RECENT FIELD EXPERIENCES WITH *BACILLUS THURINGIENSIS* IN CANADA AND RESEARCH NEEDS

Oswald N. Morris

Research Scientist, Agriculture Canada, Research Station, 195 Dafoe Road, Winnipeg, Manitoba, Canada, R3T 2M9

The CANUSA working group on the use of B.t. against the spruce budworm has prepared a document entitled "Guidelines for the operational use of *Bacillus thuringiensis* (B.t.) against the spruce budworm" following six years of extensive cooperative field trials in Canada and the U.S.A. (Morris et al 1984). The document summarized below (Table 1) describes the current state of the art and is based on the concensus of a committee of scientists actively involved in the management of this insect pest in the two countries.

TABLE I. Technical guidelines for the operational use of *Bacillus thuringiensis* against the spruce budworm - summary

1. Potency: Be sure actual potency is as claimed on label.
2. Aircraft: Calibration essential. Use actual tank mix.
3. Mixing sequence: Water then sticker then B.t.
4. Sticker: As recommended by B.t. manufacturer.
5. Dye: None for operational use.
6. Dosage rate: 12 BIU/AC (30 BIU/ha)
7. Volume rate: .25 to 0.5 gpa (2.37 to 4.7 l/ha).
8. Application timing: When buds start to flare.
9. Ground droplet density: Aim for greater than 25 drops/cm^2.

There are several items in this summary and elsewhere which to my mind requires further study with a view to improving the effectiveness of B.t. I intend to discuss briefly these items, to present some recent field results, and to offer some recommendations for research.

But firstly an update on the operational use of B.t. against the budworm in Canada.

Status of Operation Trials

The summary presented in Table 2 shows quite clearly an increasing use trend between 1979 when the cooperative CANUSA program started and the proposed 1984 use. Between 1979 and 1980, the proportion of forests sprayed with B.t. was very low (1.5 to 2%). The proportion increased to 9% in 1983 and to 14.1% (proposed) in 1984. This represents a substantial increase of B.t. use between the beginning and the end of the co-operative program on the whole, a trend which I expect will continue.

Table 2. Approximate areas (ha) of forest operationally sprayed for spruce budworm control in Canada between 1979 and 1984 (proposed).

Year	Chemical Insecticides	B.t.	% by B.t.
1979 + 1980	4,024,227	83,773	2
1982	2,265,000	34,000	1.5
1983	86,650	7,600	9.0
1984 (Proposed)	2,094,900	343,000	14.1

A summary of the use trend by province (Table 3) shows that Ontario, Nova Scotia and Quebec were the largest users of B.t. between 1979 and 1984 (proposed) based on the percentage of their sprayed areas treated with B.t. New Brunswick also increased their B.t. treated areas but in terms of acreage actually treated with the microbial insecticide the increase was not large. Newfoundland dropped precipitously from 44% in 1979 + 1980 to 1% (proposed) in 1984.

Table 3. Areas (ha) operationally treated by chemical pesticides and B.t. against the spruce budworm in 1979 + 1980 and in 1984.

Province	1979 + 1980 Total	1979 + 1980 % Bt.	1984 (Proposed) Total	1984 (Proposed) % Bt.
ONT.	49,710	9.5	10,000	80.0
N.B.	3,252,360	0.01	1,930,000	2.1
NFLD.	35,619	43.9	71,300	1.1
N.S.	63,276	47.8	35,000	100
QUE.	791,377	4.2	815,800	32.6
TOTAL	4,191,773	2.0	2,862,100	12.2

Effect of B.t. on budworm biomass

It is generally acknowledged that the non-lethal effects of B.t. treatments are important additional criteria for judging the effectiveness of the microbial agent. This type of effect was recently demonstrated in the field against the budworm in balsam fir stands (Morris and Moore 1983). The data summarized below (Table 4) were gathered from experiments carried out at Riviere du Loup, Quebec in 1981. Note that larval biomass in B.t. treated populations were significantly lower than in the control and the Dipel-vehicle-alone treated populations. There was no difference between the check and the Dipel-vehicle plot. This plus the data on population reductions and defoliations demonstrate that the vehicle had no effect on the population as was suspected by one worker. We also found that the Dipel-vehicle had no demonstrable effect on the incidence of larval and pupal parasitisim (Morris 1983).

