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Preliminary Report on the Environmental  
Impact of Nuclear Polyhedrosis Virus  
Upon Selected Forest Fauna

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## ABSTRACT

The impact of the biological insecticide nuclear polyhedrosis virus upon selected components of a forest ecosystem was studied. The insecticide was aeriaily applied to 160 hectares (400 acres) on June 3 using a small fixed wing aircraft fitted with micronairs at the rate of  $247.5 \times 10^9$  PIB/ha ( $100 \times 10^9$  PIB/acre). Native populations of song birds, small mammals, honey bees and aquatic organisms were monitored for impact associated with the virus application. No immediate or short term impact on any of these groups was attributed to the virus treatment.

## INTRODUCTION

The nuclear polyhedrosis virus of spruce budworm *Choristoneura fumiferana* (Clem) has been undergoing field testing for several years to study its potential for controlling this important forest pest species. In 1975, an experimental aerial application of this material was applied to 160 hectares (400 acres) of spruce budworm infested forest on Manitoulin Island at the rate of 247.5 polyhedral inclusion bodies/ha ( $100 \times 10^9$  PIB/acre). The Environmental Impact Section of the Chemical Control Research Institute monitored this treatment for adverse side effects on small forest song birds, small mammals, colonies of domestic honey bees *Apis mellifera* L. and aquatic invertebrates.

## METHODS

Birds:- Songbird populations were assessed on 4 hectare (10 acre) plots located on treated and untreated areas. Parallel lines 40 meters (2 chains) apart were flagged out and all birds either sighted or heard were recorded on plot maps. Populations were censused in the early morning when maximum activity is encountered. The census started 5 days prior to treatment and continued for 5 days after treatment. Plot searches for sick or distressed birds was carried out the day of application.

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 80 meters long (90 yards) was established with plastic flagging tape marking 9 meter (10 yard) intervals.

Standard snap-back kill mouse traps were located on the center line and at 1 meter (approx. 1 yard) intervals at right angles across the center line. The trapping took place over a period of 3 consecutive nights resulting in a total of 450 trap nights. All small mammal specimens trapped were preserved in a 10% formalin solution and returned to the laboratory for identification, sexing and dissection.

Honey bees:- Newly purchased 1.4 kg (3 lb.) packages of honey bees were set up in the headquarters apiary prior to their transfer to Manitoulin Island. When the colonies had become well established with healthy queens and with egg and brood production well underway, they were transferred to the Manitoulin Island experimental sites and located in openings in the forest. Five colonies were placed on each of the virus treatment and untreated plots. When the bees had adjusted to the new sites, queens and brood were checked

for any damage sustained during the transfer and environmental monitoring equipment installed on each colony. Monitoring equipment consisted of a dead bee trap attached to the outside entrance, a pollen trap which collects approximately 40% of the pollen brought into the hive, an electronic counter which counts bees leaving or entering the hive and a scale for taking hive weights. Just prior to the insecticide treatment small metal rings were embedded into comb wax containing eggs or newly hatched larvae. Each ring contained approximately 250 undamaged cells encompassing an area of 68 sq. cm (10.5 sq. in). Two rings were placed in each hive on the treated plot and a single ring in the untreated hives. The rings were monitored for a period of 22 days after application of the virus to ensure that an egg to adult life cycle was completed without interruption by the virus. Seventeen days after treatment the monitoring equipment was removed and the colonies transferred back to the headquarters apiary in order to prevent predation and damage by black bears observed in the area. Queen and brood checks continued for several days after transfer to assess any delayed effects of the virus treatment.

Aquatics:- Aquatic organisms were sampled from a stream located within the boundaries of the virus treated plot and from a stream in an untreated area located approximately 8 kilometers (5 miles) distant from the experimental plot.

A series of 5 samples of bottom dwelling fauna were taken from the same area of the treatment stream before and after the application of virus using a Surber sampler (Surber, 1936). Only post treatment samples were taken from the control stream. Sampling commenced at the bottom of the area and was repeated approximately every 5 meters (16 feet) upstream until all

5 samples had been taken. Pre-spray samples were collected just prior to treatment and the post-spray samples were taken 3 days after treatment. Samples were preserved in a 10% formalin solution and returned to the laboratory for sorting and identification.

#### RESULTS

Birds:- A total of 42 species representing 15 families of forest inhabiting birds were recorded on the virus treatment plot (Table I) and 39 species representing 14 families recorded on the untreated control plot (Table II). The family Parulidae (the warblers) comprised the largest group encountered with 15 species recorded on both treated and untreated plots. The family Fringillidae (sparrows, finches, grosbeaks etc.) recorded 5 species on the treatment plot and 4 on the untreated followed by the family Turdidae (thrushes) with 3 and 4 species respectively.

Birds were very vocal coupled with a great deal of activity during the census the day following the application. Activity declined thereafter to a level recorded prior to application. The reason for this sudden increase is not known but is thought not to be related to the treatment. A decline in singing and activity occurred on the 4th day after treatment on the untreated control plot. This decrease was probably caused by the showers and windy conditions encountered during this census. The territories of 4 species of birds, the golden-crowned kinglet, *Regulus satrapa* Lichtenstein (Fig. 1), the nashville warbler, *Vermivora ruficapella* (Wilson) (Fig. 2), the black-throated green warbler *Dendroica virens* (Gmelin) (Fig. 3) and the ovenbird *Sevirus aurocapillus* (Linnaeus) (Fig. 4) are illustrated to show the location of pre and post spray territories on each

plot. The mapping of territories does not reveal any disturbance of populations or shift in territories attributable to the application of the virus insecticide. The solid lines on the plot map indicate pre-spray territories, the broken line the post-spray territories.

