

STUDIES ON THE PROTECTION OF INSECT PATHOGENS
FROM SUNLIGHT INACTIVATION II. PRELIMINARY FIELD TRIALS

by

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ABSTRACT

Field tests of four sunlight screens for *Bacillus thuringiensis* (B.t.) spores and spruce budworm nuclear polyhedrosis virus (NPV) were conducted using potted trees and 10 m high naturally grown white spruce. B.t. spores without protectant on potted trees were totally inactivated after 1 day of weathering (570 cal/cm² sunlight radiation). With Uvinul DS49, B.t. activity lasted up to 7 days (3750 cal/cm²). The white spruce foliage itself caused 78% inactivation after 14 days in the dark. NPV alone on white spruce lost 100% activity in 1 day of weathering but with IMC sunlight protectant was still 30% active after 7 days of weathering. Mistblower applications of B.t. or NPV with DS49 as protectant resulted in negligible defoliation of white spruce carrying from 12 to 26 larvae/18" branch. DS49 will be aeriually applied with B.t. in 1976.

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II. PRELIMINARY FIELD TRIALS

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One of the major obstacles to the successful use of insect pathogens as microbial control agents is their rapid decline in activity when exposed to natural sunlight. Due to the filtering effect of atmospheric ozone, germicidal sunlight wavelengths which reach the earth's surface are primarily in the 290 nm to 380 nm range (Kleczkowski 1957, Seliger and McElroy 1965, Webb and Tai 1969). This wavelength range constitutes only about 7% of the total incident light reaching earth. There is evidence the longer wavelengths (364-366 nm) in this range have lower viricidal effects than shorter ones (308 nm) (Morris 1971, Bullock et al 1970). Morris (1975) has recently presented evidence based on measurements of spectral reflectance with monochromatic illumination of pure samples of polyhedral inclusion bodies (NPV) and *Bacillus thuringiensis* spores (B.t.) that the B.t. crystals absorb at wavelengths below 320 nm and fluoresce. The spores and crystals when combined show less fluorescence suggesting that the spores absorb below 320 nm.

The inactivating effect of sunlight on virus applied in the field is well documented (Yendol and Hamlen 1973) and some attempts have been made to increase their persistence by addition of various adjuvants to spray mixes (Yendol and Hamlen 1973; Schmidt 1974) or by

encapsulation (Ignoffo and Batzer 1971). The encapsulation method appears to be highly efficient but difficult and expensive.

The rapid decline in the viability of B.t. spores exposed to sunlight in the field has been reported by Cantwell and Franklin (1966), Pinnock et al (1971), Jaques (1972), Ahmed et al (1973), Frye et al (1973), Ignoffo et al (1974), Morris and Hildebrand (1974), Morris and McErlane (1975), Morris et al (1975). The toxic protein delta endotoxin (the crystal) on the other hand, does not appear to be affected by ultraviolet radiation (Cantwell 1967; Burgess et al 1975; Morris and McErlane 1975). Encapsulation of B.t. spores appears to be effective in increasing their longevity (Cantwell 1967; Martouret and Anglade 1971). Rapid inactivation of microsporidia by sunlight is also known to occur (Wilson 1974).

This paper presents results of preliminary field tests of some easily applied adjuvants against rapid inactivation of commercial B.t. and the NPV of the spruce budworm. The choice of adjuvants was based on laboratory tests (Morris and McErlane 1975).

MATERIALS AND METHODS

Residual Activity Test

The first of two series of experiments in this category was conducted at Ottawa, July 2, 1975. Potted white spruce (*Picea glauca*, Moench) trees about 45 cm high were hand-sprayed with 100 ml of a formulation containing 7.0 g Dipel wettable powder, 0.2 ml Chevron spray sticker and 1% Erio Acid Red dye, 10% Inositol or 10% Molasses (Cargill

Insecticide Base). Foliage from 6 replicate treatments and untreated checks sprayed with water alone was bioassayed at intervals of 0 (unexposed) 1, 2, 4, 8 and 16 days after exposure to sunlight. Third and 4th instar Douglas fir tussock moth (*Orgyia pseudotsugata*) laboratory reared on artificial diet from field collected eggs, were used (Table 1).

In the second series of tests of 3 UV absorbers conducted at Petawawa Forest Experiment Station, Ontario, 7 white spruce trees 45-60 cm in height were sprayed with various formulations (see Table 2) in a specially designed spray tower at the rate of 2 gallon U.S./acre (18.8 l/ha). Four of the trees were exposed to weathering in an open field for 1, 3, 7 and 14 days. Two trees were placed in full shade in a densely foliated grove nearby. The remaining tree from each treatment group was sampled for bioassay immediately after spray deposits had dried. Foliage from each tree was sampled at the end of its exposure period and stored in a dark cold room at 1°C for 3 to 10 days until bioassay could be performed. Meteorological conditions were monitored at the exposure site with particular emphasis on rate of solar radiation using a pyranometer (Weather Measure, model # R401).

Residual activity of the pathogens on the foliage was determined using laboratory reared 4th instar *Choristoneura fumiferana* as test organism. Larvae were allowed to feed on B.t. treated foliage for 7 days and on NPV-treated foliage for 14 days. Cadavers were examined for the presence of pathogens. Rearing conditions are summarized in Table 3.