Table 4. Effect of Thuricide 16B® and Dipel 88® on spruce budworm larval biomass, larval population reduction and defoliation of balsam fir.

Treatments	Larval Biomass (Mg/100 shoots)	% Reduction	% Defol.
Thuricide 20 BIU in 9.4 L/ha	0.86	36a	20±11a
Dipel, 20 BIU in 9.4 L/ha	0.69	36a	21±16a
Dipel vehicle 25%, 9.4 L/ha	44.2	0b	66±16b
Untreated checks	34.2	0b	65±1ab

Potency

Several researchers and operators have received in the past from manufacturers B.t. products which were below labelled potency. Rarely does one have enough time between receipt of the product and the time to apply it to assess labelled potency. This problem, in addition to causing significant errors in experimental applications can create serious credibility gaps between the product user and the manufacturer. I believe some attempt has been made by the commercial companies to collaborate on the standarization of their product assays but as far as I know there is still no agreement among them. The problem occurred as recently as 1982 when product potencies as determined by suppliers were far in excess of actual potencies as determined by independent laboratories. I want to implore the producers to come to some general agreement among themselves on bioassay technique for monitoring their products. This technique should be one that is fairly easy to use by researchers, independent laboratories, and operators.

I believe that this bioassay problem is due not only to differences in basic technqiue but also to differences between test insects. Even "minor" genetic differences between and within insect populations may result in differences in their susceptibility to microbial agents. This admitedly makes a standard bioassay procedure rather difficult. One possible way of solving this dilemma is to complement insect bioassay with the enzyme-linked immuno absorbent assay (ELISA) for B.t. endotoxin. The development of this technique while not replacing insect bioassay entirely would be valuable not only for assaying the toxin content of commercial products but also for rapidly assaying tank mixes and field deposits over the short or long term at the ground or tree level. The method is rapid, specific, sensitive and reproducible and I believe, if used, could reduce the variability normally encountered in bioassays. I have no doubt that this has been considered by B.t. and virus producers, so I would be interested to know why it has not been developed to the stage required.

During the 1970's and 1980's, the commercial companies have made a valiant effort to manufacture more highly concentrated B.t. products and have been quite successful in doing so. Products of 48 BIU/US gal. have already been field tested and 64 BIU products are already on the market or will soon be. The obvious advantage to highly concentrated products relates to cost reduction. Much of the cost of earlier materials was due to bulk shipment and this could be reduced by shipping high concentrate products. The other advantage of such high concentrates is the fact that B.t. is most effective when the target trees are densely covered with droplets containing a high concentration of the toxicants.

Table 5 presents data (from Drs. Fast and Sundaram, FPMI) on the relationship between droplet density of B.t. on coniferous foliage and spruce budworm larval mortality. Note that larval mortality at the lowest dosage (8 BIU/ U.S. gal) is highly variable regardless of

coverage. This supports my contention that 8 BIU /acre is only marginally effective for both spruce budworm and gypsy moth control.

However, just 3 drops/needle at 12 BIU/gal gave acceptable mortality. A 12 BUI/acre dosage rate is gaining general acceptance by operators. Thus if B.t. is commercially formulated in concentrated form so that the 12 BIU/acre will alow a high level of atomization (VMD circa 60 mm), low evaporation rate and dense coverage at the feeding site, then the cost-benefit relationship in forestry applications should be entirely acceptable.

