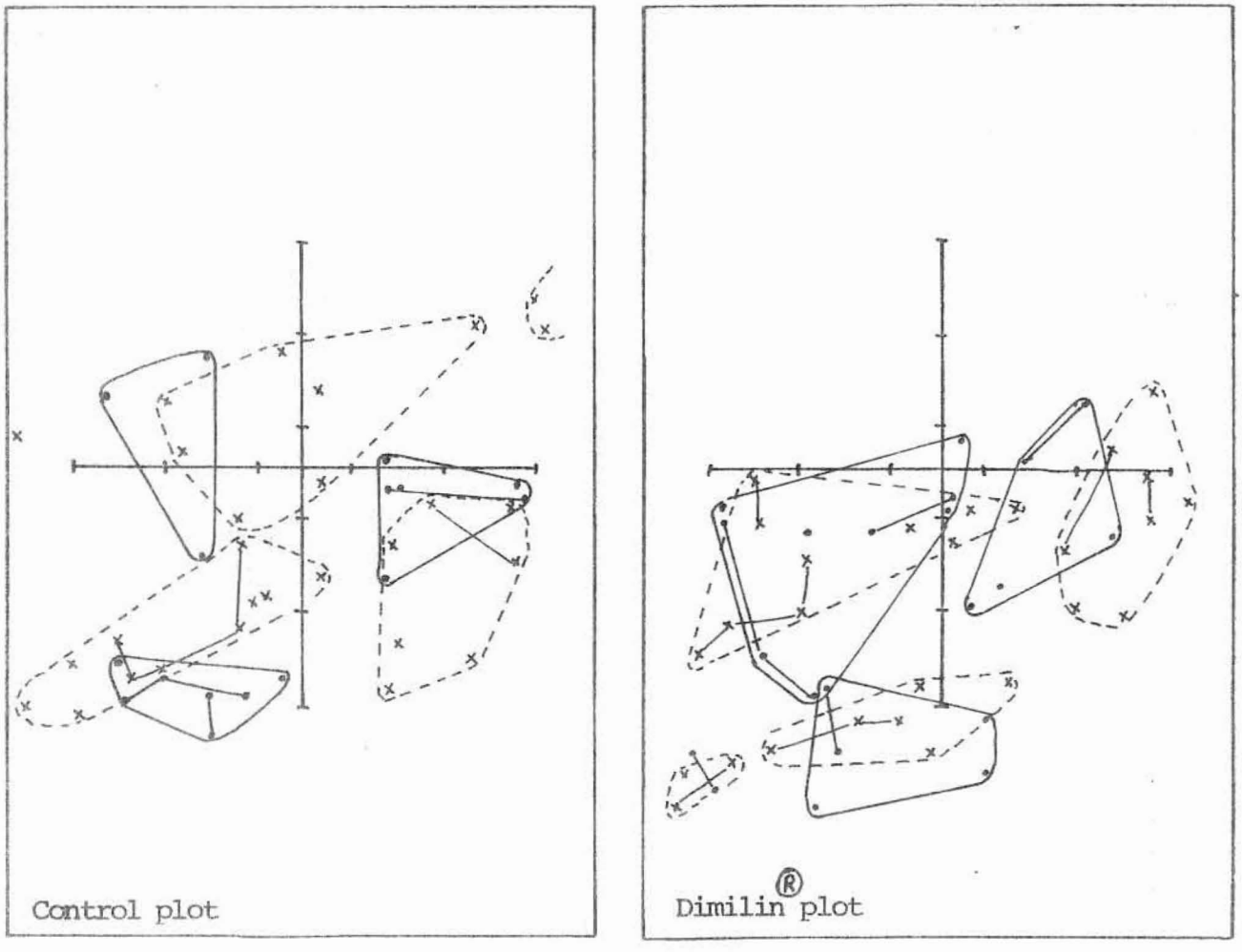

Figure 1 - Pre and post spray territories of Mniotala varia (Linnaeus) on the Dimilin® treated and untreated control plots.

