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EVALUATION OF THE TOXICOLOGICAL EFFECTS AND FIELD EFFICACY OF AN INSECTICIDAL SOAP AGAINST THE SPRUCE BUDWORM (LEPIDOPTERA: TORTRICIDAE)

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Abstract

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Fossil Flower's insecticidal soap (IS) was tested in laboratory and field studies against the spruce budworm, *Choristoneura fumiferana* (Clemens). In the laboratory, a 4% solution (v/v) of formulated IS had both acute and sublethal effects. Although there was variability in observed sublethal effects among different instars and treatments, in general, the treatments significantly (p=0.05) increased larval mortality, reduced the dry weight of frass pellets, delayed pupal development for 6th instars, reduced pupal weight and emergence, and lowered female fecundity. Sixth instars preferred the diet which had IS incorporated rather than the control diet without IS. Pupal weight was reduced more when larvae ingested IS than when IS was applied topically. Based on corrected mortality, more early instars (3rd and 4th) died when treated with IS than late instars (5th and 6th). In the laboratory, the larval parasitoid, *Apanteles fumiferana* Viereck, completed development normally and suffered mortality only when its host was killed by the treatments. In the field, single and double applications of 0.4% and 2.0% IS failed to reduce spruce budworm populations or provide foliage protection although these applications may have had a limited effect on larval development, emergence, and pupal parasitism. The current recommended rates of IS would not effectively control spruce budworm.

Introduction

The insecticidal properties of fatty acids and their soaps have been recognized since the turn of the century; however, their effectiveness against greenhouse and ornamental pests has only been demonstrated in the last 15 years (Pinnock *et al.* 1974; Puritch *et al.* 1982; Abbasi *et al.* 1984; Osborne and Petitt 1985; Wilson and Moore 1986). Recent bioassays suggest that insecticidal soaps may be useful against some insects that defoliate forests. Puritch (1978) observed significant mortality of both western spruce budworm, *Choristoneura occidentalis* Freeman, and the blackheaded budworm, *Acleris gloverana* (Walsingham), after applications of insecticidal soaps. To date, no soaps have been tested against the eastern spruce budworm (SBW), *Choristoneura fumiferana* (Clemens), and although they are considered to have low toxicity, little information is available about the effects of these soaps on beneficial species such as parasitoids.

SBW is the most damaging insect pest in Ontario's boreal forest, having caused by 1985, tree mortality on over 14 million hectares (Howse and Applejohn 1986). Current use of organophosphorous and carbamate insecticides for control of this insect in Ontario's forests, over 90% of which are crown forests, has become controversial. Alternative methods of control, one of which is the use of insecticidal soaps, are being investigated. Here, we describe results of experiments which examined the: 1) effects of Fossil Flower's insecticidal soap (IS) (Fossil Flower Natural Bug Controls, Mississauga, Ontario) on SBW feeding, development and survival after ingestion or topical application; 2) impact of IS on one larval parasitoid of the SBW; and 3) effectiveness of IS for control of SBW in the field.

Materials and Methods

Laboratory Studies

SBW were obtained from the Forest Pest Management Institute, Sault Ste. Marie, Ontario. All larvae were exposed during the 2nd instar to parasitism by *Apanteles fumiferanae* Viereck. Parasitoids were obtained from a colony maintained by V. Nealis (Great Lakes Forestry Centre, Sault Ste. Marie, Ontario). Newly moulted larvae were selected by stage according to Retnakaran (1980) and reared individually in 6 ml vials containing 1 g plugs of synthetic diet (Grisdale 1970). The vials containing the larvae were ventilated daily and each diet plug was replaced at 48-h intervals. All studies were done under a regime of 19±1°C, 60-80% RH and 16:8 L:D photoperiod.

The insecticidal activity of Fossil Flower's IS was studied by 1) incorporating a solution contain-

ing 40 ml formulated IS (50% potassium salts of fatty acids) per litre water into the rearing diet; 2) dipping nontreated diet plugs into the solution; and 3) spraying each larva to runoff with the solution and then removing each to nontreated diet plugs. Topical sprays of IS were applied using a mist applicator calibrated to deliver 0.04 ml/cm^2. This rate, although 10 times higher than that recommended for field application against tortricid species, was used in order to magnify subtle effects and observe readily apparent differences. Because we suspected that IS could have insect growth regulator (IGR) activity such as deformed larvae and pupae, the compound was compared with a bonafide moult-inhibiting IGR, Hoechst's (Regina, Saskatchewan) HOE-00522 (4% v/v 1-(3,5-dichloro-2-4-difluorophenyl)-3-(2,6-difluorobenzoyl)-urea) incorporated into the diet as in 1). With the exception of HOE-00522, each of these 4 treatments was applied to 3rd, 4th, 5th and 6th instars so that there were 24 larvae in each stage per treatment. HOE-00522 was not applied to 3rd instar larvae. An additional 24 larvae in each instar were left on artificial diet not treated as controls. This design was replicated twice and analyzed with the aid of 2-way Analysis of Variance (ANOVA) and Duncan's (1955) multiple range test following arcsine transformation of the percentage data.