Table 5. Relationship between droplet density on coniferous foliage and spruce budworm larval mortality following aerial application of Bacillus thuringiensis[a]

Concentration Sprayed	Drops/Needle	% Mortality (50 larvae)
2.1 BIU/l	1	20 (16-24)
(8 BIU/US gal)	3	38 (30-46)
	6	31 (8-70)
3.2 BIU/l	1	55
(12 BIU/US gal)	3	84
5.3 BIU/l	1	78 (70-88)
(20 BIU/US gal)	3	96 (92-100)
	6	100

[a]Courtesy of Drs. P. Fast and A. Sundaram (FPMI)

The question of the most appropriate droplet size for maximum effectiveness against the spruce budworm is also unanswered at this point. It would be useful to know what droplet size one could expect with a particular set of formulation, atomizer and weather conditions but we are still in a state of confusion on this score. To illustrate the problem, Table 6 gives the droplet analyses of undiluted Thuricide and Dipel of drops collected on Kromekote cards during aircraft calibration in 1982.

Table 6. Droplet analysis of spray deposits of undiluted Thuricide and Dipel collected on kromekote cards during aricraft calibration[a]

Criteria	Thuricide 48B	Dipel 48B	Thuricide 32B	Dipel 32B
Spread Factor Mean±SE	1.2±0.09	1.2±0.01	1.2±0.09	1.2±0.08
D_{min}(M)	<25	17	<25	17
D_{max}(M)	233	302	323	417
NMD (M)	50	41	155	163
VMD (M)	100	66	205	207

[a]Applied with AU3000 Micronairs

We see that the droplet maximum diameters (D_{max}) of undilated Dipel 48B and 32B were larger that those of undiluted thuricide 48B and 32B suggesting that the atomization of Thuricide was superior to that of Dipel. Note, however, that the differences between NMD and VMD (which reflect uniformity of the droplet spectrum) were identical for the two products (i.e. 50 um) indicating that the atomization of the sprays were in fact similar, in spite of Dmax differences. This unexpected similarity is a bit of a conondrum but may have something to do with the viscosities of the products. Viscosity will directly affect droplet size range produced during atomization and also affects indirectly droplet size at the impingement site because it affects the rate of evaporation during descent of the droplet. Dipel acts like a typical non-Newtonian fluid. The apparent viscosities of such fluids can be dramatically reduced depending on sheer rate. Research is needed with non-Newtonian products to determine the most suitable apparent viscosities for a variety of nozzle types and sheer rates. Some has already been done but to my knowledge has not been published. Such information would help to predict drop-size spectra for a variety of spray systems.

Lastly, when 32B amd 48B preparations are compared, the difference between NMD and VMD for Dipel 48B is 25 but is 44 for Dipel 32B. The reason for the apparent difference in behavior between 32B and 48B products is not clear to me. Perhaps manufacturing representatives can explain this.

Stickers

In the CANUSA technical guidelines, we recommended that a sticker be used in all presently manufactured B.t. product tank mixes. We could not find enough well documented information on the effectiveness of various stickers for aerial application and on their compatibility with B.t. to make any general recommendation. We were also aware that different types of formulations may require different types of stickers. We therefore, suggested that operators should follow the recommendation of the product manufacturers.

Table 7. Some stickers used in the aerial applications of microbial agents over forests.

Product	Sticker	Conc./US gal.
Thuricide, Dipel	Chevron	2.365-3.785 Ml
	Dupont	
	Spread sticker	2.42 ml
	Biofilm	4.73 ml
	Rhoplex B60A	75.7 ml
NPV	Biofilm	4.73 ml

Table 7 presents some stickers commonly used in aerial applications of microbial agents over forests. None of these have been fully accepted by operators or researchers in terms of their effectiveness. There is evidently an information gap here which B.t. producers in particular would be well advised to explore. Acrylic stickers (e.g. Rhoplex) are now entering the market and they do appear to be superior to older types of stickers in terms of effectiveness. Other acrylics like 5% Bond and 3% Acrylocoat are highly effective under laboratory conditions (Win McLane, Otis Air Base) when applied with B.t. or NPV against gypsy moth on red oak seedlings. There are, however, several criteria which should be assessed before a sticker is recommended:

1. It must be determined that the sticker does not delay or inhibit the availability of the microbial agent when it is ingested by the target pest. It should be easily degradable in various liquid environments simulating the insect gut contents of a variety of pest species.