. solid line = pre spray territory boundary

x broken line = post spray territory boundary

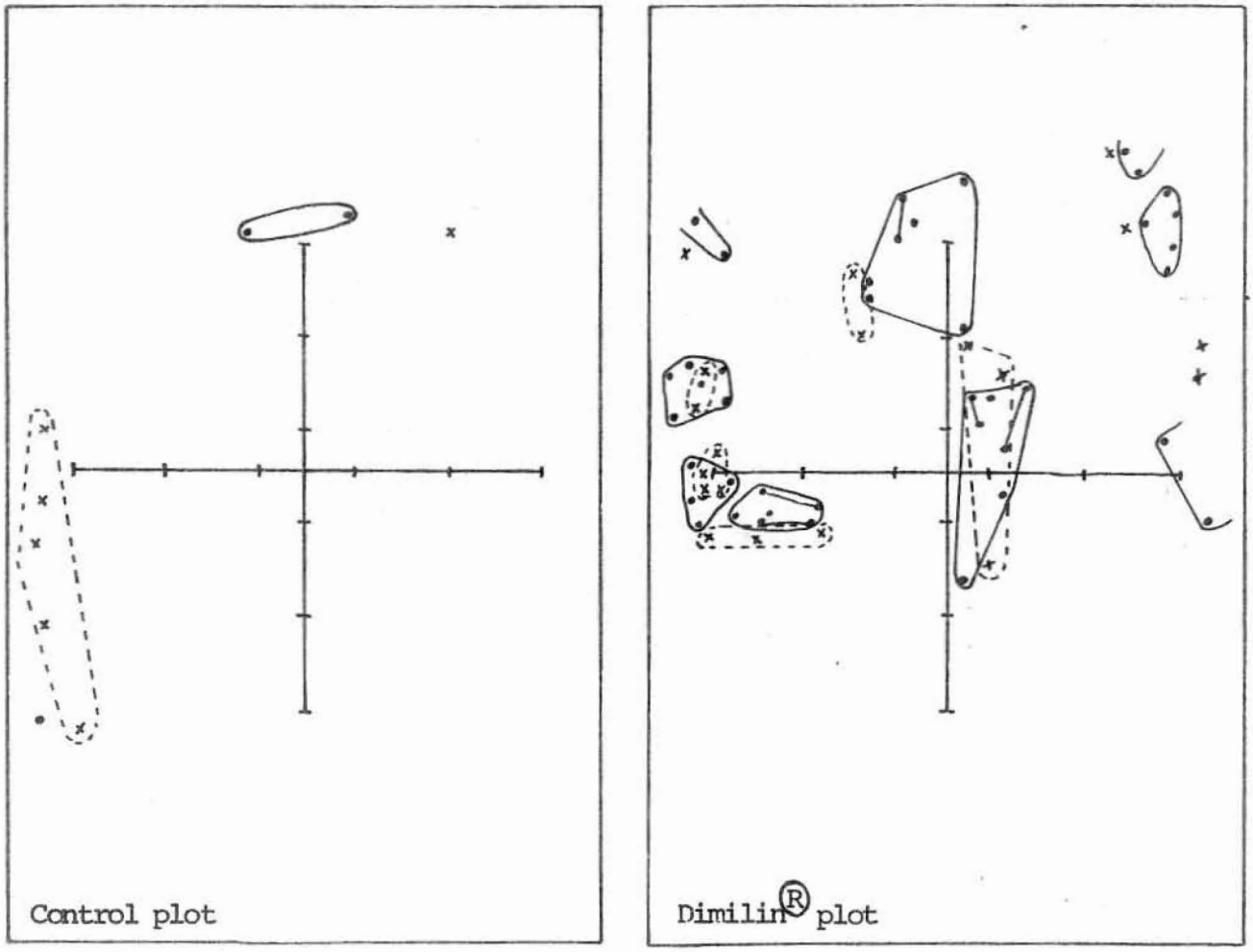

Figure 2 Pre and post spray territories of Vermivora ruficopilla (Wilson) on Dimilin® treated and untreated control plots.