Mistblower Field Trials

In 1975, a series of small scale spray trials of several formulations of B.t. or NPV + protectants (Table 9) were conducted at

Rankin, Ontario, using a back-pack mistblower. The sprays were applied in the evening of May 28 at 1 l/tree when larvae were mainly in the 3rd and 4th instars. Three replications of each formulation were tested on white spruce trees about 8-10 m high. Population densities and larval mortality were determined 1 day pre-treatment and 13 and 21 days post-treatment. Larvae were counted on two 45 cm branch tips per tree at each sample period.

The residual activities of the pathogens applied were determined at 20 and 27 days post-treatment with B.t. alone, B.t + Orthene^(R) and B.t. + NPV. Pupae were collected for emergence studies from the NPV alone, NPV + chitinase and untreated check only because these were the only trees on which sufficient pupae survived. Defoliation was assayed on two 45 cm branch tips per tree on September 25.

RESULTS AND DISCUSSION

Residual Activity Test

Results of the first residual activity test (Table 1) indicated that Erio Acid Red and 10% Inositol and 10% molasses conferred no protection of B.t. from sunlight inactivation over the 16 day test period.

Meteorological data collected during the exposure of the sprayed potted trees (Table 4) indicated that solar radiation and temperature were normal for the time of year. Rainfall was very low throughout the test period.

Of the three sunlight protectants tested with B.t., Univul DS49 (Chemical Developments of Canada Limited, Montreal), was the most effective, followed by benzyl cinnamate & sodium ascorbate (Table 5, Fig. 1). A 50%

loss of activity after 3 days exposure (about 2 Kcal/cm² of radiation) was considered a decided improvement in persistence but not adequate. B.t. with or without the protectant on spruce trees kept in the shade also lost activity steadily after 3 days (Fig. 2) indicating that the tree foliage itself does cause some loss of B.t. activity. The phenomenon of inactivation of B.t. spores by phytonicides has been documented (Smirnoff 1968; Morris 1972).

IMC sunlight screen which is available commercially from Sandoz, Homestead Florida, was superior to DS49 and benzyl cinnamate for protecting NPV (Fig. 3). However, even with this protectant, 50% activity was lost soon after the first full day of exposure. Unprotected virus lost 100% activity by the end of the first day.

Data from the diagnosis of dead larvae in these tests corroborate the mortality data particularly in the B.t. tests where the incidence of B.t. infection decreased steadily over the test period for all tests (Table 5). The incidence of B.t. infection on the shaded trees was consistently higher than on those exposed to sunlight, indicating that the spores were the main target of the radiation. Natural incidence of NPV was generally very low (Table 6) and except for IMC treatment, there was a substantial reduction in NPV incidence on treated trees after 1 day of exposure. No tests for NPV activity in the shade were made.

The incidences of microsporidia and pathogenic fungi were generally high on all the tests (Tables 7 and 8).

Mistblower Field Trials

The formulations used in mistblower trials are summarized in Table 9. Larval mortality was generally high with the exception of NPV + chitinase, NPV + Dipel, and Orthene alone which gave low to

moderate affects (Table 10). Surprisingly, the B.t. + DS49 gave 100% mortality. The effects of NPV and Orthene were additive in combination as were chitinase and NPV. B.t. + NPV were apparently antagonistic as the combined mortality was lower than with either agent applied alone.

Results of the defoliation assessment (Table 11) showed that all treatments protected the current year's growth. When expected defoliation is compared with actual at similar or near similar larval densities, it is seen that even the NPV + B.t. treatment was highly effective in protecting the tree from excessive defoliation. An analysis of variance for the defoliation assessment is presented in Table 12. The incidence of bacterial infection among dead insects was generally high (Table 13) (especially at 13 days post-spray) except with B.t.-NPV treatment reflecting the apparent antagonistic effect of the combination noted above. NPV incidence among virus sprayed populations (Table 14) was high only at 21 days post-treatment indicating a very delayed effect of the virus pathogen. The high incidence in the B.t.-NPV treatment suggests that the observed mortality was due mainly to the virus. Incidence of microsporidia and fungi was moderate to high in these populations (Tables 15 and 16).

Data on the persistence of the sprayed pathogens (Table 17) show that the foliage remained highly infectious up to 27 days post-spray due mainly to the deposition of B.t. on the sunlight protected under-surface of foliage. At both 13 and 27 post spraying days, the incidence of B.t. was higher among B.t. or B.t.-Orthene treated insects than among B.t.-NPV treated ones. Microsporidia incidence was high among all groups of test insects. Results of rearing of pupal survivors of the NPV and NPV-chitinase treatments (Table 18), indicated that these treatments

had no effect on pupal survival or on oviposition rate.

To assess the true protective effect of DS49 on B.t. and NPV, defoliation figures from the present tests with protectants were compared with similar ones from an earlier test (Morris 1972) without protectants carried out in the same area. The data (Table 19) are consistent with the above observation that DS49 is more effective as a B.t. protectant than as an NPV protectant and that benzyl cinnamate is less effective than DS49. The expected defoliation was calculated from a curve expressing the relationship between spruce budworm larval density and defoliation of current year growth (Fig. 4).

SUMMARY AND CONCLUSIONS

Experiments were designed to test the efficacy of IMC, Univul DS49, sodium ascorbate and benzyl cinnamate for reducing sunlight inactivation of *Bacillus thuringiensis* (B.t.) and nuclear polyhedrosis virus (NPV). Potted and open grown white spruce trees were sprayed at known rates of B.t. and/or NPV suspension into which the protectants were incorporated. The amount of incident sunlight was recorded and correlated with biological persistence of the pathogens. DS49 sufficiently prolonged the viability of B.t. in these tests to warrant aerial trials in 1976.