2. It should be non-toxic for the microbial agent. Some microbial agents seem to be particularly sensitive to chemical agents. Microsporiadia in my experience is one such group and nematodes another.

3. Its use should not be limited to certain water types or qualities because the operator often has little choice in terms of the water he must use as diluent.

4. Its chemical nature should not be altered significantly under conditions of high shear produced by aircraft spray systems. In 1982, the use of Phoplex in B.t. tank mixes in Canada put out of action our aircraft flow meter and gummed up the spinning disc cage of our Micronairs delaying our first-application by one day under good weather conditions. Since polymers tends to introduce thixotropic properties to a liquid formulation, some knowledge of their effect on viscosity-shear rate would be helpful for producing desired drop-size spectra during aerial application.

In summary, it seems to me essential that the physical, chemical, biological and toxicological properties of stickers should be tailor-made to achieve maximum spray efficiency with microbial insecticides.

Tracer dyes

The CANUSA guidelines recommended that no tracer dyes be used in operational applications, however, if a dye is needed the recommendation of the manufacturer should be followed. Dyes are sometimes called for in semi-operational amd often in experimental trials. As far as I know, the commercial suppliers are not fully agreed on an acceptable tracer dye. A large number of dyes have been tested as B.t. tracer, but, as in the case of stickers, very little information exists on their compatibility with microbial agents, their chemical stability following release from the aircraft, the

---

Footnote:

NMD = The diameter that divides the number of droplets into two equal groups - 50% of the droplets have a diameter above the NMD and 50% below.
VMD = the droplet size diameter that divides the spray volume into equal parts - 50% of the volume is in droplets below the VMD and 50% above.

maintenance of their integrity on the surface of foliage and their environmental acceptability.

Table 8 gives a list of the most commonly used tracer dyes for aerial applications of microbials over forests.

Table 8. Some tracer dyes used in aerial applications of microbial agents over forests.

Product	Dye	Con./US gal
Thuricide,Dipel[a]	Rhodamine B	3.785 g
	Nigrosine	7.57 g
	Brilliant Sulphoflavine	3.785 g
	Erio Acid Red X B400	0.095 g
Tussock Moth-NPV	Rhodamine B	3.785 g
	BSF	3.785 g

[a]Water-based

Table 9 presents data on the compatibility of the dyes with B.t. under laboratory conditions (Morris 1980).

Of the 5 dyes shown, only Rhodamine B had any apparent deleterious effect on spore viability but the half-life was so long, that this effect is considered practically insignificant. Rhodamine B, however, has been reported to induce mutations in Salmonella and DNA damage in hamster ovarian cells (Nestmann and Kowbel 1979; Nestman et al 1979) making it suspect environmentally.

Table 9. Decay rates of viable spores of Thuricide 16B diluted 1:1 in water and exposed to 0.1% of various tracer dyes for 21 days.

Dye	Decay Constant (R)	1/2 life-days (95% CL)
Thuricide alone	0	-
Rhodamine B	0.036±0.015	19(10-150)
Brilliant Sulpho Flavine	0	-
Nigrosine	0	-
Erio Acid Red X B400[a]	0	-

[a]No significant decay for 69 days.

I have used successfully Erio Acid Red XB, a red fluorescent dye chemically related to Rhodamine B (4,5 dihydroxy-3-(P-nitro benzyl) azo 12,7 naphthalene disulphonic acid disodium salt. It is not mutagenic for bacteria. Toxicity data from Ciba Geigy indicate rat oral acute toxicity of 5000 mg/kg; 96 hr fish toxicity-LC_{50}, 1000 gm/l; bacterial activity - no inhibition at 300 mg/l.