Table I

Forest bird population census  
Nuclear polyhedrosis virus treated plot  
Manitoulin Island Ontario  
May 29 - June 9, 1975

Family	Species	Pre-spray					Daily ave	Post-spray					Daily ave
		May 29	May 30	May 31	June 1	June 2		June 4	June 6	June 7	June 8	June 9	
		-5	-4	-3	-2	-1		+1	+3	+4	+5	+6	
Tetraonidae	Ruffed Grouse	0	0	0	0	0	0.0	2	0	0	1	0	0.6
Trochilidae	Ruby-throated Hummingbird	1	0	0	0	0	0.2	0	0	0	0	0	0.0
Picidae	Yellow-shafted Flicker	0	0	2	4	2	1.6	5	4	1	0	0	2.0
	Yellow-bellied Sapsucker	0	0	0	0	0	0.0	0	0	2	0	0	0.5
	Hairy Woodpecker	0	0	0	0	0	0.0	2	0	0	0	2	0.8
Tyrannidae	Great-crested Flycatcher	0	0	0	2	2	0.8	2	2	1	2	0	1.4
	Eastern Phoebe	0	0	0	0	0	0.0	0	0	0	4	0	0.8
	Least Flycatcher	0	0	2	0	0	0.4	0	0	0	0	0	0.0
Corvidae	Blue Jay	1	1	1	0	0	0.6	1	0	0	0	0	0.2
	Common Crow	2	0	1	0	1	0.8	0	0	0	0	0	0.0
Paridae	Black-capped Chickadee	2	0	0	2	0	0.5	0	2	2	0	0	0.8
Sittidae	Red-breasted Nuthatch	0	0	0	2	0	0.4	2	0	0	0	0	0.4
Troglodytidae	House Wren	0	0	0	2	0	0.4	0	0	0	0	0	0.0
Mimidae	Catbird	0	0	0	0	0	0.0	2	0	0	0	0	0.4
	Brown Thrasher	0	0	0	0	0	0.0	2	0	0	2	0	0.8



Table I (Cont'd)

Family	Species	Pre-spray					Post-spray						
		May 29	May 30	May 31	June 1	June 2	Daily ave.	June 4	June 6	June 7	June 8	June 9	Daily ave.
		-5	-4	-3	-2	-1		+1	+3	+4	+5	+6	
Turdidae	American Robin	0	0	1	1	0	0.4	2	0	2	1	2	1.4
	Wood Thrush	0	0	0	0	0	0.0	0	2	2	2	2	1.6
	Hermit Thrush	4	2	0	2	2	2.0	0	0	2	2	0	0.8
Sylviidae	Golden-crowned Kinglet	4	2	4	6	4	4.0	9	6	8	10	8	8.2
	Ruby-crowned Kinglet	2	0	2	2	4	2.0	2	0	0	0	2	0.8
Vireonidae	Red-eyed Vireo	4	0	0	4	4	2.4	6	2	4	2	2	3.2
Parulidae	Black and White Warbler	0	0	0	4	6	2.0	4	0	0	0	0	0.8
	Nashville Warbler	12	6	14	12	18	12.4	24	16	18	22	4	16.8
	Magnolia Warbler	0	0	2	2	2	1.2	2	2	2	4	2	2.4
	Cape May Warbler	0	2	0	0	2	0.8	2	0	0	0	2	0.8
	Myrtle Warbler	0	0	0	0	0	0.0	0	0	0	2	0	0.4
	Black-throated Green Warbler	2	0	0	2	0	0.8	2	2	2	0	0	0.6
	Blackburnian Warbler	0	0	0	0	0	0.0	2	2	2	4	2	2.4
	Chestnut-sided Warbler	2	2	2	0	2	1.6	2	4	4	2	4	3.2
	Bay-breasted Warbler	0	0	2	0	0	0.4	0	0	0	0	0	0.0

Table I (Cont'd)

Family	Species	Pre-spray					Daily ave.	Post-spray					Daily ave.
		May 29	May 30	May 31	June 1	June 2		June 4	June 6	June 7	June 8	June 9	
		-5	-4	-3	-2	-1		+1	+3	+4	+5	+6	
Parulidae (cont'd)	Blackpoll Warbler	0	0	0	0	2	0.4	0	0	0	0	0	0.0
	Ovenbird	8	8	10	8	11	9.0	12	12	8	18	8	11.6
	Mourning Warbler	0	0	0	0	0	0.0	0	0	0	2	2	0.8
	Yellowthroat	0	0	0	4	2	1.2	0	0	0	0	0	0.0
	Canada Warbler	0	0	0	0	0	0.0	2	0	0	0	0	0.4
	American Redstart	0	0	2	0	0	0.4	0	0	0	0	0	0.0
Icteridae	Brown-headed Cowbird	2	0	2	6	0	2.0	2	0	1	2	2	1.4
Fringillidae	Indigo Bunting	0	0	0	0	0	0.0	0	2	0	0	0	0.4
	Purple Finch	2	3	4	2	4	3.0	2	0	4	4	0	2.0
	Slate-coloured Junco	0	0	0	0	0	0.0	0	0	2	0	0	0.4
	Chipping Sparrow	2	0	2	0	0	0.8	4	0	2	4	4	2.8
	White-throated Sparrow	4	2	8	6	2	4.4	12	3	4	3	2	4.8
Totals		54	28	61	73	70	56.9	109	61	73	93	50	76.7