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Table 1

Residual Activity of Dipel Wettable Powder Applied with
and without Sunlight Protectants to Potted Trees and Exposed to Direct Sunlight¹

No. of Days of Weathering	Controls-No Protectant Corrected % Mortality ²	Percent of Original Activity		
		0.1% Erio Acid Red	10% Inositol	10% Molasses
0 (Treatment Day)	100	100	100	100
1	100	98	100	95
2	91	76	74	56
4	98	71	86	61
8	62	33	18	38
16	51	21	23	50

¹ Means of 6 replicate trees per treatment and 50 third instar of Douglas Fir tussock moth (*Orgyia pseudotsugata*) larvae per tree. Mortality among untreated checks was 15% of 189 larvae.

² Corrected from mortality among checks.

Table 3

REARING CONDITIONS FOR BIOASSAYS - SUNLIGHT PROTECTANT TESTS

NO. OF LARVAE PER TEST : ~200 (20 LARVAE x 10 CAGES)

LENGTH OF BIOASSAY : B.t. FORMULATIONS - 7 DAYS
NPV FORMULATIONS - 14 DAYS

TEMPERATURE : 22 \pm 3°C

HUMIDITY : 65 \pm 5% R.H.

LIGHTING : 24 h./day, FLOURESCENT LIGHTS

Table 4

Meteorological Conditions During Exposure Periods - Sunlight Protectant Tests

INCLUSIVE DATES	CUMULATIVE SOLAR RADIATION (Kcal/cm ²)	TEMPERATURE		CUMULATIVE RAINFALL (cm.)
		MEAN MIN. (°C)	MEAN MAX. (°C)	
B.t. FORMULATIONS				
JULY 4 ONLY	0.57	11.7	27.2	0.0
JULY 4 - 6	1.76	13.3	29.6	0.1
JULY 4 - 10	3.75	12.9	28.5	0.5
JULY 4 - 17	7.12	13.1	27.8	1.6
NPV FORMULATIONS				
JULY 23 ONLY	0.60	15.0	27.8	0.0
JULY 23 - 25	1.10	15.2	23.9	1.3
JULY 23 - 29	2.78	11.5	23.7	3.0
JULY 23 - AUG. 5	6.06	-	-	-

Table 5

INCIDENCE OF B. t. IN CADAVERS - SUNLIGHT PROTECTANT TESTS
(EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED) *

TREATMENT	LENGTH OF EXPOSURE (DAYS)						
	IN SUNLIGHT					IN SHADE	
	0 d.	1 d.	3 d.	7 d.	14 d.	3 d.	14 d.
B.t. alone	85	56	0	8	-	64	44
B.t. + DS49	64	57	50	27	0	68	56
B.t. + BENZYL CINNAMATE	54	52	39	18	0	56	68
B.t. + SODIUM ASCORBATE	80	80	39	10	18	78	39
NPV alone	4	0	0	-	-	-	-
NPV + DS49	2	0	0	0	-	-	-
NPV + BENZYL CINNAMATE	0	0	12	-	-	-	-
NPV + IMC	0	0	0	0	-	-	-