Table 10 gives data on the toxicity of EAR for spruce budworm larvae using a foliage dipping technique. No significant toxicity to the larvae could be detected even at a concentration 1000x that used in field tank mixes (25 mg/l).

Table 10. Toxicity of Erio Acid Red XB 400 for L_4-L_5 larvae of the spruce budworm; 5 replicates.

Conc. of Dye (g/l)	Pooled No. of larvae	Adjusted % Mortality
0.025	109	0
0.25	106	3.6
2.5	115	0
25.0	103	0
Check	99	-

Table 11 gives data on the bioassay of Thuricide with and without the dye against L_5 larvae. The "needle inoculation" method was used in which known dosages of B.t. were fed to the larvae on balsam fir needles. The dye had no apparent effect on larval mortality. Because this dye is fluorescent, it has been used successfully in detecting very small droplets of B.t. on coniferous foliage under fluorescent optics. It should be tested with other microbial agents.

Sunlight screens

Several workers have demonstrated that B.t. and other entomopathogens are rapidly inactivated by sunlight and some have shown that the residual activity of some of these agents can be enhanced by the addition of

Table 11. Bioassay of Thuricide 16B with and without Erio Acid Red XB400 (0.25 g/l) against 5th instar spruce budworm[a].

Dosage/larva	No.	Adjusted % Mortality	Statistics
3.1 IU	49	49	
3.1 IU + EAR	49	49	x^2=2.026, IDF
31.0 IU	49	88	
31.0 IU + EAR	49	93	x^2=0.544, IDF
Check	49	-	

[a]Single needle inoculation method

sunlight-screens to microbial formulations. Nearly all the published reports so far relate to agricultural systems and only a few of them have done their tests under natural sunlight. Most recently Shapiro et al (1983) demonstrated that benzylidine sufonic acid enhanced the persistence of NPV 3-fold under sunlamps in the 320 to 400 nm range. We designed experiments to find protective materials which could be added to commercial products or to tank mixes intended for aerial application over forests.

Table 12. The effect of sunlight screens on the residual insecticidal activity of Dipel 36B applied to 2 replicate white spruce trees.

Formulation	Slopes[a]	R1/2 in Kcal/ cm^2[b]	Absorb Max. (nm)
Dipel alone	-0.202a	1.8	270
1% DS49+CYASORB + 0.5% CMC	-0.148ab	2.4	282,320
1%DS49 = 1% UVITEX + 0.5% CMC	-0.087ab	4.1	282,330
DS49 + 0.1% EAR + 6% MOLASSES	-0.069b	5.3	282,320 400,565
1% DS49 + 0.1% EAR + 1% PVP + 0.5% CMC	-0.129ab	2.8	282,328 565

[a]Estimates followed by the same letter are not significantly different at 5% level (SNK)
[b]R1/2 represents the amount of radiation required to reduce the potency by 50%.

The data in Table 12 summarizes the results of experiments designed to test the effect of sunlight screens on the residual activity of Dipel 36B applied to white spruce trees. Of the four formulations of sunscreens tested, DS49+ EAR+ molasses had the largest number of absorbance peaks within the solar wavelengths, the highest R/2 (the amount of solar radiation measured in KCal/cm^2 required to reduce the potency of B.t. by 50%) and a slope that was significantly different from that of Dipel without sunlight screens.

This sunscreen formulation was aerially tested against the spruce budworm in balsam fir stands. The results are summarized in Table 13.

Table 13. The effect of the addition of sunlight screens[a] to commercial Bacillus thuringiensis (Thuricide 32BX) on larval population reduction of and defoliation by the spruce budworm in balsam fir stands.