. solid line = pre spray territory boundary

x broken line = post spray territory boundary

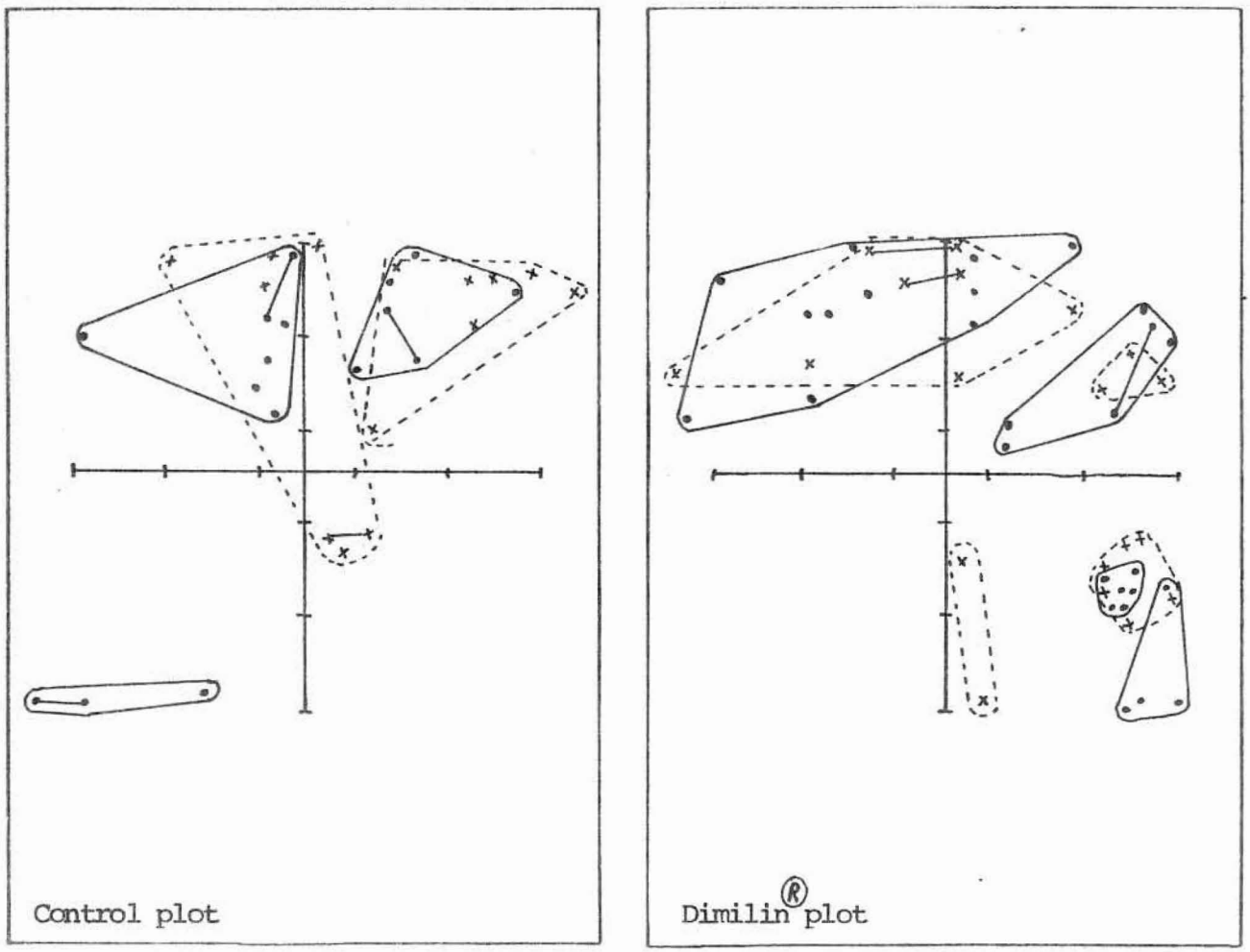

Figure 3 Pre and post spray territories of Dendroica virens (Gmelin) on the Dimilin® treated and untreated control plots.