Table II

Forest bird population census  
Nuclear polyhedrosis virus untreated control plot  
Manitoulin Island Ontario  
May 29 - June 9 1975

Family	Species	Pre-spray					Daily ave..	Post-spray					Daily ave.
		May 29	May 30	May 31	June 1	June 2		June 4	June 6	June 7	June 8	June 9	
		-5	-4	-3	-2	-1		+1	+3	+4	+5	+6	
Trochilidae	Ruby-throated Hummingbird	0	0	0	0	0	0.0	0	0	0	0	2	0.4
Picidae	Yellow-shafted Flicker	0	2	2	0	0	0.8	2	0	0	0	0	0.4
	Yellow-bellied Sapsucker	0	0	2	0	0	0.4	0	0	0	0	0	0.0
	Hairy Wood- pecker	0	0	0	1	0	0.2	0	0	0	0	0	0.0
Tyrannidae	Great-crested Flycatcher	2	2	2	4	2	2.4	2	2	2	0	0	1.2
	Least Flycatcher	0	0	2	0	0	0.4	0	0	0	0	0	0.0
	Eastern Wood Pewee	0	2	0	2	0	0.8	0	0	0	0	0	0.0
Corvidae	Blue Jay	0	0	0	0	0	0.0	0	0	0	2	0	0.4
	Common Crow	0	0	1	0	1	0.4	0	0	0	0	0	0.0
Paridae	Black-capped Chickadee	2	2	0	0	4	1.6	2	0	2	0	2	1.2
Sittidae	Red-breasted Nuthatch	2	2	0	0	0	0.8	0	0	0	0	0	0.0
Troglodytidae	Winter Wren	4	2	2	2	2	2.4	0	2	0	0	2	0.8
Mimidae	Catbird	0	0	2	2	0	0.8	0	0	0	0	2	0.4
Turdidae	American Robin	1	0	1	2	0	0.8	2	3	2	1	1	1.8
	Wood Thrush	2	2	2	0	0	1.2	2	0	0	0	0	0.4

Table II (Cont'd)

Family	Species	Pre-spray					Daily ave.	Post-spray					Daily ave.
		May 29	May 30	May 31	June 1	June 2		June 4	June 6	June 7	June 8	June 9	
		-5	-4	-3	-2	-1		+1	+3	+4	+5	+6	
Turdidae (cont'd)	Hermit Thrush	0	0	0	0	2	0.4	0	0	0	2	0	0.4
	Veery	3	0	2	2	3	2.0	0	0	0	0	0	0.0
Sylviidae	Golden-crowned Kinglet	2	2	4	4	2	2.8	0	6	2	4	4	3.2
Vireonidae	Red-eyed Vireo	4	4	2	0	0	2.0	4	2	0	0	2	1.6
Parulidae	Black and White Warbler	2	6	2	4	4	3.6	10	2	2	4	6	4.8
	Tennessee Warbler	0	4	0	0	0	0.8	0	0	2	2	4	1.6
	Nashville Warbler	4	0	2	2	6	2.8	0	2	2	0	2	1.2
	Magnolia Warbler	2	0	0	2	2	1.2	2	0	0	0	0	0.4
	Cape May Warbler	0	0	0	0	2	0.4	2	2	0	2	2	1.6
	Myrtle Warbler	2	0	2	2	0	1.2	0	2	0	0	0	0.4
	Black-throated Green Warbler	8	4	2	2	4	4.0	4	4	4	0	4	3.2
	Blackburnian Warbler	0	0	4	2	4	2.0	2	6	2	4	2	3.2
	Chestnut-sided Warbler	6	2	4	2	4	3.6	4	2	0	2	0	1.6
	Bay-breasted Warbler	0	0	0	0	0	0.0	0	0	2	0	0	0.4
	Ovenbird	12	10	8	12	4	9.2	6	6	2	8	4	5.2

Table II (Cont'd)

Family	Species	Pre-spray					Daily ave.	Post-spray					Daily ave.
		May 29	May 30	May 31	June 1	June 2		June 4	June 6	June 7	June 8	June 9	
		-5	-4	-3	-2	-1		+1	+3	+4	+5	+6	
Parulidae (cont'd)	Northern Waterthrush	2	2	0	0	2	1.2	2	2	2	2	2	2.0
	Connecticut Warbler	0	0	0	0	0	0.0	0	0	0	2	2	0.8
	Mourning Warbler	0	0	0	0	0	0.0	0	0	0	2	0	0.4
	Canada Warbler	0	4	0	2	6	2.4	4	6	2	2	2	3.2
	Icteridae	Brown-headed Cowbird	4	4	0	4	4	3.2	2	4	0	2	0
Fringillidae	Rose-breasted Grosbeak	4	4	4	4	4	4.0	0	2	2	0	2	1.2
	Purple Finch	2	0	0	2	0	0.8	0	0	4	0	2	1.2
	Chipping Sparrow	2	0	0	2	0	0.8	0	0	0	0	0	0.0
	White-throated Sparrow	0	0	0	0	0	0.0	0	2	0	0	0	0.4
Unidentified	Species	0	0	2	1	0	0.6	0	0	0	0	0	0.0
Totals		72	60	54	62	62	62.0	52	57	34	41	49	46.6

