

**EFFICACY OF SINGLE AND DOUBLE
APPLICATIONS OF FORAY 48B
(*BACILLUS THURINGIENSIS*)
AGAINST THE GYPSY MOTH
(*LYMANTRIA DISPAR* L.) IN ONTARIO**

*J.H. Meating, S. Schyff¹, G.M. Howse
and J.J. Churcher¹*

Forestry Canada
Ontario Region
Great Lakes Forestry Centre

1993

Information Report O-X-423

¹Ontario Ministry of Natural Resources Forest Resources Branch,
Silviculture Section Sault Ste. Marie, Ontario



The printing of this report was funded by the Northern Ontario Development Agreement



This report is printed on recycled paper

Canadian Cataloguing in Publication Data

J.H. Meating, S. Schyff, G.M. Howse and J.J. Churcher
Efficacy of single and double applications of Foray 48B (*Bacillus thuringiensis*)
against the gypsy moth (*Lymantria dispar* L.) in Ontario
(Information report ; ISSN 0832-7122 ; O-X-423)
Includes an abstract in French.
Includes bibliographical references.
ISBN 0-662-20528-6 DSS cat. no. Fo46-14/423E

1. Gypsy moth – Control – Ontario. 2. Spraying and dusting
in forestry – Ontario 3. Insecticides – Ontario.

I. Meating, Joseph Hazen, 1951-

II. Great Lakes Forestry Centre.

III. Series: Information report (Great Lakes Forestry Centre); O-X-423.

SB945.G9E33 1993 634.9'6781 C93-099543-0

©Minister of Supply and Services Canada 1993

Catalogue No. Fo 46-14/423E

ISBN 0-662-20528-6

ISSN 0832-7122

Copies of this publication are available at no charge from:

Communications Services
Forestry Canada
Ontario Region
Great Lakes Forestry Centre
P.O. Box 490
Sault Ste. Marie, Ontario
P6A 5M7

Microfiches of this publication may be purchased from:

Micro Media Inc.
Place du Portage
165, Hotel-de-Ville
Hull, Quebec J8X 3X2

Meating, J.H., Schyff, S., Howse, G.M. and Churcher, J.J. 1993. Efficacy of single and double applications of Foray 48B (*Bacillus thuringiensis*) against the gypsy moth (*Lymantria dispar* L.) in Ontario. For. Can., Ont. Region, Sault Ste. Marie, Ont. Inf. Rep. 0-X-423. 5 p.

ABSTRACT

In May 1991, an experimental aerial spraying program was conducted in Pembroke District to compare the efficacy of single and double applications of Foray 48B (*Bacillus thuringiensis* Berliner) (B.t.) against the gypsy moth (*Lymantria dispar* L.).

Two blocks were treated with a double application of B.t. (30 BIU/2.4 L/ha), and six blocks (three "early" and three "late") were treated with a single B.t. application (50 BIU/4 L/ha).

The treatments appeared to have had little effect on gypsy moth populations, but defoliation levels in all spray blocks were lower than in untreated check plots with similar pre-spray egg-mass densities. Protection of foliage was greatest in the blocks treated with two applications of B.t., intermediate in the blocks treated "early" with the single B.t. application and least in the blocks treated "late".

RÉSUMÉ

En mai 1991, un programme expérimental de pulvérisations aériennes a été mené dans le district de Pembroke afin de comparer l'efficacité d'une seule application et de deux applications de Foray 48B (*Bacillus thuringiensis* Berliner) (B.t.) pour lutter contre la spongieuse (*Lymantria dispar* L.).

Deux blocs ont reçu deux applications de B.t. (30 MUI/2,4 L/ha) et six blocs ont reçu une application unique, soit 50 MUI/2,4 L/ha (trois blocs, tôt en saison, et les trois autres, tard en saison).

Les traitements semblent avoir eu peu d'effet sur les populations de la spongieuse, mais les degrés de défoliation de tous les blocs traités étaient plus faibles que dans les parcelles témoins ayant des densités prétraitement similaires de masses d'œufs. La protection du feuillage procurée par les traitements était plus élevée dans les blocs qui avaient reçu une double dose, moyenne dans les blocs traités tôt en saison et plus faible dans les blocs traités tard en saison.