* INCIDENCE IN POOLED CONTROLS = 0%

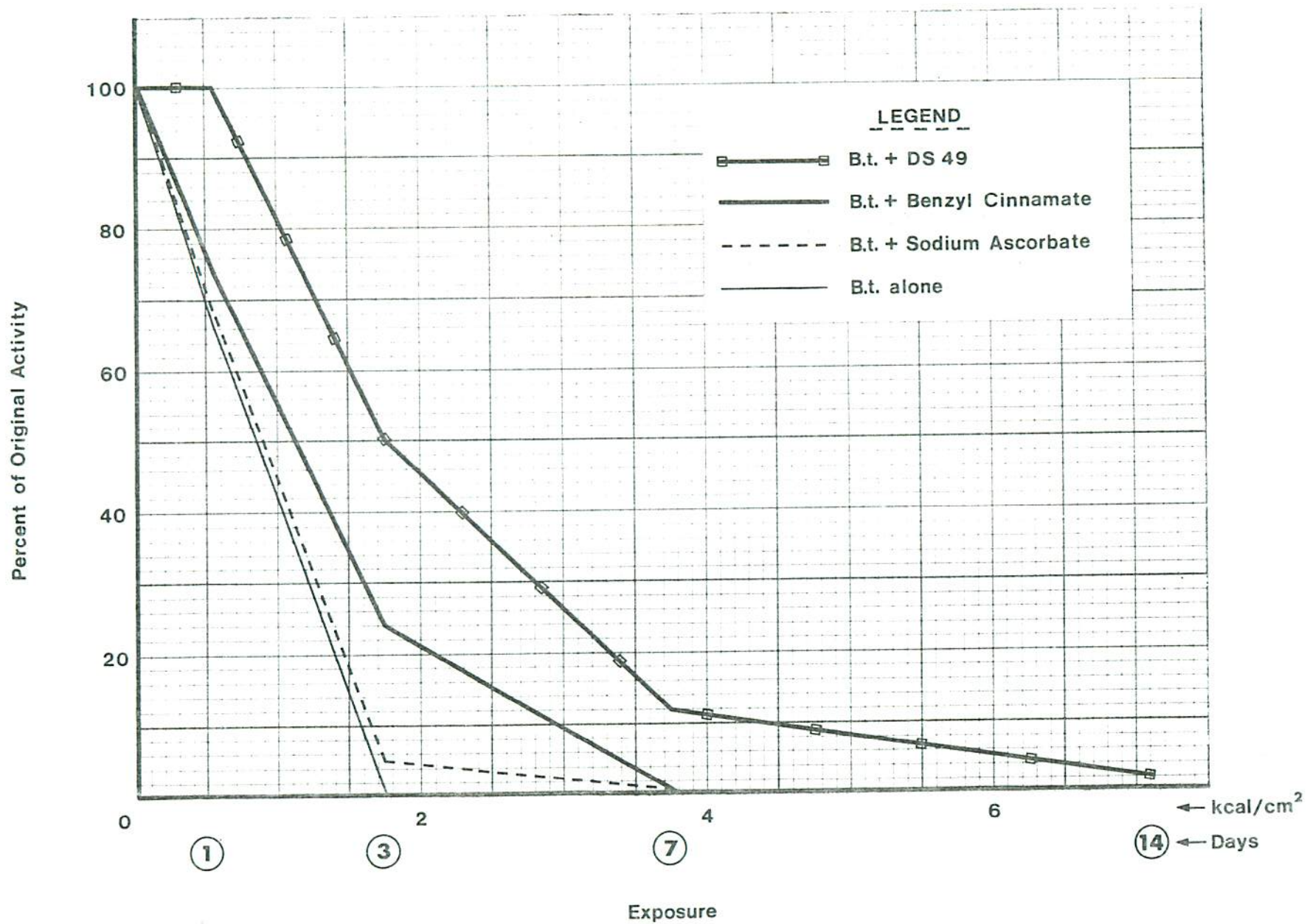


FIG. 1 Protection of *Bacillus thuringiensis* from sunlight inactivation

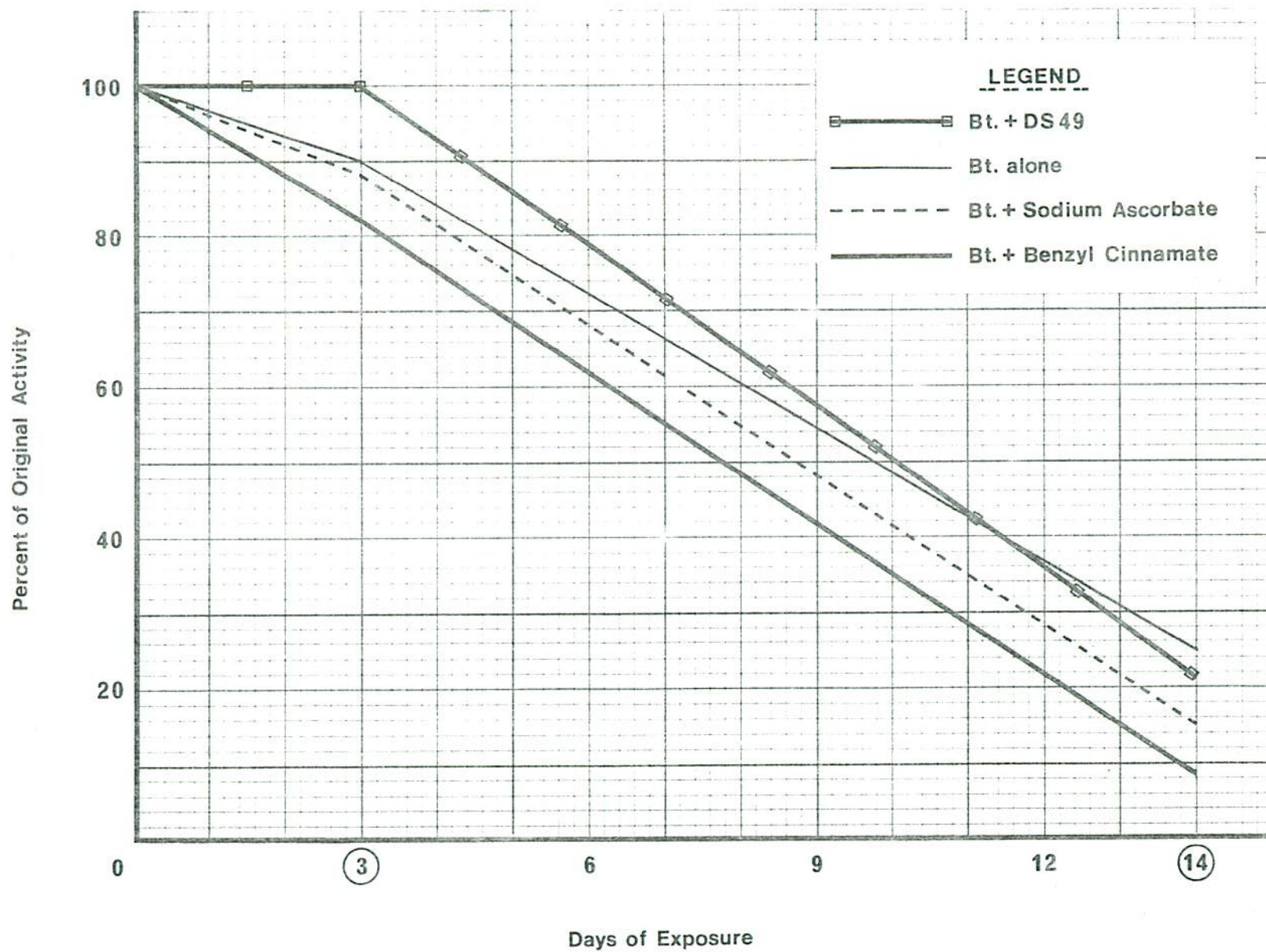


FIG. 2 Inactivation of *Bacillus thuringiensis* on white spruce unexposed to sunlight