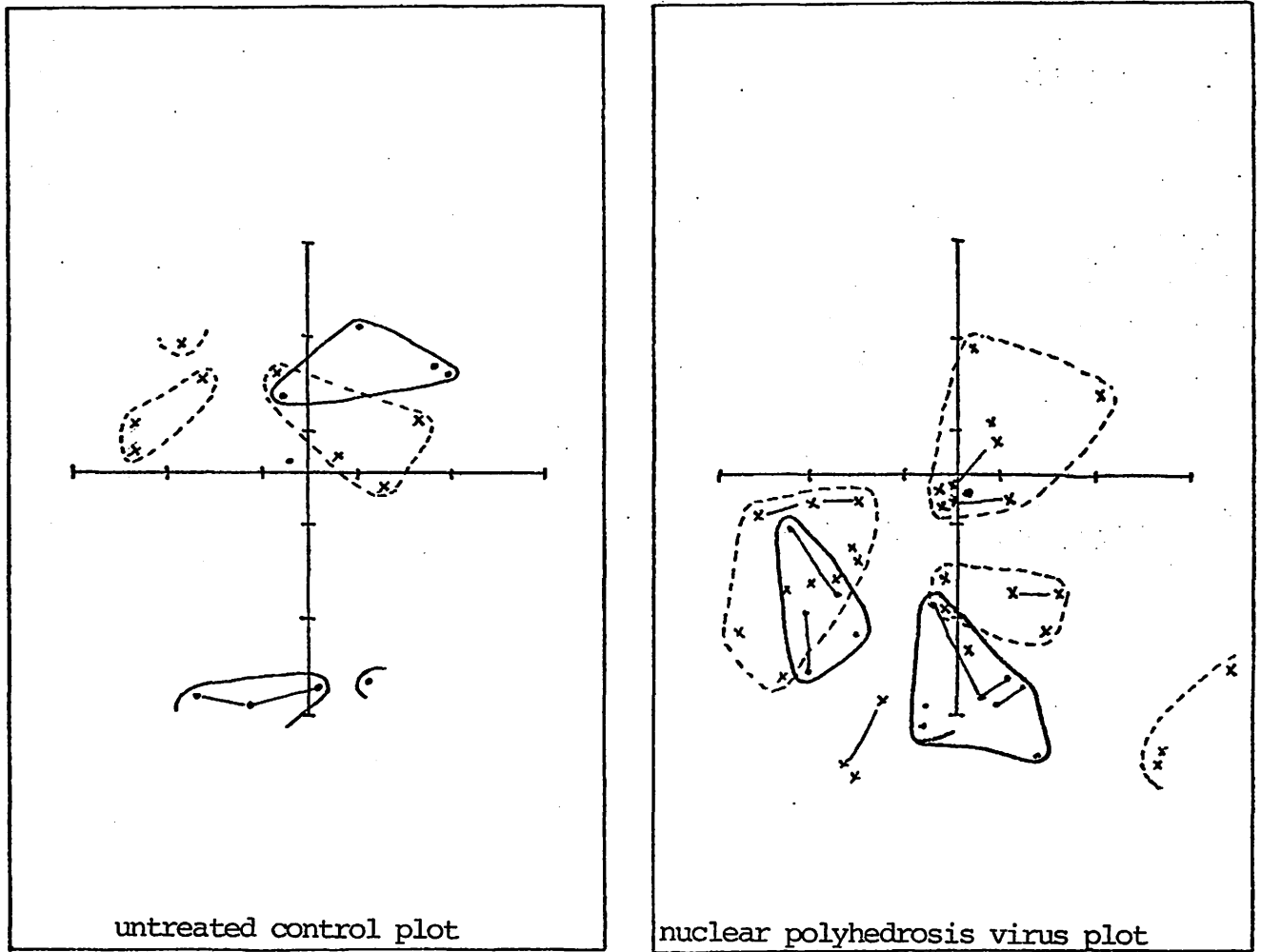


Fig. 1 Pre and post-spray territories of the golden-crowned kinglet, *Regulus satrapa* Lichenstein on the virus treated and untreated plots.