The effect of the treatments on SBW was determined by comparing overall larval mortality, time to pupation (larval development), time to adult eclosion (pupal development), pupal weight, and adult longevity for each sex. When female SBW were mated to males that underwent the same treatment, the fecundity (number of eggs laid and percentage of eclosed eggs per female) of the females was also measured. Feeding by individual 5th and 6th instars, 3-4 days after ecdysis, was determined from weights of frass pellets produced over a 24-h period and air dried at 19±1°C for 2 weeks. The effect of the treatments on the larval parasitoid *A. fumiferanae* was measured by examining those parasitoids that emerged successfully or were found upon dissection of dead SBW larvae. For emerged parasitoids, survival and developmental time were compared in the 4 treatment groups and the controls using ANOVA.

In trials with foliage, 20 4th instars were allowed to establish on randomly selected buds of newly flushed balsam fir (*Abies balsamea* (L.) Mill.) (1 larva/bud). After 24 h, half of the buds (each with a single larva attached) was sprayed with 0.04 ml/cm^2 IS while the remaining half of the buds were treated with the same volume of water to serve as controls. Each treated bud was placed in an individual 10-mL cup which was then covered with a cardboard lid and inverted. Larval mortality was assessed after 48 h with moribund larvae considered alive. This experiment was replicated twice under the standard laboratory conditions.

The effect of IS on SBW pupae was also studied by spraying 38 pupae 24 h after ecdysis in the manner previously described. Another 38 pupae were sprayed with water, to serve as controls. The treated and nontreated pupae where placed individually into 6 mL holding vials and the percentage survival was assessed daily until ecdysis occurred. This procedure was replicated twice under the standard laboratory conditions.

Feeding preference tests were done by placing 4 diet plugs in a 10-cm-diameter petri dish; 2 of these plugs had IS incorporated (4% v/v) and 2 were not treated. The treated plugs alternated with the control plugs, each 3 cm apart from one another. Two 6th instars were placed in the centre of each of 9 dishes and their distribution on the diet plugs was recorded after 48 h; 3 replicate tests were conducted. Fluorescent bulbs placed ca. 0.5 m above the dishes provided uniform overhead lighting. This experiment and the previous two were analyzed using Ostle and Mensing's (1975) Chi-square test (χ) for proportions with binomial distributions.

Field Studies

Field trials were conducted in a 22-year-old plantation of white spruce (*Picea glauca* (Moench) Voss) near Hearst, Ontario (84°W,50°N). To estimate SBW populations, 38 dominant or codominant trees, evenly divided between white spruce and volunteer balsam fir, were randomly selected. On 14 June IS was applied at the recommended rate of 0.4% v/v (in water) on 7 of the balsam fir and 7 of the white spruce. Because no change in SBW populations was evident in the week following the first application, IS was reapplied 11 days later at the same concentration to 8 of these 14 trees and to an additional 3 balsam fir and 3 white spruce. Again, as no effects were apparent, 7 days after the second treatment, IS was applied at 2% v/v to an additional 5 balsam fir and 5 white spruce.

Five treatment regimes were thus established. These included: 1) 6 trees treated once with 0.4% IS on 14 June; 2) 6 trees treated once with 0.4% IS on 25 June; 3) 8 trees treated twice with 0.4% IS on 14 and 25 June; 4) 10 trees treated once with 2% IS on 2 July; and 5) 8 trees left as controls. The controls included 4 nontreated trees and 4 trees sprayed with water alone on 2 July. The initial spray date was selected to coincide with that developmental stage of the SBW when operational applications of aminocarb or *Bacillus thuringiensis* Berliner (*B.t.*) were conducted by provincial agencies.

IS was applied until runoff (ca. 10 min) on each date between 19:00 and 22:00 h, from a backpack mistblower calibrated to release at 3.5 L/min. Rates of application (mL/m^3 foliage) were calculated on the basis of applicator flow rate, individual tree volume and the time required. Tree volume was derived by measuring the height and the diameter of the crown at the base of each tree and then assuming that the tree had a perfect conical shape to estimate the volume of a cone. Weather conditions were recorded daily 15 km from the site.