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INTRODUCTION

In 1981, 23 pockets of gypsy moth (*Lymantria dispar* L.) defoliation, totaling approximately 1,450 ha, were detected in the Eastern Region of Ontario (Howse and Applejohn 1982). In 1982, the Ontario Ministry of Natural Resources (OMNR) aerially sprayed 353 ha with Dipel 88 (*Bacillus thuringiensis* Berliner) (B.t.) and Sevin-4-Oil (Carbaryl). Researchers from Forestry Canada's Forest Pest Management Institute (Sault Ste. Marie, Ont.) treated an additional 63 ha with Gypchek (nuclear polyhedrosis virus) (Meating et al. 1983). By 1985, the gypsy moth infestation in Ontario covered more than 246,000 ha (Howse and Applejohn 1985). In 1986, OMNR conducted the largest aerial spraying program in its history against the gypsy moth. More than 103,000 ha were treated with double or triple applications of B.t. (Meating et al. 1986). Gypsy moth populations declined in 1986 and by 1987, moderate-to-severe defoliation was detected over only 12,678 ha. Over the next few years, however, the gypsy moth infestation expanded steadily in the province until, in 1991, more than 347,000 ha of defoliation was reported (Howse and Applejohn 1991). During the period from 1987 to 1991, OMNR aerially treated an additional 137,517 ha of gypsy moth-infested stands with B.t.

There are several potential benefits to reducing operational spray programs from double applications to single. Of primary concern to most government foresters and entomologists is the necessity of reducing program costs or maximizing treatment area when budgets are fixed. Programs involving single applications are generally simpler to coordinate and are less time-consuming. There is also less risk of accident or injury to aircraft, pilots and ground personnel when the number of spray sessions can be reduced.

Results of annual surveys conducted in Ontario since 1981 show that gypsy moth egg hatch at specific locations normally occurs over a 3- to 4-week period in late April and early May. This prolonged period of egg hatch has meant that double and sometimes triple applications of B.t. have been necessary to protect host foliage. The use of double applications has been recommended by Dubois (1991) in the United States when egg hatch is extended beyond 14 days. Therefore, a cooperative experimental spraying program involving OMNR, Forestry Canada and Novo Nordisk was undertaken in 1991 to compare the efficacy of a single

B.t. treatment to the standard operational double application of B.t. Two timing regimes were used for the single B.t. treatments. An "early" treatment was scheduled to coincide with the first application of the operational program and a "late" treatment coincided with the second operational spray approximately 5 days later (Table 1).

METHODS AND MATERIALS

All blocks in the 1991 experimental program were located in Pembroke District approximately 40 km west of the town of Pembroke near Bonnechere Provincial Park (45°42'N, 77°35'W). Rocky, shallow-soiled ridges are typical of the area, with elevations ranging between 180 and 360 m above sea level. Stand composition in all treatment and control plots was primarily red oak (*Quercus rubra* L.) and trembling aspen (*Populus tremuloides* Michx.), with a minor component of white birch (*Betula papyrifera* Marsh.), sugar maple (*Acer saccharum* Marsh.), jack pine (*Pinus banksiana* Lamb.) and eastern white pine (*Pinus strobus* L.). White oak (*Quercus alba* L.) was relatively common in one block. Tree heights ranged from 10 to 15 m.

Gypsy moth defoliation was first detected in the area south of Bonnechere Provincial Park in 1988. Moderate-to-severe defoliation was first observed in 1990 in the stands selected for this study and was, therefore, considered to be on the "leading edge" of the gypsy moth outbreak in Ontario.

Eight experimental spray blocks, each 10 ha in size, were established within an area of 50 km². However, because of the locations of susceptible stands and access limitations, spray blocks were sometimes clustered into groups of two or three. In these instances, blocks were separated by a minimum distance of 400 m. Block boundaries were marked with helium-filled weather balloons before each treatment. Control plots were located a minimum of 1 km from the nearest spray block.