. solid line = pre spray territory boundary

x broken line = post spray territory boundary

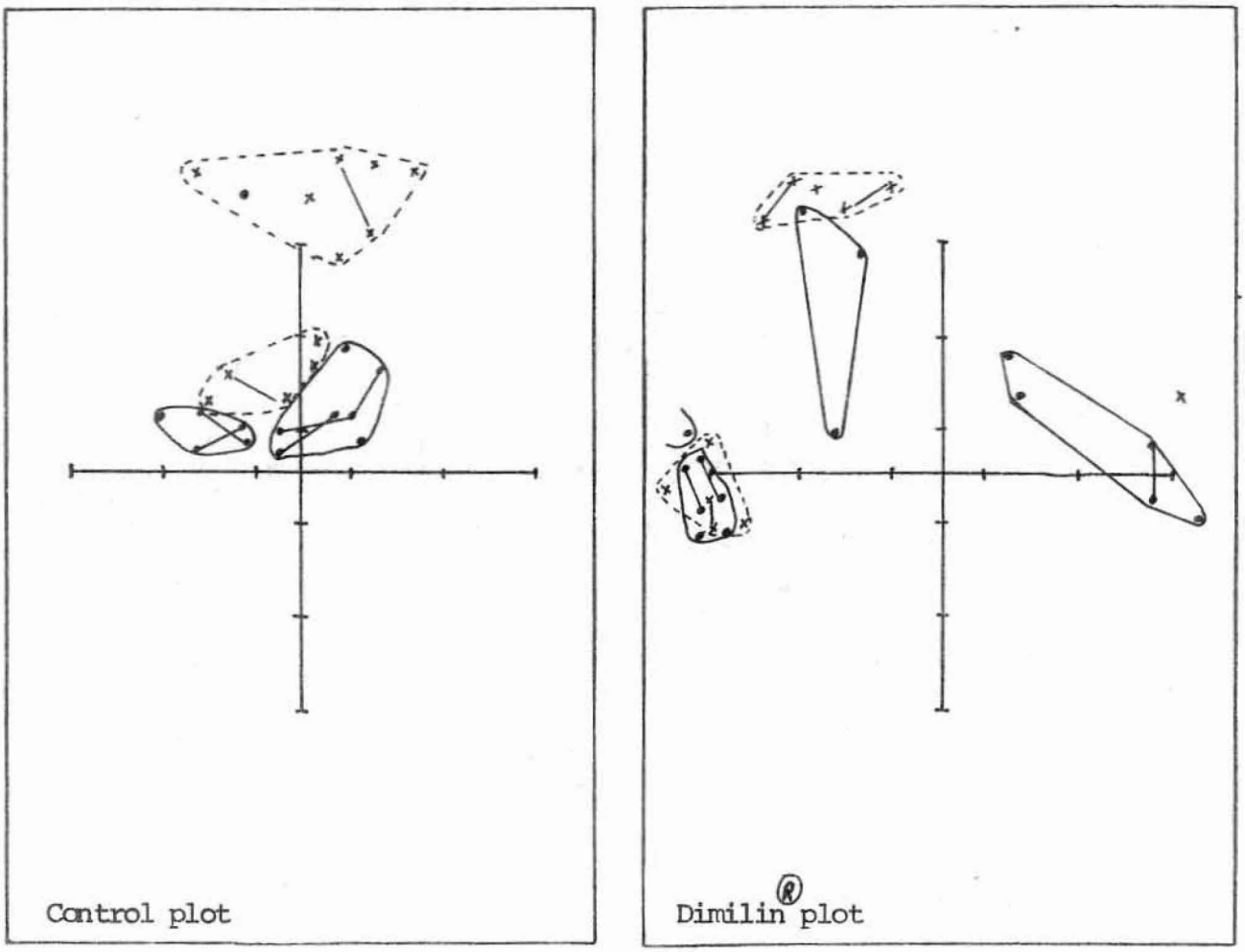

Figure 4 Pre and post spray territories of Dendroica fusca (Muller) on Dimilin® treated and untreated control plots.