Populations of SBW were estimated by counting insects on one branch sample taken from the upper mid-crown of each tree, 1-3 days before and after each spray. To assess SBW development, the number of larvae and their instars were recorded for all sample dates (McGugan 1954). An instar index was used to summarize SBW development. This value was obtained by multiplying the frequency of SBW collected at each stage by the stage number (i.e., 1-6, 1st to 6th instars; 7, pupae; 8, adults), summing these values and then dividing the sum by the total number of SBW collected. Defoliation was estimated using the Dorais-Hardy method of assessment on a single branch taken from the midcrown of each sample tree on 10 July (Sanders 1980). Population reductions resulting from the application of IS were computed by means of a modified Abbott's formula (Retnakaran 1982) based on pre- and post-spray population densities of SBW larvae. Pupae collected from the branches were held in ventilated vials under field conditions until adult eclosion or until pupal parasitoids emerged. The parasitoids were identified as described by Tilles and Woodley (1984).

Pre- and post-spray densities of SBW found on trees of each species and the resulting defoliation of those trees were compared for each application date by means of 2-tail Student's t-test. The total percentage of emergence, pupal parasitism and SBW larvae in each instar over the season were analyzed following arcsine transformations with ANOVA and Duncan's (1955) multiple range test.

Results

Laboratory Studies

Treatments with 4% Fossil Flower's IS (50% potassium salts of fatty acids) and with the IGR, HOE-005522, were not selectively toxic to either male or female SBW. Both sexes were represented in approximately equal numbers, therefore, the results were pooled for further analysis of each treatment group. In each case, when applied to 3rd, 4th and 5th instars, all treatments significantly increased larval mortality over that in the controls (Table I). This was particularly apparent with HOE-00522 which provided 100% mortality. During the 6th instar, only IS applied topically and HOE-00522 resulted in significant larval mortality. Several treatments reduced the dry weight of frass pellets over that of the controls.

The pupal stadium was extended slightly following treatment with IS but the delay was significant only when IS was applied topically to 6th instars (Table I). To a limited extent, pupal weights were reduced when SBW larvae ingested IS with the diet and when IS was applied topically to the diet during the 5th instar or to the larvae during 5th and 6th instars. Where female larvae completed development to the adult stage, treatment with IS generally reduced fecundity although there was considerable variation observed. These differences were significant (P=0.05) where IS was administered topically on the 4th and 6th instars and through ingestion by 3rd and 5th instars. In general, adult longevity was not affected by the IS treatments, while the percentage of eggs hatching was reduced significantly following most of the treatments.

IS incorporated in the diet or applied topically and HOE-00522 reduced successful emergence of *A. fumiferanae* from its SBW host (Table II). Parasitoid development in the host was extended significantly by HOE-00522 but not by either of the IS treatments. In most cases, dissections of dead SBW larvae showed that the parasitoid failed to emerge because the host died before parasitoid development was complete.

TABLE I. Effects of an insecticidal soap and chitin inhibitor growth regulator on spruce budworm when applied to 3rd, 4th, 5th and 6th instars.

Instar	Treatment	Larval mortality (%)	Larval development (days)	Frass dry weight (mg)	Pupal development (days)	Pupal weight (mg)	Adult longevity (days)	No. of eggs laid/ femal[eA]	Eclosed eggs (%)
3rd	Control	45.8a[B]	22.1a	-	7.5a	91a	15.5a	58.0a	53.4a
	IS-diet[C]	75.0b	24.0a	-	12.0a	41a	5.0b	0b	-
	IS-dip	100.0b	-	-	-	-	-	-	-
	IS-topical	83.3b	21.0a	-	9.7a	95a	15.0a	27.0a	1.0b
4th	Control	29.2a	16.8a	-	8.1a	89a	15.3a	16.4a	82.5a
	IS-diet	62.5b	18.2a	-	9.3a	43a	13.6a	10.0a	16.7b
	IS-dip	50.0b	16.4a	-	8.3a	78a	15.2a	11.5a	0b
	IS-topical	62.5b	18.8a	-	8.7a	77a	15.5a	9.7b	9.7b
	HOE-00522	100.0b	-	-	-	-	-	-	-
5th	Control	8.3a	14.8a	0.7a	8.1a	97a	16.4a	103.0a	38.6a
	IS-diet	45.8b	13.8a	0.1b	8.1a	46c	13.1b	7.0b	0b
	IS-dip	62.5b	14.4a	-	9.0a	72b	13.7ab	23.0ab	0b
	IS-topical	62.5b	12.6a	0.3b	8.8a	76b	13.9ab	47.0a	1.0b
	HOE-00522	100.0b	-	-	-	-	-	-	-
6th	Control	4.2a	6.3a	3.4a	8.0a	87a	15.8a	139.2a	65.7a
	IS-diet	4.2a	6.2a	1.1b	8.8ab	66b	15.8a	82.2ab	57.3ab
	IS-dip	4.2a	6.5a	-	8.6ab	99a	16.3a	82.3a	43.5b
	IS-topical	54.2b	5.6a	2.3ab	9.6b	80ab	17.0a	10.0b	15.9c
	MOE-00522	95.8b	6.0a	2.1ab	-	-	-	-	-