Foray 48B, an aqueous formulation of the bacterial insecticide *Bacillus thuringiensis* Berliner (B.t.), was provided by Novo Nordisk for all treatments. Basazol Red (Rhodamine B dye, 0.25%) was mixed with each treatment to facilitate assessment of spray deposition. A Piper Pawnee aircraft equipped with four Micronair (AU 3000) rotating nozzles, with a blade setting of 35°, was used for all treatments. The flow meter used to set flow rates for the

treatments was ground checked and determined to be accurate to within 2% after three trials. Mixing and loading of the aircraft took place at Bonnechere Airfield, which was within 8 km of the most distant block. Airspeed over each block was approximately 160 km/hour. Swath width was 30 m.

Table 1. Gypsy moth spray treatments with Foray 48B in the Pembroke District of Ontario, 1991.

Treatment	Number of applications	Timing	Number of blocks	Dates sprayed
30 BIU/2.4 L/ha	2	Operational	2	23 and 28 May
50 BIU/4 L/ha	1	Early	3	23 May
50 BIU/4 L/ha	1	Late	3	28 May

Weather conditions before, during and after each treatment were monitored using a portable weather station located at the airstrip. Air speed, temperature and relative humidity were recorded at 15-minute intervals during each spray session.

Four plots were established in treatment and control blocks to monitor gypsy moth egg hatch. Because of the rolling topography, egg hatch was monitored on both northern and southern exposures. Fifty egg masses, 25 on the northern sides of trees and 25 on the southern sides, were tagged in each plot and assessed for hatch daily from 7 to 17 May.

Gypsy moth larval development and host foliar development were monitored at two locations before and during the trials. Larvae and foliage were also evaluated in some individual spray blocks as they were treated.

Spray deposition was assessed at each of five plots established within each spray block. One mid-crown branch was sampled from each of two trees per plot approximately 1 to 3 hours after treatment. Ten leaves were removed from each branch, placed in individually labeled Petri dishes, and stored at 4°C until they could be examined. In all, 100 leaves were assessed for spray deposition from each block. In the laboratory, five areas (0.36 cm² each) from the upper leaf surface and five from the lower surface were microscopically examined and the number of spray droplets was recorded.

Spray efficacy was evaluated in terms of the reduction in the numbers of gypsy moth egg masses and the protection of host foliage at five 0.01-ha plots established in each spray

block. An additional 16 plots were monitored outside the spray blocks to assess natural population changes and defoliation in untreated areas.

Surveys were conducted in the spring to determine the 1990 gypsy moth egg-mass densities in each plot. The same plots were surveyed again in October to assess the 1991 egg-mass densities and thus provide estimates of population change.

Upon completion of larval feeding in July, all plots were surveyed to provide estimates of host defoliation. A single mid-crown branch was removed from each of 10 trees in or near each plot. The numbers of damaged and undamaged leaves, and estimates of defoliation for each damaged leaf, were recorded for each branch. Red oak was the primary host evaluated in all plots except in Block 3, where white oak was also a major component of the stand.

RESULTS

Gypsy Moth Egg Hatch

Gypsy moth egg hatch was monitored in four plots in the experimental area. Two plots were located on northern exposures and two on southern exposures. Fifty egg masses were marked in each plot and checked daily for evidence of egg hatch. Hatch generally appeared to begin earlier, and was initially more advanced, in the south-facing plots. However, there was little difference between north- and south-facing plots as hatch approached 100% (Fig. 1). Eggs had started to hatch in all egg masses in all four plots by 13 or 14 May.