————— . pre-spray  
----- x post-spray

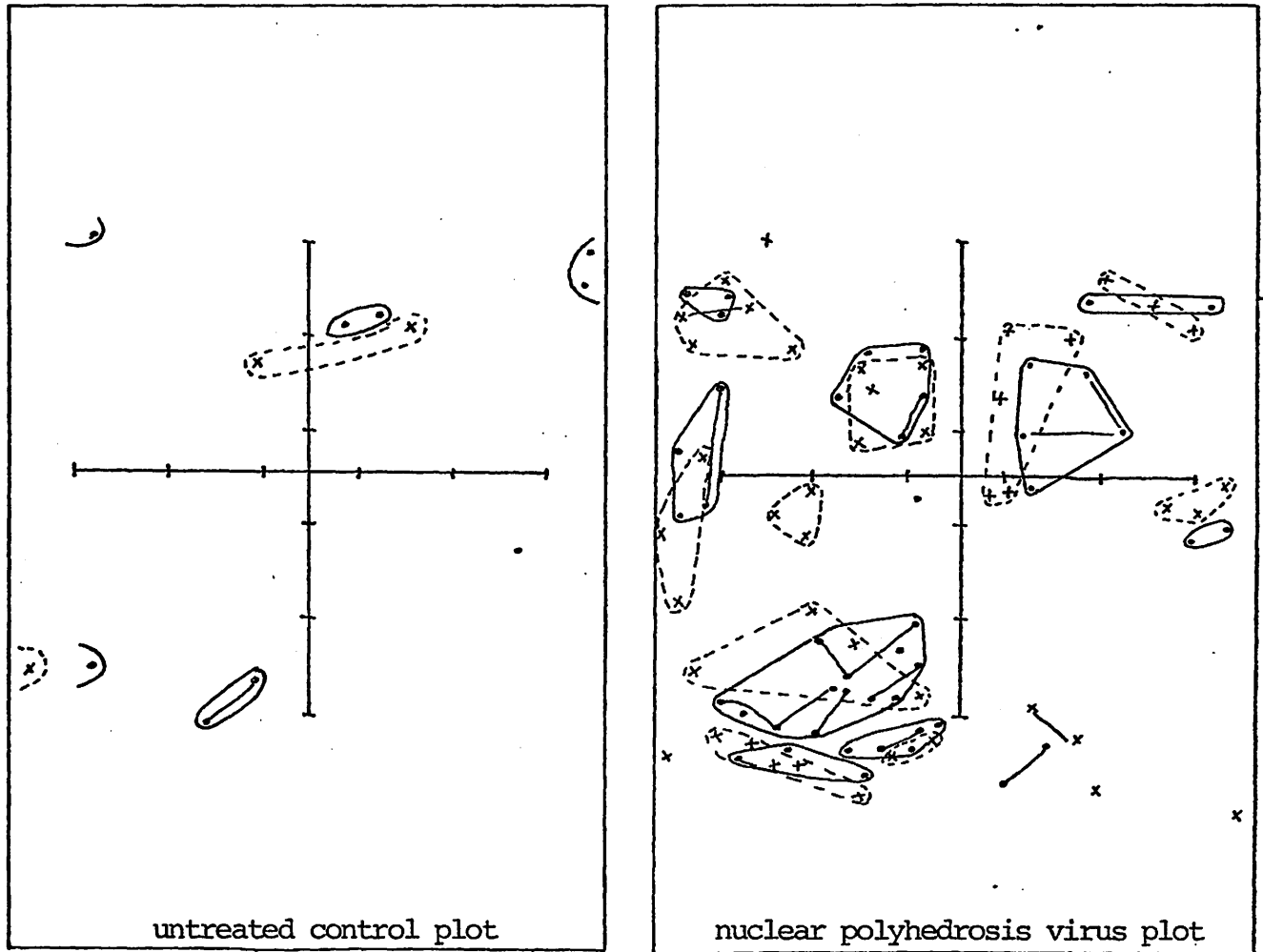


Fig. 2 Pre and post-spray territories of the nashville warbler, *Vermivora ruficapilla* (Wilson) on the virus treated and untreated plots.