Dosage rate BIU/ha	% Larval Population Reduction at Days Post-application 14	21	Percent Defoliation[b]
10	0b	9b	69d
10+screen	67a	79a	39ab
20	63a	68a	45bc
20+screen	67a	88a	55cd
Check I	0b	0b	71d
Check II	0b	0b	89d

[a]1% Uvinul DS49 (Sodium 2,2'-dihydroxyl-4,4'-dimethoxy-5-sulfobenzophenone) +0.1 Erio Acid Red B 100 (sodium salt of formyl-m-benzenedisulphonic acid).
[b]Defoliation values followed by the same letter are not significantly different at 5% level.

In these tests molasses was omitted from the mixture because of the already high sugar content of the Thuricide used.

The results indicate that at the lower dosage rate of B.t. (10 BIU/ha), the effectiveness of Thuricide containing the sunlight screens was significantly greater than that of Thuricide without the screens. The screening effect was apparently masked by the higher level of active ingredient at the 20 BIU/ha application rate. Thus, it may be possible to reduce the dosage rate applied by extending the residual life of the B.t. on forest trees.

In summary, while the effectiveness of currently produced B.t. has improved greatly over that of the older products, greater effort on the part of manufacturers to formulate their products with sunlight screens to enhance residual activity seems fully justified to me.

Table 14. Effect of single vs. double application of the same dosage rate on population density reduction in and defoliation of balsam fir stands.

Treatment	Colonies per cm^2	% Population Reduction[b]	% Defoliation
Dipel 2x10	82	91ab	35a
Dipel 1x20	42	84ab	24a
Thuricide 2x20	71	81ab	35a
Thuricide 1x40	77	95ab	36a
CH-I	--	0c	71b
CH-II	--	0c	89d

[a]Dipel population density/100 shoots, 23 and 24; Thuricide, 23 and 27.
[b]Final post-spray corrected for pupal mortality. Values followed by the same letter are not significantly different at 5% level (ANOVA, Tukey's).

Single vs. double applications

Double or split applications of B.t. are generally accepted as necessary in mixed stands of white spruce, Picea glauca and balsam fir, Abies balsamea, but the strategy of applying split applications of B.t. in unmixed stands has been in question. Aerial applications of Dipel and Thuricide in spruce budworm infested balsam fir stands were designed to answer this question. The results summarized in Table 14 indicate that split applications are not significantly more effective than single ones of the same dosage as measured by defoliation and population reduction (Morris 1984).

Dosage and Volume

The question of the most effective and appropriate dosage and volume for aerial application against the spruce budworm was the subject of a cooperative field research project between Canada and the U.S.A. two years ago. In Canada, doses of 10, 20, 40 and 80 BIU in 9.4 l/ha and 30 BIU in 2.4, 4.7 and 9.4 l/ha were applied against budworm populations ranging in density between 10 and 36 fourth instar larvae per 45 cm branch in balsam fir stands. The results of the dosage test (Table 15) show that while larval population reduction and defoliation were statistically similar between 20 and 40 BIU/ha, considering the acceptable limits of 75% population reduction and 50% defoliation of current year's growth, the 20 BIU/ha treatment was only marginally acceptable. When the values were adjusted for differences in population densities by calculating the ratios of larval density to population reduction and % defoliation, it is apparent that the efficacy of the treatments increased with dosage applied. This supports the recommendation of 30 BIU/ha as being likely to give more consistently acceptable results than 20 BIU/ha.

Results of the tests on volumes (Table 16) indicate that the volumes applied between 2.4 and 9.4 l/ha had no significantly different effect on population reduction and defoliation. In fact only the treatment at 2.4 l/ha gave less than 50% defoliation. These data support the CANUSA recommendation of applying B.t. in volumes betweem 2.4 and 4.7l/ha.

Table 15. Population reduction and defoliation in balsam fir stands treated with various doses of Thuricide 32B in 9.4 l/ha.