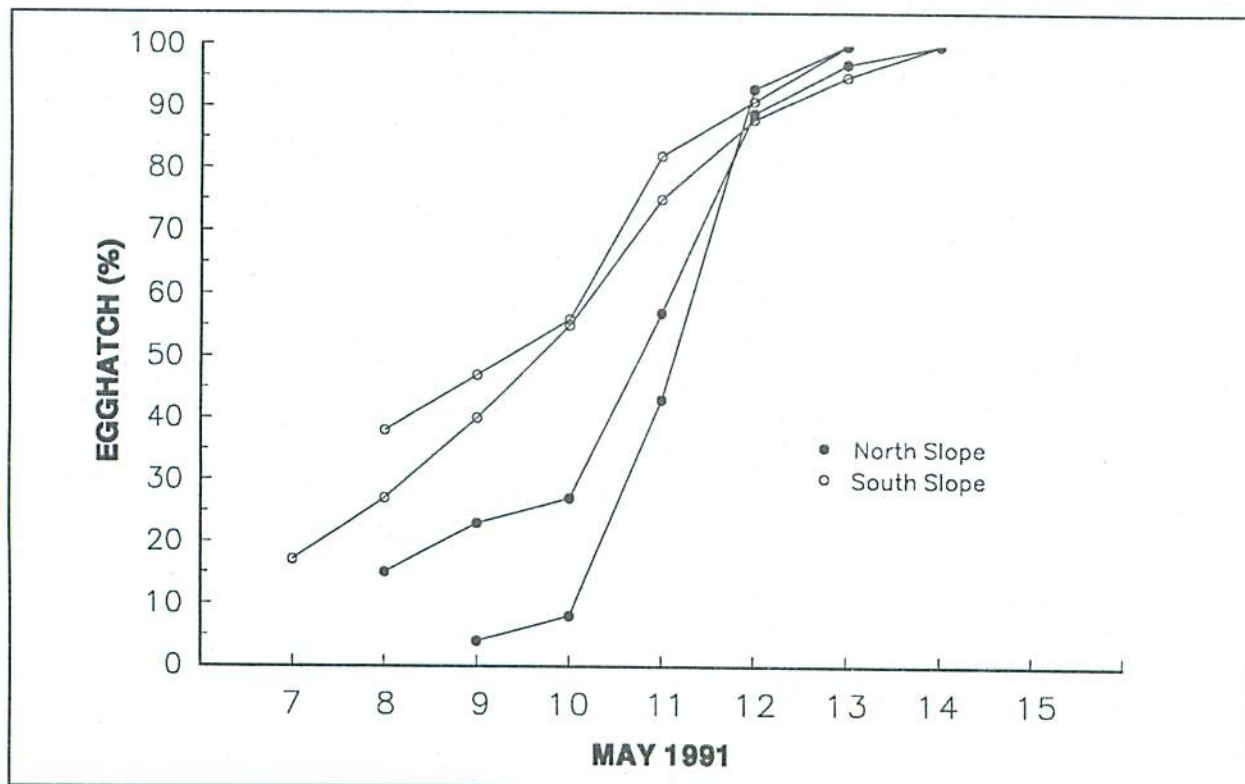


Figure 1. Gypsy moth egg hatch on north- and south-facing slopes in the Pembroke District of Ontario in 1991.

Insect and Host Development

Unusually warm temperatures in April and May 1991 reduced the normal egg-hatch period of 3 to 5 weeks to approximately 1 week and resulted in rapid larval and host development in May. The results of the gypsy moth larval and host development surveys are presented in Table 2. The first experimental spray session occurred on 23 May, when most gypsy moth larvae were in the first and second instars and the red oak foliage was 50 to 60% expanded. By 25 May, larvae were predominantly second and third instars and foliar expansion was approximately 65%. Between 25 and 28 May, temperatures were cool, with showers and overcast conditions. Thus, there was little change in larval development. On the final day of the program (28 May), the majority of larvae were still second and third instars. Foliar expansion was approximately 65 to 70%.

Table 2. Gypsy moth larval and host development in the Bonnechere area of Pembroke District, 1991.

Date	Plot	Proportion (%) in instar				Red oak foliage (% expansion)
		I	II	III	IV	
May 15	1	100	—	—	—	27
	2	100	—	—	—	28
May 17	1	90	10	—	—	40
	2	90	10	—	—	42
May 20	1	83	17	—	—	49
	2	67	28	5	—	52
May 22	1	52	44	4	—	60
	2	29	57	13	—	52
May 23 ^a	3	41	50	19	—	50
May 25	4	3	21	71	5	65
May 27	1	3	55	42	—	71
	2	—	19	80	1	62
May 28 ^a	3	—	40	58	2	65

^a spray dates

Weather

Weather information was collected at the Bonnechere Airstrip during each spray session. Air temperature, relative humidity and wind speed data were recorded at 15-minute intervals and precipitation was monitored for 48 hours after spraying (Table 3).