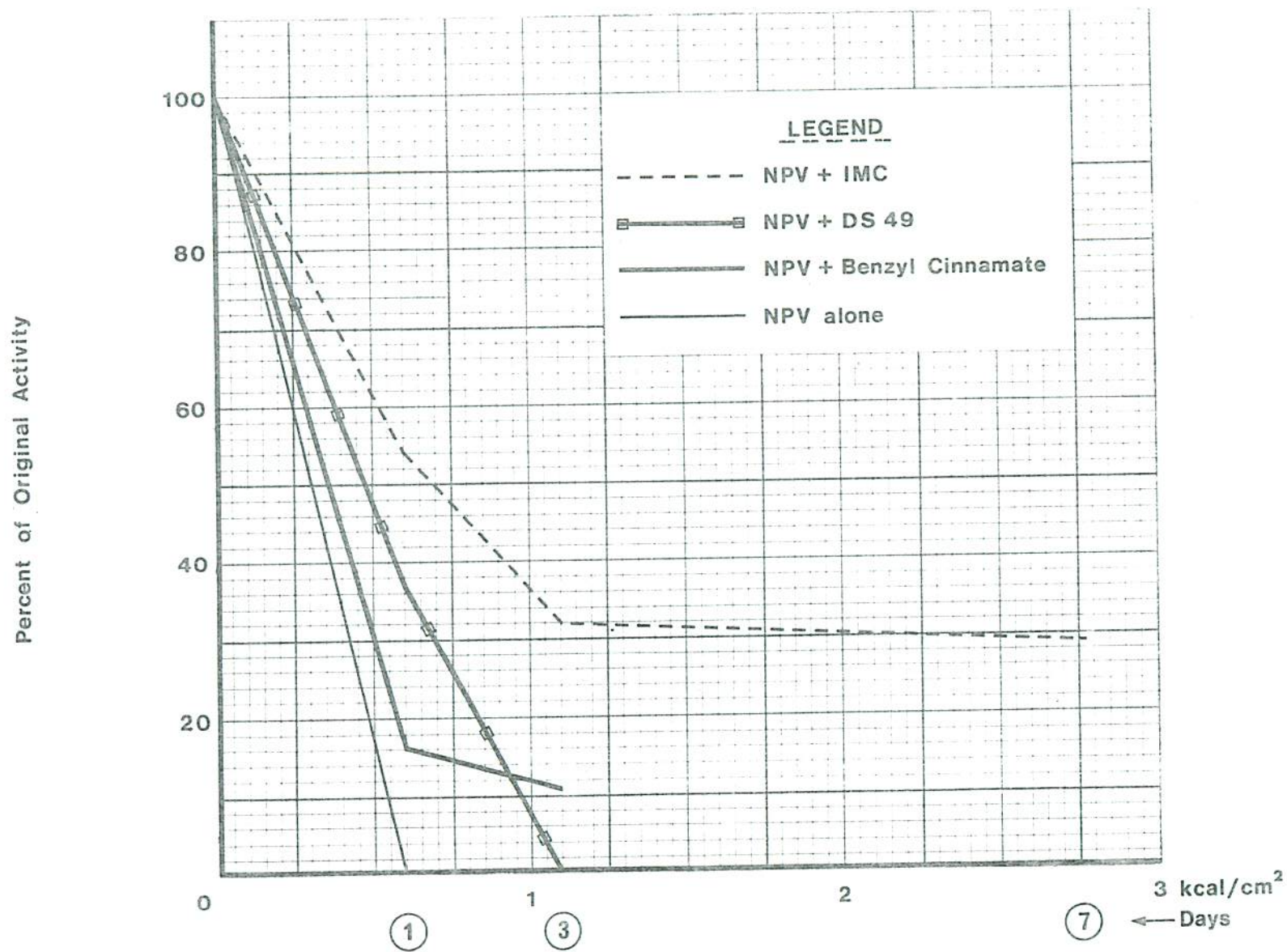


FIG. 3 Protection of spruce budworm nuclear polyhedrous virus from sunlight inactivation

Table 7

INCIDENCE OF MICROSPORIDIA IN CADAVERS - SUNLIGHT PROTECTANT TESTS
 (EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED) *

TREATMENT	LENGTH OF EXPOSURE (DAYS)							
	IN SUNLIGHT					IN SHADE		
	0 d.	1 d.	3 d.	7 d.	14 d.	3 d.	14 d.	
B.t. alone	61	72	44	25	-	20	33	
B.t. + DS49	82	57	10	12	29	16	39	
B.t. + BENZYL CINNAMATE	64	54	13	42	14	22	9	
B.t. + SODIUM ASCORBATE	72	20	22	30	45	10	11	
NPV alone	30	33	32	-	-	-	-	
NPV + DS49	28	38	18	33	-	-	-	
NPV + BENZYL CINNAMATE	21	36	38	-	-	-	-	
NPV + IMC	54	41	67	71	-	-	-	