Dosage BIU/ha	Larval density/45 cm branch	Population reduction (%) 21 days post-application	Defoliation (%)	Ratio Larval density/ pop. red.	Ratio Larval density/ % defol.
Check	14	1 d	80 b	14.0	0.18
10	16	9 cd	69 b	1.8	0.23
20	12	68 ab	45 a	1.8	0.27
40	16	95 a	36 a	1.7	0.44
80	36	95 a	61 b	0.4	0.59

Table 16. Population reduction and defoliation in balsam fir stands treated with various volumes of Thuricide 48B at 30 BIU/ha.

Volume/ha	Larval density/45 cm branch	% population reduction 21 days post-application	% Defoliation	Ratio: Larval density/pop. reduction	Ratio: Larval density/% defoliation
Check	18	75 a	86 a	0.24	0.21
2.4	10	68 a	45 b	0.15	0.22
4.7	17	72 a	59 ab	0.24	0.29
9.4	16	76 a	56 ab	0.21	0.29

[a]Three replicates

To summarize this discussion as a whole, I offer the following recommendations:

1. Every effort should be made by B.t. manufacturers to guarantee the potency of their product to the satisfaction of their clients.
2. Manufacturers of B.t. ought to continue their already valiant efforts to formulate their products in highly concentrated form in order to reduce application costs to users.
3. Stickers should be well researched for their effectiveness, bioavailability of microbial agents with which they are mixed and their compatibility with various formulations and spray equipment before they are marketed or recommended.
4. While tracer dyes for B.t. are not generally recommended for operational B.t. sprays, they are sometimes needed for semi-operational and experimental trials. Such dyes should be non-toxic to target insects, to the microbial agents with which they are mixed and should be environmentally acceptable. Erio Acid Red is suggested as an acceptable dye.
5. I recommend that a concerted research effort be put in place by B.t. manufacturers to find effective sunlight screens which can be added to their commercial formulations or to tank mixes.
6. Information on the most effective droplet sizes for spruce budworm control is sparce. More research is needed in this area.

LITERATURE CITED

Morris, O.N. Report of the 1979 CANUSA cooperative Bacillus thuringiensis spray trials. Canadian Forestry Service Rept. #FPM-X-40, 61-63. 1980.

Morris, O.N. Comparative efficacy of Thuricide 16B® and Dipel 88® against the spruce budworm, Choristoneura fumiferana (Lepidoptera:Tortricidae) in balsam fir stands. Can. Ent. 115: 1001-1006; 1983.

Morris, O.N., Moore, A. Changes in spruce budworm, Choristoneura fumiferana (Lepidoptera:Tortricidae), biomass in stands treated with commercial Bacillus thuringiensis var. kurstaki. Can. Ent. 115: 431-434; 1983.

Morris, O.N. Comparative efficacies of single and double applications of commercial Bacillus thuringiensis against the spruce budworm, Choristoneura fumiferana in balsam fir stands. Can. Ent. 116: 101-102; 1984.

Morris, O.N., Dimond, J.B., Lewis, F.B.. Guidelines for the operational use of Bacillus thuringiensis against the spruce budworm. U.S. Department of Agriculture, Forest Service Handbook #621; 1984.

Nestmann, E.R., Douglas, G.R., Matulka, T.I., Grant, C.E., Kowbel, D.J. Mutagenic activity of Rhodamine dyes and their impurities as delected by mutation induction in Salmonella and DNA damage in Chinese hamster ovary cells. Cancer Res. 39: 4412-4417. 1979.

Nestmann, E.R., Kowbel, D.J., Mutagenieity in Salmonella of Rhodamine WT, a dye used in environmental water-tracing studies. Mutation Res. 68: 389-392, 1979.

Shapiro, M., Agin, P.P., Bell, R.A., Ultraviolet protectants of the gypsy moth (Lepidoptera:Lymantriidae) nuclear polyhediosis virus Environ. Entomol. 12: 982-985. 1983.