. solid line = pre spray territory boundary

x broken line = post spray territory boundary

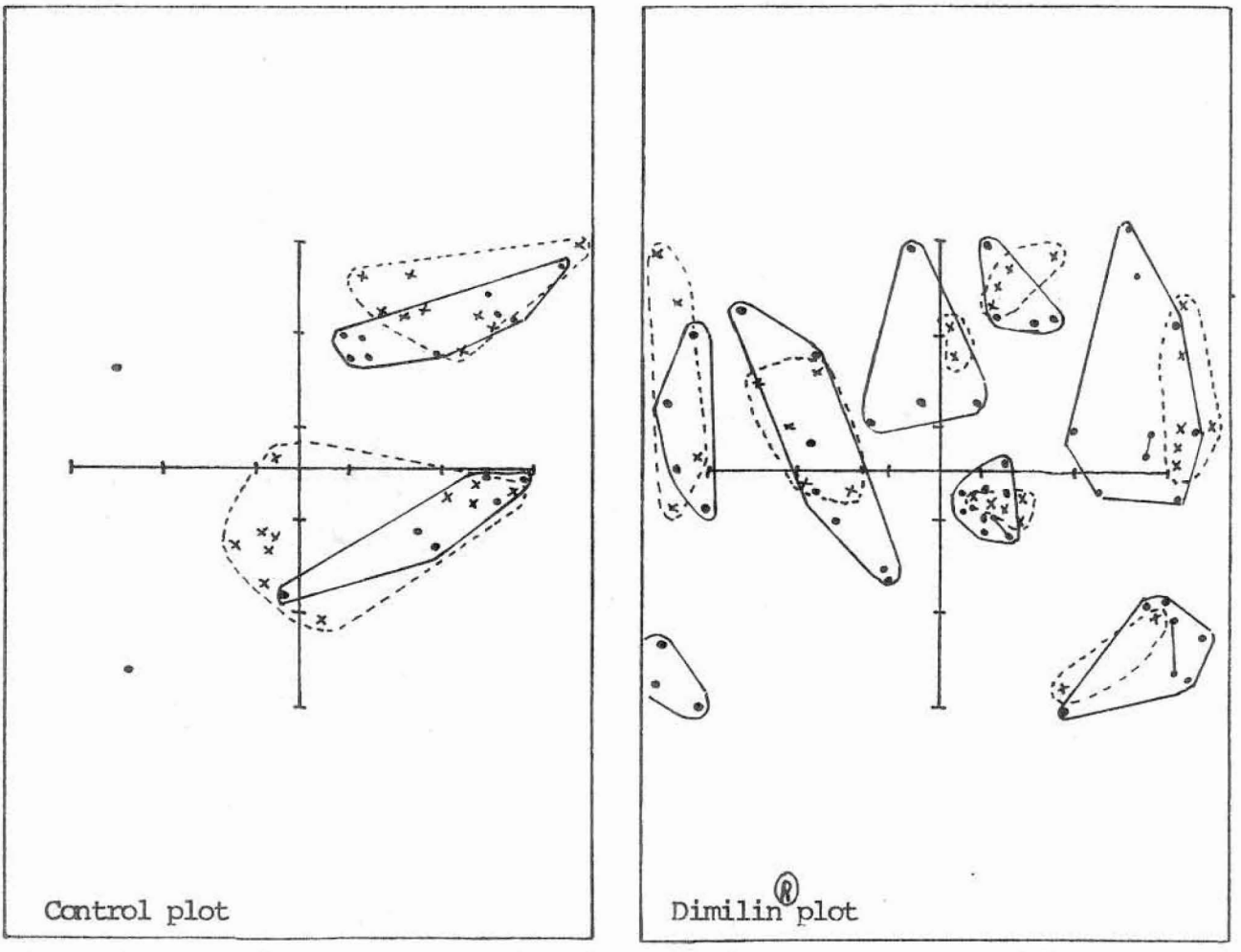

Figure 5. Pre and post spray territories of Seiurus aurocopillus (Linnaeus) on Dimilin® treated and untreated control plots.