Table 3. Weather data recorded at the Bonnechere Airstrip during the experimental gypsy moth control trials in Ontario, 1991.

Date	Time	Start – finish			Precipitation 48 hours after spraying (mm)
		Temperature (°C)	Relative humidity (%)	Wind speed (km/hour)	
23 May	0810–0940	18–26	86–62	3–4	<1
28 May	0715–0845	16–17	93–81	0–10 ^a	0

^a gusts to 15 km/hour

During the first spray session (23 May), the temperature rose (from 18° to 26°C) and relative humidity decreased (from 86% to 62%) rapidly. Wind speed remained low (3 to 4 km/hour) during the entire session. Some 1.5 hours after the last treatment, light showers passed through the area. Less than 1 mm of rain was recorded and there was no further precipitation until 0500 on 25 May, when heavy showers passed through the area. These showers lasted for approximately 1 hour. The last spray session began at 0715 on 28 May. Temperatures (16 to 17°C) and relative humidity (93 to 81%) remained fairly constant, but wind speeds fluctuated from 0 to 10 km/hour, with gusts to 15 km/hour. Reports from the pilot and observers in the spray blocks indicated that the spray cloud was deposited in the blocks. No precipitation was recorded during the following 48 hours.

Spray Deposition

The upper and lower surfaces of 20 mid-crown leaves collected from each plot were examined under a stereomicroscope. Droplet densities were determined in five randomly selected fields on each surface for a total search area of 3.6 cm² per leaf. Droplet densities were converted to "drops per cm²" and are presented in Table 4. Overall block means varied considerably, with more than a 10-fold difference between the highest (21.4 drops/cm²) and the lowest (2.0 drops/cm²) depositions. Variability between plots in each block was also generally high, suggesting a very irregular distribution of B.t. within each block.

Spray Efficacy

Changes in gypsy moth egg-mass densities and host defoliation were assessed in each of the five 0.01-ha plots that transected each spray block. An additional 16 plots located in unsprayed stands were monitored to assess natural population changes and defoliation rates. Summaries of gypsy moth egg-mass reductions and foliar protection for each block are presented in Table 5.

With the exception of Block 1 (a single "late" application of 50 BIU/ha), there seems to have been little effect of spraying on gypsy moth egg-mass densities. Generally, egg-mass densities in spray and check plots that had high 1990 densities decreased in 1991, and densities in plots that had low 1990 densities increased in 1991.

Defoliation levels prior to treatment ranged between 5 and 10% in all plots except Plot 1, Block 2 (a single "early" application of 50 BIU/ha), where prespray defoliation was approximately 25%. The final estimated defoliation rate in this plot was 65%, the highest level of defoliation observed in any of the 56 plots assessed. The red oak within

Table 4. Individual plot means for spray droplet density for each block treated with Foray 48B in 1991.

Treatment	Block	Drops per cm ² for plot					\bar{X}	SE
		1	2	3	4	5		
Single, 50 BIU/ha, early	1	44.4	2.0	7.8	1.7	4.9	12.2	2.9
	2	1.0	0.9	1.6	30.8	8.1	8.5	2.7
	3	0.9	4.5	5.8	0.4	62.1	14.7	3.2
Single, 50 BIU/ha, late	1	2.2	3.8	0.7	2.9	1.4	2.2	0.3
	2	0.4	0.1	18.1	57.7	3.2	16.0	4.2
	3	0.5	0.9	26.4	8.3	1.9	7.6	2.1
Double, 30 BIU/ha	1	2.9 ^a	1.4	3.9	8.8	6.6	4.7	0.7
		1.0 ^b	2.1	1.4	3.0	2.4	2.0	0.3
	2	49.6 ^a	1.2	17.9	30.6	7.6	21.4	3.6
		14.3 ^b	0.4	1.2	2.3	0.5	3.7	1.6

^a first application

^b second application

Table 5. Gypsy moth population reductions and foliar protection on red oak (rO) and white oak (wO) in blocks treated with single and double applications of Foray 48B in Pembroke District, Ontario, in 1991.