* INCIDENCE IN POOLED CONTROLS = 67%

Table 8

INCEDENCE OF FUNGUS IN CADAVERS - SUNLIGHT PROTECTANT TESTS
(EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED) *

TREATMENT	LENGTH OF EXPOSURE (DAYS)						
	IN SUNLIGHT					IN SHADE	
	0 d.	1 d.	3 d.	7 d.	14 d.	3 d.	14 d.
B.t. alone	23	10	56	25	-	18	35
B.t. + DS49	22	28	20	31	71	40	6
B.t. + BENZYL CINNAMATE	16	20	46	17	43	52	18
B.t. + SODIUM ASCORBATE	28	34	33	70	82	26	32
NPV alone	34	67	42	-	-	-	-
NPV + DS49	18	44	18	22	-	-	-
NPV + BENZYL CINNAMATE	25	14	25	-	-	-	-
NPV + IMC	32	22	62	0	-	-	-

* INCIDENCE IN POOLED CONTROLS = 41%

Table 9

Mistblower Ground Spray 1975 - PFES

Spray Volume/tree - 1000 ml

Formulations for 500 ml of spray

	pH
Dipel 36B alone:	4.2
250 water + 5 gm PVP + 5 gm DS49	
250 ml Dipel 36B	
1 ml Biofilm (emulsifier)	
0.5 ml Chevron sticker	
Virus alone (mix thoroughly in blender just before use)	6.4
1 - gm Virus powder	
500 ml water + 5 gm PVP + 5 gm DS49	
1 ml Biofilm	
0.5 ml Chevron Sticker	
Virus + B.t.	6.3
250 ml water + 5 gm PVP + 5 gm DS49	
250 ml Dipel	
-1 gm Virus powder	
1 ml Biofilm	
0.5 ml Chevron sticker	
Virus + Chitinase	6.5
500 ml water + 5 gm PVP + 5 g DS49	
-1 gm Virus powder	
1 ml Biofilm	
0.5 ml Chevron sticker	
10 mg chitinase	
B.t. + Orthene technical	4.1
250 ml Dipel 36B	
250 ml water + 5 gm PVP + 5 gm DS49	
1 ml Biofilm	
0.5 ml Chevron	
0.5 gm Orthene	
NPV + Orthene Technical	6.4
500 ml water + 5 gm PVP + 5 gm DS49	
- 1 gm Virus powder	
1 ml Biofilm	
0.5 ml sticker	
0.5 gm Orthene	

Table 9 (Contd.)

B.t. + Pyrenone EC	4.1
250 ml Dipel 36B	
250 ml water + 5 gm PVP + 5 gm DS49	
1 ml Biofilm	
0.5 ml Chevron	
5 ml (0.5 gm A1) Pyrenone	
B.t. + pH 60 - 40 WP	4.1
250 ml Dipel 36B	
250 ml water + 5 DS49	
1 ml Biofilm	
0.5 ml Chevron	
7.8 gm pH 60 - 40	
"Untreated"	6.1
500 ml water + 5 gm PVP + 5 gm DS49	
0.5 ml Chevron	
1.0 ml Biofilm	
Pyrenone alone	6.1
500 ml water + 5 gm PVP + 5 g DS49	
1 ml Biofilm	
0.5 ml Chevron	
5 ml (0.5 g A1) Pyrenone	
Orthene alone	6.0
500 ml water + 5 g PVP + 5 g DS49	
1 ml Biofilm	
0.5 ml Chevron	
0.5 g Orthene	

Table 10

Percent Population Reduction of Spruce
Budworm Larvae on White Spruce Sprayed
by Mistblower with Bacillus thuringiensis
and Nuclear Polyhedrosis Virus Formulations Containing
Sunlight Protectants.

Treatment	Pre-Spray Larval Density per Bud	Corrected Population Reduction 21 Days Post-Spray ¹
Dipel 36 B + Orthene	0.27*	100.0
Dipel 36B + pH 60-40	0.16*	96.0
NPV alone	0.15*	56.0
NPV + Chitinase	0.32**	79.5
NPV + Orthene	0.51**	98.2
NPV + Dipel 36B	0.48**	50.0
Orthene alone	0.31**	67.2
Dipel alone	0.16**	100.0
Dipel + Pyrenone	0.16**	82.9
Pyrenone alone	0.25**	80.8
Untreated Check <u>I</u>	0.24**	(52.7)
Untreated Check <u>II</u>	0.35**	(77.1)

Treatment followed by * or ** are compared with each other.

¹ Corrected for mortality among checks by Abbot's formula.

Table 11

Defoliation Current Growth of Spruce Budworm
 Infested White Spruce Sprayed by Mistblower
 With Bacillus thuringiensis and Nuclear Polyhedrosis
 Virus Formulations Containing Sunlight Protectants

Treatment	Pre Spray Density Per Bud	Theoretical* Density	Theoretical % Defoliation	Actual Density	% Actual Defoliation **
Dipel 36B + Orthene	0.27	0.30	76.7	0.27	19.0 ab
Dipel 36B + pH 60-40	0.16	0.24	68.0	0.16	12.0 ab
NPV alone	0.15	0.24	68.0	0.15	26.0 abc
NPV + Chitinase	0.32	0.30	76.7	0.32	22.0 abc
NPV + Orthene	0.51	0.36	90.0	0.51	26.0 abc
NPV + Dipel	0.48	0.36	90.0	0.48	41.0 c
Orthene alone	0.31	0.30	76.7	0.31	25.0 abc
Dipel alone	0.16	0.24	68.0	0.16	9.0 a
Dipel + Pyrenone	0.16	0.24	68.0	0.16	11.0 ab
Pyrenone alone	0.25	0.24	68.0	0.25	13.0 ab

* Expected defoliation based on data from aerial spray untreated check plots, 25 white spruce trees 2 branches per tree.