. solid line = pre spray territory boundary

x broken line = post spray territory boundary

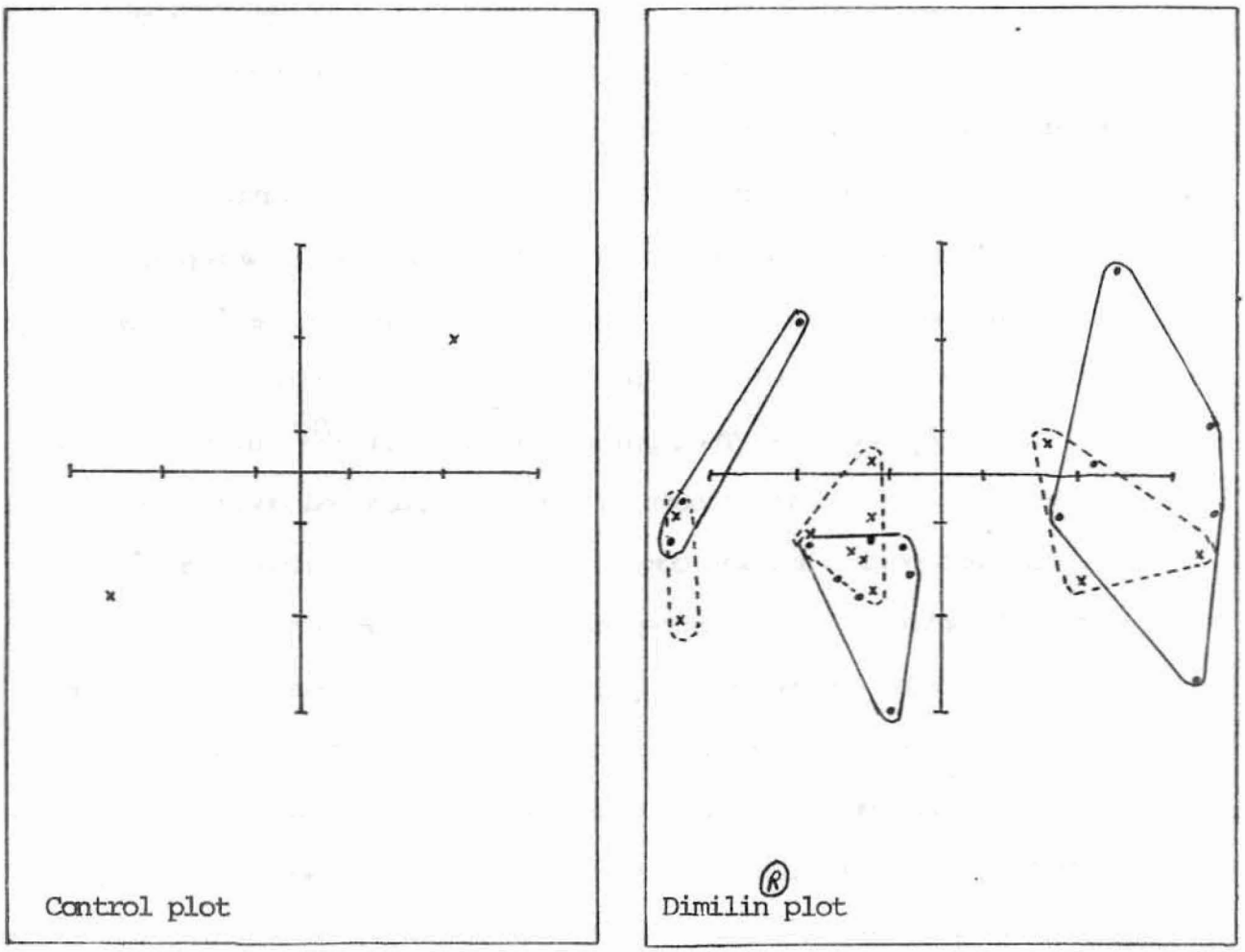

Figure 6 Pre and post spray territories of Hylocichla fuscescens (Stephens) on Dimilin® treated and untreated control plots.

. solid line = pre spray territory boundary

x broken line = post spray territory boundary

Small Mammals: - Very low numbers of small mammals were trapped on Manitoulin Island. A single specimen of Peromyscus maniculatus (Wagner) was taken from the treatment plot and two specimens of the same species from the untreated control plot (Table III). The female mouse taken from the treatment plot was pregnant indicating normal breeding.

The low populations are a result of a normal low population density at that particular time and in that particular habitat. No evaluation of impact could be made under these conditions.

Honey bees: - The application of Dimilin® did not present a hazard to the field force bees as mortality remained constant throughout the treatment period and was comparable with that obtained from the untreated control hives. Discrepancies in daily pollen collection and activity recorded can be attributed to the variable weather conditions encountered throughout the test period (Table IV). The metal rings embedded in the comb containing eggs and young brood showed that the eggs hatched and the brood proceeded through the larval stage and finally pupated and emerged as young nurse bees. The empty cells were then filled with honey and capped with wax, (Table V). On August 20, all treated colonies were checked for overall strength and honey production and were found to be as healthy as colonies remaining in the headquarters apiary. Honey production was equivalent in treated and control hives.

TABLE III

Small mammal populations trapped on

Dimilin® treatment and control plots

Manitoulin Island Ontario

July 1975

Plot	Species	Males			Females					
						Adults			Total	Total
		Sub Adult	Adult	Total Males	Sub Adult	Pregnant	Pregnant with scars	Scars only	Females	Animals
Dimilin®	P. maniculatus	0	0	0	0	1	0	0	1	1
Control	P. maniculatus	0	2	0	0	0	0	0	0	2

TABLE IV

Pesticide impact measurements on honey bee colonies on Dimilin® treatment and untreated plots
Manitoulin Island Ontario
June, 1975

Days from treatment	Untreated control plot				Dimilin® treatment plot				Remarks
	Adult mortality	Adult activity trips/day	Pollen collected (gms)	Hive weights (kg)	Adult mortality	Adult activity trips/day	Pollen collected (gms)	Hive weights (kg)	
-7	2	32512	23.5	17.2	4	27008	75.1	17.9	
-6	2	20736	18.3		5	8576	30.4		rain in am, cloud & fog
-5	3	18560	2.1	18.6	4	10496	4.2	17.4	heavy rain
-4	3	24960	33.4		4	30976	46.0		windy - showers
-3	3	20352	41.7	18.1	2	17280	42.0	17.4	overcast, cool, drizzle in pm
-2	4	41088	87.0		3	40960	200.0		sunny & windy, high of 18°C.
-1	4	36352	93.0	17.7	4	34176	142.6	17.4	warm & sunny, high of 23°C.
+0	3	61312	39.7		6	31744	77.7		sunny, warm, high of 23°C
+1	3	23040	27.3	16.8	3	52736	102.0	17.5	sunny in am, winds light, 20°C
+2	1	23808	38.3		4	16512	31.2		cloudy, high of 18°C
+3	1	29056	13.7	18.1	2	30720	10.1	17.6	showers in am, 18°C
+4	2	45184	60.0		6	40448	136.6		clear & windy, high of 20°C
+5	2	14208	4.0	18.1	4	35072	78.4	16.8	heavy showers in am, 18°C
+6	2	44288	52.5		3	36480	57.8		cloudy, cool in am, sunny pm
+7	4	20224	2.3	17.2	3	13440	1.5	17.2	rain all day, 20°C

TABLE V

Results of monitoring "brood rings" placed in honey bee colonies on Dimilin® treatment and untreated plot
Manitoulin Island, Ontario
June, 1975