\_\_\_\_\_ . pre-spray  
----- x post-spray

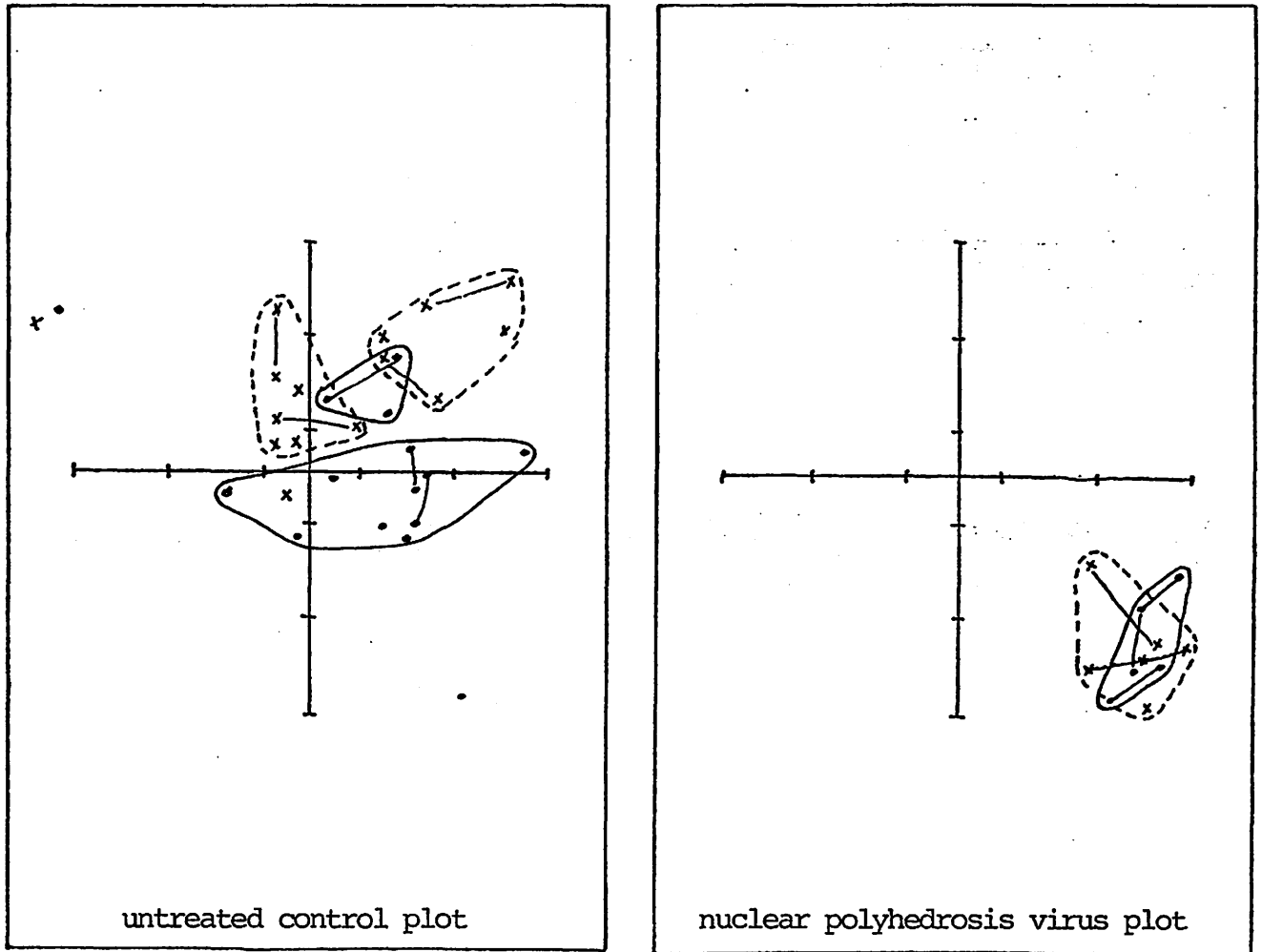


Fig. 3 Pre and post-spray territories of the black-throated green warbler, *Dendroica virens* (Gmelin) on the virus treated and untreated plots.