Treatment	Block number	Number of egg masses/ha		Change (%)	Defoliation (%)			
		1990	1991		red oak	Range	white oak	Range
Single, 50 BIU/ha, early	1	17,461	6,620	-62	28	18-36	-	-
	Checks	17,032	8,067	-53	46	32-52	-	-
	2	19,330	5,720	-70	32	30-39	-	-
	Checks	19,186	2,733	-86	42	29-51	-	-
	3	216	2,100	+872	17	13-19	-	-
	Checks	1,632	2,276	+39	34	30-37	-	-
Single, 50 BIU/ha, late	1	9,237	3,500	-62	35	21-41	-	-
	Checks	7,507	9,933	+32	43	40-46	-	-
	2	14,214	2,980	-79	38	27-45	-	-
	Checks	17,032	8,067	-53	46	32-52	-	-
	3	1,613	2,100	+30	19	17-22	-	-
	Checks	1,789	2,475	+38	34	30-37	-	-
Double, 30 BIU/ha	1	48,271	3,620	-92	15	12-21	13	6-21
	Checks	36,064	6,175	-83	44	30-52	-	-
	2	1,915	2,120	+10	19	17-27	-	-
	Checks	1,789	2,475	+38	34	30-37	-	-

this plot were thin-crowned and sustained varying degrees of branch mortality. By late summer, whole-tree mortality was common in and around this plot and it was deleted from the assessment.

Defoliation levels in all spray blocks were lower than in untreated check plots with similar pre-spray egg-mass densities. Protection was greatest in the two blocks that received the operational application of B.t. (two sprays of 30 BIU/ha). Defoliation rates in the six blocks treated with the single application of 50 BIU/ha were generally greater than in the two operational blocks, but were kept below 50% in all blocks. Differences in defoliation between the treatments were not significant, but defoliation was marginally lower in the blocks treated early ($\bar{X}=26\%$) than in those treated late ($\bar{X}=31\%$).

CONCLUSIONS

The condensed period of gypsy moth egg hatch recorded in Ontario in 1991 was not typical of what has been observed in the province since the pest became established in 1981. In Pembroke District, egg hatch occurred over approximately 7 days in 1991, as opposed to the 3 to 5 weeks that had been the norm in previous years. Therefore, the objective of testing the efficacy of single versus double applications of B.t. under normal Ontario conditions was not achieved. However, the results of the 1991 experimental program demonstrate what can be expected when the hatch period is unusually short. The best results, in terms of foliar protection, were achieved with the standard operational double application of B.t. Defoliation in the blocks treated with a single B.t. application was generally higher, but the results may be acceptable considering that loss of foliage did not exceed 50% in any of the assessed spray plots. Therefore, in years when the gypsy moth egg hatch period is unusually condensed, the tactic of using single B.t. applications to protect foliage appears to be a practical option in Ontario. The practicability of the single-application option still remains to be tested in a more typical year in this province, when egg hatch is extended over a normal period. The comparison between the "early" versus "late" single-application treatment indicates that protection of foliage is most likely to be achieved using an early timing regime.

Additional testing is necessary to evaluate the efficacy of a single B.t. treatment at different points in the gypsy moth outbreak cycle, especially in years when the egg-hatch period is more extended.

ACKNOWLEDGMENTS

We are grateful to OMNR's Forest Resources Branch for funding and support of this study. We wish to thank Temple Bowen and Novo Nordisk for providing the Foray 48B used in this program. Special thanks are due to Garth Sargentson from the Pembroke District OMNR and Tom Donaghy from OMNR's Southern Ontario Forest Health Unit for their support and ideas. We also thank Jerry Losee of Farm Air for his cooperation. The assistance of Dani Landry, Daniela Gebhart, J.P. Beaupre and the other students who were involved in the program was greatly appreciated.

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