Days from treatment	Control plot		Dimilin® plot		
	Hive no.	Hoop "A"	Hive no.	Hoop "A"	Hoop "B"
-1	25	7/8 eggs, 1/8 empty	30	all eggs	¾ eggs, 1/8 larvae, 1/8 empty
	22	3/4 eggs, 1/4 larvae	23	all eggs	7/8 eggs, 1/8 empty
	27	3/4 eggs, 1/4 larvae	26	2/3 eggs 1/3 empty	3/4 eggs, 1/4 empty
	31	3/4 eggs, 1/4 larvae	32	queenless	
	16	3/4 eggs, 1/8 larvae, 1/8 capped	21	7/8 eggs, 1/8 larvae	3/4 larvae, 1/8 eggs, 1/8 empty
+6	25	1/2 capped, 1/2 empty	30	7/8 capped brood, 1/8 empty	1/2 capped brood, 1/2 empty
	22	3/4 capped, 1/8 larvae, 1/8 empty	23	2/3 capped brood 1/3 larvae	2/3 capped, 1/6 larvae, 1/6 empty
	27	1/2 capped, 1/4 larvae, 1/4 empty	26	1/3 capped 1/3 larvae 1/3 empty	1/4 capped, 1/2 larvae, 1/4 empty
	31	7/8 capped 1/8 empty	32	queenless	
	16	7/8 capped 1/8 empty	21	2/3 capped larvae 1/6 empty 1/6 larvae	2/3 capped, 1/3 empty
+14	25	1/3 capped 2/3 honey	30	7/8 capped 1/8 honey	1/2 capped, 1/4 larvae, 1/4 empty
	22	7/8 capped 1/8 honey & pollen	23	7/8 capped 1/8 honey	7/8 capped, 1/8 honey
	27	----	26	3/4 capped 1/4 honey	3/4 capped, 1/4 honey
	31	2/3 capped 1/3 honey	32	queenless	
	16	7/8 capped 1/8 honey	21	7/8 capped 1/8 honey	2/3 capped, 1/6 empty, 1/6 honey
+17	25	¼ larvae ¼ honey ¼ eggs ¼ empty	30	all honey	1/8 capped, 7/8 honey
	22	2/3 honey 1/3 empty	23	1/2 capped 1/3 honey 1/6 empty	2/3 capped, 1/6 honey, 1/6 empty
	27	1/3 capped, 1/2 empty 1/6 eggs	26	¾ honey, ¼ capped	1/3 capped, 2/3 honey
	31	3/4 eggs, 1/4 larvae	32	queenless	
	16	1/3 capped, 2/3 honey	21	5/6 honey, 1/6 capped	all honey

Aquatic fauna: - Bottom fauna populations in the Dimilin® treatment and control streams are presented in Table VI. The type of organisms present in the treatment stream (snails, tadpoles, amphipods) reflect its sluggish flowing, swampy nature. The control stream is somewhat larger and faster flowing with a very limited bottom fauna and few of the slow water forms found in the treatment stream. The Dimilin® treatment did not significantly alter the total number of organisms present in the treatment stream but appears to have reduced amphipod and aquatic beetle larvae populations. The absence of amphipods and low beetle larvae populations in the control stream make it difficult to assess natural changes in the populations of these two groups over the sampling period. Both of these groups could be vulnerable to a chitin synthesis blocking compound such as Dimilin® as amphipods moult frequently and coleoptera larva complete their development into adults at this period of the year. Several of the amphipods taken in the pre-treatment Surber samples were carrying young and these may be extremely sensitive to Dimilin® during their period of rapid growth and frequent moulting. In examining the bottom samples collected, it was noted that small numbers of copepods and ostracods were seen in pre-spray Surber samples from the treatment stream but none were observed in post-spray bottom samples. This indication of an effect on zooplankton in the treatment stream should be tested by further studies employing more suitable methods for studying plankton than Surber sampling.

The results from the laboratory tests on the effects of various concentrations of Dimilin® on juvenile crayfish and fish were complicated by difficulties in keeping concentrations of Dimilin® of 100 ppm or

greater in solution. At these high concentrations the solution in the test vessels remained turbid and a fine precipitate gradually settled on the bottom of the containers. This would indicate that the actual concentration at these levels were less than the calculated concentrations. The survival and moulting success of juvenile crayfish in the various concentrations of Dimilin® is shown in Table VII. Mortality amongst the control group related and unrelated to moulting were both greater or equal to mortality suffered by the Dimilin® exposed groups. Successful moults occured over the entire range of Dimilin® concentrations. One crayfish in the 100 ppm group successfully moulted and survived for over five days during which its carapace appeared to harden normally from its soft post-moult condition.