\_\_\_\_\_ . pre-spray  
----- x post-spray



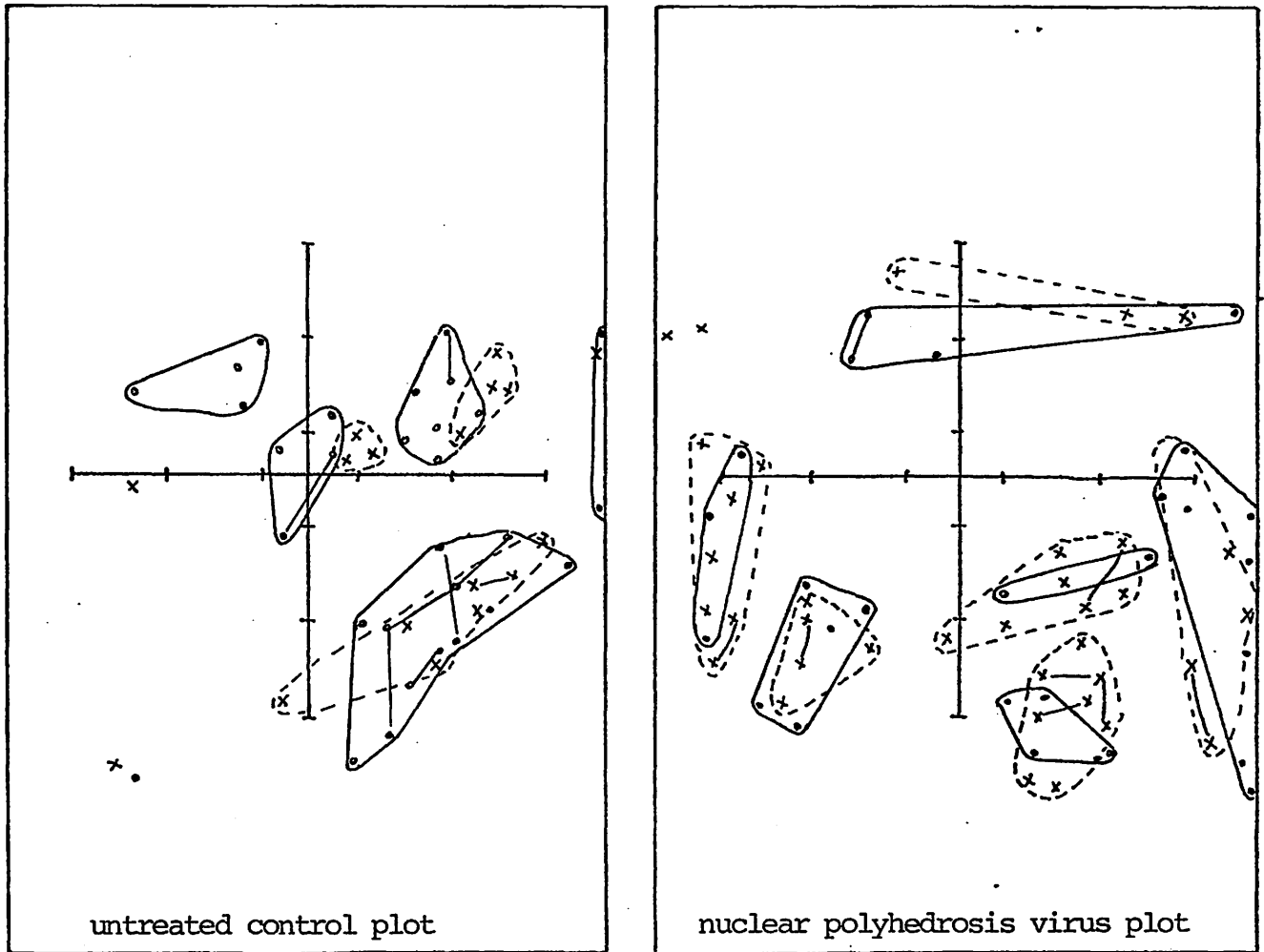


Fig. 4 Pre and post-spray territories of the ovenbird, *Seiurus aurocapillus* (Linnaeus) on the virus treated and untreated plots.

— . pre-spray  
- - - x post-spray

Small mammals: Only 1 species of small mammal, *Peromyscus maniculatus* (Wagner) was trapped on the experimental plots. Seven specimens were trapped on the virus treated plot and two on the untreated control plot (Table III). The small numbers of animals encountered no doubt reflect natural population levels rather than pesticide impact. All 4 females trapped on the treated plot carried embryos indicating no disruption of breeding.

Table III

Small mammal populations trapped on nuclear  
polyhedrosis virus treatment and control plots  
Manitoulin Island, Ontario  
July 1975

Plot	Species	Males			Females					Total animals
		Sub adult	Adult	Total males	Sub adult	Adults			Total females	
						Pregnant	Pregnant with scars	Scars only		
N.P.V.	<i>Peromyscus maniculatus</i>	0	3	3	0	3	1	0	4	7
Control	<i>Peromyscus maniculatus</i>	0	2	2	0	0	0	0	0	2

Honey bees:- The aerial application of nuclear polyhedrosis virus did not cause any mortality to the foraging component of the colonies located on the treated plot (Table IV). Adverse weather caused reductions in pollen collection on the third day after treatment. Monitoring of the brood rings located in each hive indicated no interruption of the normal metamorphosis from egg to adult (Table V). Monitoring continued for 22 days after treatment in order to assess any delayed impact but none was found. On August 20 the colonies were examined for general health and honey production and were found to compare favorably with untreated colonies in these respects.

Table IV

Pesticide impact measurements of honey bee colonies on  
nuclear polyhedrosis virus treated and untreated plots  
Manitoulin Island, Ontario  
June 1975  
(average of 5 colonies on each plot)

Days from treatment	Untreated plot				N.P.V. treated plot				Remarks
	Adult bee mortality	Adult activity trips/day	Pollen collected gms	Hive weights kg	Adult bee mortality	Adult activity trips/day	Pollen collected gms	Hive weights kg	
-3	8	12,800	0.0		7	27,264	0.0		Cloudy with showers, 25°C
-2	4	38,400	31.1	17.2	3	96,384	25.2	19.3	rain in am - clearing pm
-1	4	21,376	17.6		4	13,824	20.6		sunny, cool & windy, 16°C
-0	3	30,336	21.3	17.8	5	18,304	23.3	19.5	sunny periods, high of 18°C
<u>Spray day</u>									
+1	2	32,512	23.5		5	10,624	33.4		rain in am, cloud & fog
+2	2	20,736	18.3	18.1	2	10,496	20.8	19.2	heavy rains
+3	3	18,560	2.1		3	10,240	6.0		windy - showers
+4	3	24,960	33.4	17.7	2	12,032	18.3	19.8	overcast - cool
+5	3	20,352	41.7		3	10,496	27.5		sunny, windy, high of 18°C
+6	4	41,088	87.0	17.2	3	20,224	25.5	18.3	sunny, windy, high of 23°C

Table V

Results of monitoring "brood rings" placed in honey bee colonies on  
nuclear polyhedrosis virus treated and untreated plots  
Manitoulin Island, Ontario  
May-June 1975