** Means followed by the same letter and not significantly different at the 5% level.

Table 12

ANALYSIS OF VARIANCE - DEFOLIATION OF TREATED AND UNTREATED TREES AT RANKIN 1975 (GROUND SPRAY)

SOURCE OF VARIANCE	SUMS OF SQUARES	DEGREES OF FREEDOM	MEAN SQUARES	F RATIO
BETWEEN TREATMENTS	5880.9645	10	588.0965	9.2004 **
BETWEEN TREES WITHIN TREATMENTS	2552.8467	22	116.0385	1.8154 ns
BETWEEN BRANCH SAMPLES WITHIN TREES (ERROR)	2109.3750	33	63.9205	
TOTAL	10543.1862	65		

Table 13

INCIDENCE of BACTERIAL INFECTION in CADAVERS
(EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED)

	PRE-SPRAY	POST-SPRAY 1	POST-SPRAY 2
B.t. + ORTHENE	0	71	-
B.t. ALONE	2	90	67
NPV + ORTHENE	-	7*	-
B.t. + PH 60 - 40	0	83	67
B.t. + PYRENONE	8	89	57
B.t. + NPV	11	6	4
ORTHENE ALONE	5	19*	-
PYRENONE ALONE	6	7*	0
NPV + CHITINASE	10	0	0
NPV ALONE	0	30*	6*
UNTREATED	-	0	0

* Mostly non-spore forming bacteria. Probably some contamination.

Table 14

INCIDENCE of NPV in CADAVERS

(EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED)

	PRE-SPRAY	POST-SPRAY 1	POST-SPRAY 2
B.t. + ORTHENE	0	0	-
B.t. ALONE	0	0	0
NPV + ORTHENE	-	33	-
B.t. + PH 60-40	0	0	0
B.t. + PYRENONE	0	0	0
B.t. + NPV	0	44	73
ORTHENE ALONE	0	0	-
PYREONONE ALONE	1	14 *	45 *
NPV + CHITINASE	0	35	73
NPV ALONE	0	30	67
UNTREATED CHECK	-	0	12 *

* Source maybe contaminated during spraying or stimulation of latent virus infection by the chemical.

Table 15

INCIDENCE of MICROSPORIDIA in CADAVERS

(EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED)

	PRE-SPRAY	POST-SPRAY 1	POST-SPRAY 2
B.t. + ORTHENE	31	11	-
B.t. ALONE	26	0	33
NPV + ORTHENE	-	19	-
B.t. + PH 60 - 40	22	25	50
B.t. + PYRENONE	18	11	0
B.t. + NPV	44	39	32
ORTHENE ALONE	15	38	-
PYRENONE ALONE	27	43	36
NPV + CHITINASE	15	41	35
NPV ALONE	34	10	22
UNTREATED	-	30	12

Table 16

INCIDENCE of FUNGI in CADAVERS

(EXPRESSED AS A PERCENTAGE OF THE NO. OF CADAVERS EXAMINED)

	PRE-SPRAY	POST-SPRAY 1	POST-SPRAY 2
B.t. + ORTHENE	2	25	-
B.t. ALONE	17	10	17
NPV + ORTHENE	-	16	-
B.t. + PH 60 - 40	39	17	17
B.t. + PYRENONE	24	0	28
B.t. + NPV	22	17	18
ORTHENE ALONE	25	50	-
PYRENONE ALONE	19	43	0
NPV + CHITINASE	29	41	31
NPV ALONE	6	10	17
UNTREATED	-	50	29

Table 17

RESIDUAL ACTIVITY - RESULTS OF BIOASSAY OF BRANCH SAMPLES FROM MISTBLOWER TESTS COLLECTED 20 AND 27 DAYS
AFTER SPRAY APPLICATION

	% MORTALITY ^a	INCIDENCE ^b OF PATHOGENS IN CADAVERS (%)			
		B. t.	NPV	Micros.	Fungus
<u>20 DAYS POST-SPRAY</u>					
B. t. alone	95	42	0	89	13
B. t. + Orthene	100	36	0	89	11
B. t. + NPV	90	13	2	88	12
<u>27 DAYS POST-SPRAY</u>					
B. t. alone	95	34	0	92	8
B. t. + ORTHENE	81	24	0	89	4
B. t. + NPV	100	6	6	96	10
UNTREATED	0	4	4	100	0

^a Corrected for mortality in untreated checks by Abbott's formula.

^b Expressed as a percentage of the number of cadavers examined for each treatment.

Table 18

RESULTS OF REARING OF SURVIVING PUPAE FROM RESIDUAL ACTIVITY TEST

TREATMENT	SEX	PUPAL SURVIVAL			OVIPOSITION		
		MEAN PUPAL WT. (mg)	% MORTALITY*	% PARASITISM	% ADULT EMERGENCE	EGG MASSES PER ♀	% VIABLE
NPV ALONE	0	97	0	7	93	3.4	100
	0 ⁷	68	0		90		
NPV +CHITINASE	0	95	10	0	73	3.5	100
	0 ⁷	69	19		72		
UNTREATED	0	89	0	6	83	1.1	94
	0	59	0		89		

* Corrected by Abbott's formula; includes mortality due to parasitism.