Results from the bullhead and crayfish groups revealed no toxic effects on these species caused by exposure to up to 500 ppm (125 ppm active ingredient) Dimilin® wettable powder. No mortality occured over sixteen days to the bullheads exposed to the highest Dimilin® concentration. Mortality among the other groups was light and similar to that experienced by the control group except for the lowest Dimilin® concentration group where heavy mortality was caused by severe fungus growth on many of the fish. No mortality occured among the sunfish until the ninth day of exposure and mortality from this point until the end of the experiment seven days later, was similar among the control and exposed groups.

TABLE VI

Bottom fauna populations in the Dimilin® treatment and control streams as numbers and standard deviations of organisms/sq. ft. (0.092 sq. m.) Manitoulin Island, June 5 to 15, 1975

	Treatment stream						Control stream					
	June 5			June 15			June 6			June 15		
Ephemeroptera	---			---			---			0.2	±	0.5
Trichoptera	0.2	±	0.5	---			1.0	±	0.8	0.8		1.5
Odonata	1.0	±	1.4	0.2	±	0.5	---			---		
Coleoptera	5.5	±	1.7	0.8	±	1.1	0.2	±	0.5	0.8	±	1.1
Diptera-Chironomidae	1.2	±	1.5	2.5	±	1.7	2.0	±	2.8	6.2	±	10.5
Diptera-Simuliidae	0.2	±	0.5	---			---			0.2	±	0.5
Diptera-Tipulidae	0.5	±	1.0	0.5	±	0.6	---			0.8	±	1.1
Other Diptera	---			---			---			1.0	±	1.2
Nematoda	---			---			0.2	±	0.5	---		
Oligochaeta	3.8	±	2.5	13.8	±	9.8	4.5	±	3.4	8.5	±	8.9
Hirudinea	---			---			0.5	±	0.6	0.5	±	0.6
Amphipoda	11.5	±	6.8	1.8	±	3.5	---			---		
Isopoda	0.5	±	0.6	0.2	±	0.5	0.8	±	1.1	---		
Gastropoda	23.2	±	16.9	13.5	±	5.1	---			0.2	±	0.5
Pelecypoda	5.2	±	5.5	16.5	±	18.5	5.8	±	9.0	9.5	±	7.0
Amphibia	3.5	±	1.3	1.2	±	1.0	---			---		
Pisces	1.0	±	0.8	0.2	±	0.5	3.0	±	5.4	1.2	±	1.5
Total	57.5	±	25.2	51.2	±	20.8	18.0	±	10.4	30.0	±	7.9

TABLE VII

Survival and moulting success of juvenile crayfish Orconectes virilis in various concentrations of Dimilin® wettable powder

Elapsed time	Control	1 ppm (0.25 ppm a.i.)	10 ppm (2.5 ppm a.i.)	100 ppm (25 ppm a.i.)	1000 ppm (250 ppm a.i.)
0 hr	10	10	10	10	10
8 hr	10	10	10	10	10
16 hr	10	6 ++ΔΔ	10	10	10
24 hr	7+ΔΔ	6	10	10	9Δ
36 hr	6+	5Δ	9Δ	10	8Δ
48 hr	6	5	9	8ΔΔ	7Δ
60 hr	6	5	7ΔΔ	6ΔΔ *	6Δ *
72 hr	6	5	7	6	3ΔΔ
4 days	4ΔΔ	5 *	6Δ	4ΔΔ	1ΔΔ
5 days	1ΔΔΔ	4 *	6	4	1
6 days	0+	2Δ	4Δ+	4	1
7 days	-	1Δ	4	4	1
8 days	-	1	4	3	1
9 days	-	0+	4	3	1
10 days	-	-	4	3	1
13 days	-	-	3Δ	1Δ+	1
16 days	-	-	2Δ	0Δ	1

* - Successful moult, animal survived for period shown.

\+ - Unsuccessful moult, animal died during moult.

Δ - Mortality apparently unrelated to moulting.

CONCLUSIONS

The monitoring studies conducted revealed no immediate or short term effects of the Dimilin® treatment on small forest songbirds or colonies of domestic honey bees. Small mammal populations in the treatment area were too low to assess insecticide impact. The treatment did appear to affect amphipod and coleoptera larva populations within the treatment stream and incidental observations on the presence of zooplankton in pre and post spray bottom samples indicate that this group of aquatic organisms may have been affected. Laboratory studies revealed no effect of high concentrations of Dimilin® on juvenile crayfish, bullheads or sunfish. Further studies of the short and long term effects of this compound on non-target organisms are recommended, particularly with respect to components of aquatic ecosystems.

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