Days from treatment	Untreated plot		Virus treated plot		
	Hive no.	Brood ring "A"	Hive no.	Brood ring "A"	Brood ring "B"
-0	16	3/4 young larvae, 1/4 mature larvae	19	2/3 young larvae, 1/3 eggs	1/2 young larvae, 1/3 eggs, 1/6 capped brood
	25	2/3 young larvae, 1/3 eggs	29	3/4 young larvae, 1/8 eggs, 1/8 capped brood	3/4 young larvae, 1/4 capped brood
	22	2/3 young larvae, 1/3 mature larvae	18	7/8 young larvae, 1/8 1/8 capped brood	1/3 young larvae, 2/3 mature larvae
	27	1/2 young larvae, 1/2 mature larvae	20	1/2 young larvae, 1/6 eggs, 1/3 mature larvae	1/3 young larvae, 1/2 mature larvae, 1/6 capped brood
	31	2/3 young larvae, 1/3 mature larvae	15	2/3 young larvae, 1/6 empty, 1/6 mature larvae	1/3 young larvae and eggs, 2/3 empty
+7	16	7/8 capped, 1/8 empty	19	7/8 capped, 1/8 empty	3/4 capped, 1/4 empty
	25	2/3 capped, 1/3 empty	29	2/3 capped, 1/3 empty	1/3 capped, 1/3 eggs, 1/3 empty
	22	7/8 capped, 1/8 empty	18	7/8 capped, 1/8 empty	3/4 capped, 1/8 larvae, 1/8 eggs
	27	7/8 capped, 1/8 empty	20	3/4 capped, 1/8 empty, 1/8 larvae	3/4 capped, 1/8 empty, 1/8 larvae
	31	all capped	15	1/2 capped, 1/2 empty	1/2 capped, 3/4 empty
+14	16	7/8 capped, 1/8 empty	19	3/4 capped, 1/4 empty	7/8 capped, 1/8 empty
	25	1/2 capped, 1/8 larvae, 3/8 empty	29	2/3 capped, 1/3 larvae	3/4 capped, 1/4 larvae
	22	1/2 capped, 3/4 empty	18	7/8 capped, 1/8 empty	1/2 eggs, 1/4 capped, 1/4 empty
	27	1/3 capped, 1/3 empty, 1/3 eggs	20	3/4 capped, 1/4 empty	2/3 capped, 1/3 empty
	31	2/3 capped, 1/6 honey, 1/6 empty	15	7/8 empty, 1/8 capped	7/8 empty, 1/8 capped

Table V (Cont'd)

Days from treatment	Untreated plot		Virus treated plot		
	Hive no.	Brood ring "A"	Hive no.	Brood ring "A"	Brood ring "B"
+22	16	all larvae	19	all larvae	all larvae
	25	1/8 capped, 1/4 eggs, 5/8 larvae	29	3/4 larvae, 1/4 capped	5/6 larvae, 1/6 empty
	22	7/8 larvae, 1/8 capped	18	all larvae	--
	27	1/4 capped, 3/4 larvae	20	all larvae	all larvae
	31	3/4 eggs, 1/8 empty, 1/8 honey	15	3/4 larvae, 1/4 honey	all larvae

Aquatics: The stream sampled within the virus treatment plot was very narrow (about 30 cm wide) and slow flowing with a silty bottom covered with organic debris. The control stream was similar but with more gravel in the stream bed. Populations of aquatic organisms in these streams are presented in Table VI. There are no significant indications of adverse effects of the virus treatment on any group of aquatic organisms. Tadpoles were observed to be as abundant in the treatment stream after the spray application as before treatment, even though none were collected in the post-spray Surber samples. The disappearance of caddisfly larvae (Trichoptera) after treatment can not be considered significant because of their low populations and wide variation in abundance in pre-treatment samples.



Table VI

Bottom fauna populations in the nuclear polyhedrosis virus  
treatment and control streams as numbers and standard deviations  
of organisms/0.092 sq. m. (square foot)  
Manitoulin Island, Ontario  
June 1 to 7, 1975

	Treatment stream		Control stream
	June 1	June 7	June 7
Ephemeroptera	4.5 ± 3.1	11.8 ± 7.7	0.8 ± 1.5
Trichoptera	1.2 ± 1.9	-----	3.0 ± 4.7
Odonata	-----	0.8 ± 1.1	1.2 ± 1.2
Coleoptera	-----	-----	0.2 ± 0.5
Hemiptera	-----	-----	0.2 ± 0.5
Diptera-Chironomidae	4.8 ± 2.5	6.5 ± 3.0	2.2 ± 2.0
Diptera-Heleidae	-----	-----	0.2 ± 0.5
Diptera-Tipulidae	0.2 ± 0.5	0.5 ± 0.6	-----
Nematoda	-----	0.2 ± 0.5	-----
Oligochaeta	2.0 ± 1.6	2.5 ± 3.8	1.0 ± 0.8
Hirudinea	0.8 ± 1.1	1.2 ± 1.2	-----
Amphipoda	2.8 ± 3.8	4.8 ± 2.8	-----
Isopoda	0.2 ± 0.5	-----	-----
Hydracarina	-----	0.2 ± 0.5	-----
Gastropoda	0.2 ± 0.5	0.8 ± 1.1	-----
Pelecypoda	9.0 ± 8.8	8.5 ± 8.3	1.8 ± 2.9
Amphibia	13.8 ± 27.5	-----	-----
Pisces	0.5 ± 1.0	0.2 ± 0.5	0.2 ± 0.5
TOTAL :	40.0 ± 24.0	38.0 ± 18.6	11.0 ± 9.0

### CONCLUSIONS

The safe nature of nuclear polyhedrosis viruses with respect to their lack of effects on vertebrates has been reviewed by Heimpel (1970). The results of the studies conducted on Manitoulin Island in 1975 indicate that under the conditions of application this virus had no immediate or short term effects on forest songbirds, small mammals, domestic honey bees or aquatic organisms.

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