Table 19

Mistblower Applications of B.t. and NPV with and without Sunlight Protectants
against Spruce Budworm on White Spruce

Treatment	Pre-spray Larval Density		Percent Defoliation		Positive Difference
	Per 18" branch	Per Bud	Expected	Observed	
B.t. alone	54.4	0.51	78.0	67.0	11.0
B.t. + 1% DS49 + 1% PVP	11.5	0.15	37.0	9.0	28.0
NPV alone	68.0	0.88	86.0	97.4	-
NPV + 1% DS49 + 1% PVP	16.5	0.33	48.0	26.0	22.0
B.t. + NPV alone	52.0	0.83	75.0	90.9	-
B.t. + NPV + 1% DS49 + 1% PVP	26.2	0.47	58.0	41.0	17.0
B.t. + NPV + 1% Benzyl Cinnamate	45.7	0.40	73.0	59.0	14.0
Check	35.1	0.34	72.0	80.0	-

DEFOLIATION

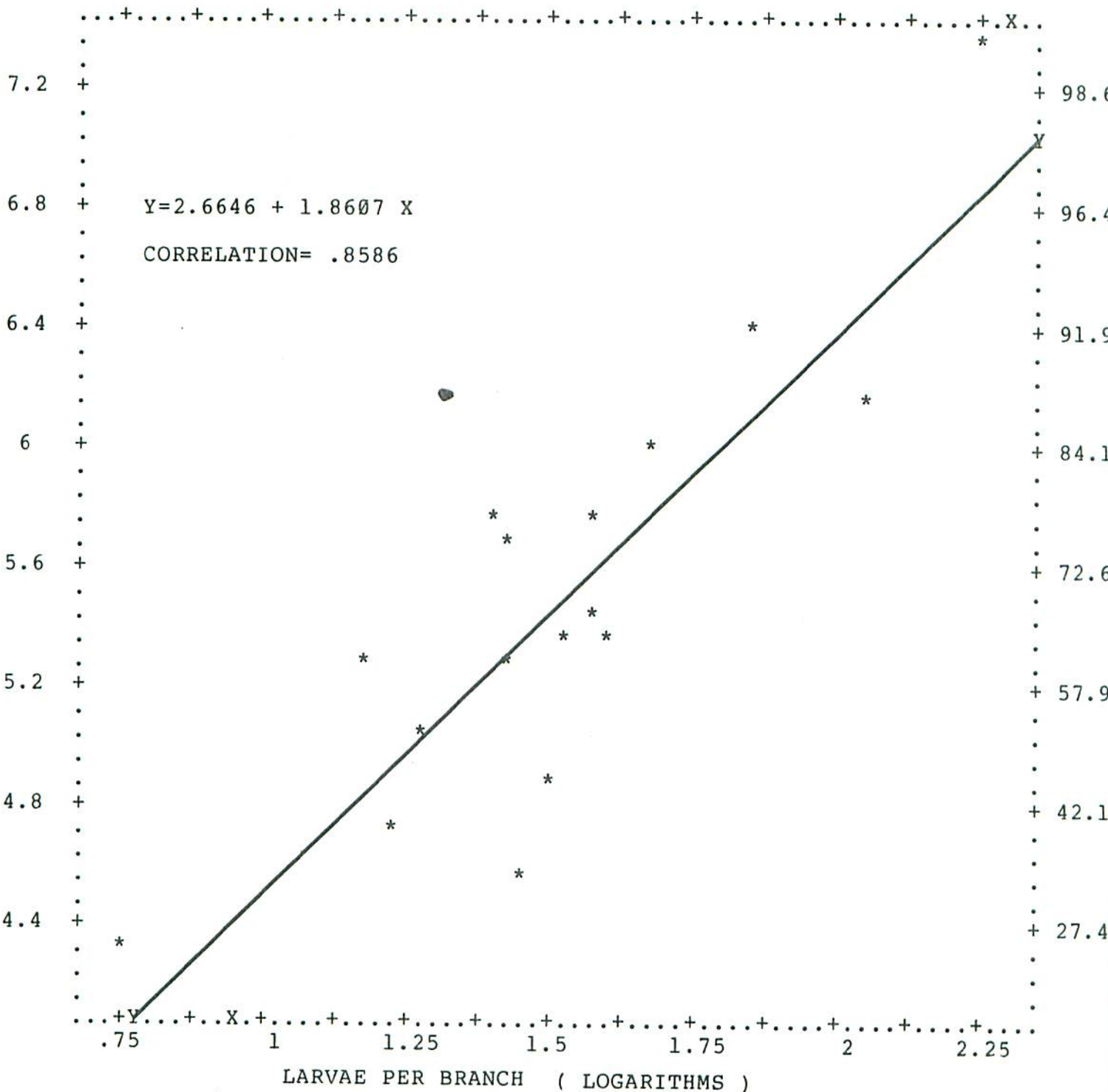


FIG. 4 Relationship of spruce budworm population density to percent defoliation of white spruce (current year's growth) based on data from 17 unsprayed plots in Ontario. (50-100 18 inch branch tips per plot)