[A]n = mean of 14 females per treatment (range 8-22) based on matings producing fertile eggs.

[B]Instars analyzed separately. Means followed by the same letter within each instar and category are not significantly different (P 0.05; Duncan's (1955) multiple range test). Represents pooled values for both replicates; n = 48 larvae per treatment.

[C]IS, Fossil Flower's insecticidal soap (4% v/v) 50% potassium salts; IS-diet, IS incorporated into synthetic diet; IS-dip, synthetic diet dipped in 4% IS; IS-topical, IS sprayed on spruce budworm larvae fed synthetic diet not treated; HOE-00522, Hoechst's chitin-inhibitor growth regulator (4% v/v) incorporated in synthetic diet.

TABLE II. Effect of an insecticidal soap and a chitin inhibitor growth regulator on *Apanteles fumiferanae* when applied to parasitized 4th instar spruce budworm.

Treatment	n	Parasitoid survival (%)[A]	Parasitoid development (days)
Control	18	89a	8.6a
IS-diet[B]	19	37c	8.6a
IS-topical	19	53b	10.0a
HOE-00522	9	33c	18.0b

[A]Means followed by the same letter within each category are not significantly different (P >0.05; Duncan's (1955) multiple range test).

[B]See footnote B, Table I.

When 4th instars were allowed to establish themselves on tips of balsam fir and were sprayed with IS, larval mortality increased significantly over that of the controls; 55.0% of the treated larvae died in comparison with 0% of the control larvae (χ^2_s=9.05; df=1; P=0.01).

Of those pupae sprayed directly with 4% IS, only 29.8% emerged successfully; 86.4% of the pupae sprayed with water alone emerged, representing a 65% reduction in adult emergence (χ^2_s=9.82; df=1; P=0.01). There were no morphological or juvenile hormone effects noted in the pupae or adults of these treated insects as might be expected if the IS had acted as an IGR.

Of the 54, 6th instars allowed to choose nontreated diet versus 4% IS incorporated diet, 42 larvae were found feeding on the IS diet after 48 h while only 12 larvae were established on the nontreated diet (different at χ^2_s=14.36; df=1; P=0.01).

Field Studies

Weather conditions at the time of all 3 applications were similar; daily temperatures and relative humidities averaged 15.9 ± 1.5°C and 56 ± 12%, respectively. No rainfall was recorded during, or for 2 days following, any of the applications. At the time of the first application on 14 June, the greatest proportion of SBW larvae were 4th or 5th instars (index = 4.7). By 25 June, the larval index had progressed to 5.7 and by 2 July, most larvae had reached the 6th instar; 5 to 12% had already pupated (index = 6.0).

SBW populations were generally not affected by the soap applications (Table III). Natural mortality of SBW larvae was similar in the 2 control groups, ranging from 14.3 to 32.2%, while larval mortality on treated trees ranged from 0 to 37.4% and was not significantly different from that of the controls. Only the second, single application of IS on 25 June reduced SBW populations by 31% on balsam fir and by 30% on white spruce. The third spray on 2 July also significantly reduced larval populations but only on balsam fir. Despite fewer larvae, there was no evidence that the IS treatments provided significant foliage protection. Defoliation was generally higher on the treated trees than on the control trees (Table III).

In the field, the applications of IS produced extremely variable responses in SBW development, adult emergence, and pupal parasitism (Table IV). On balsam fir, more larvae were found on the treated trees than on the nontreated trees, suggesting a slight delay in larval development as a result of the IS application. On white spruce, those trees sprayed on 2 July had lower adult emergence and greater pupal parasitism by *Apecthis ontario* (Cress.), *Meterous tachynotus* Vier. and *Winthemia amoena* (Mg.) than the control trees or those trees sprayed earlier (i.e., on 14 June). These two effects of IS application on SBW feeding in white spruce are probably linked, in that greater pupal parasitism will always lead to lower adult emergence. Both effects should be considered sublethal, however, as they contribute, either separately or in combination, to a general reduction in SBW populations.

Table III. Experimental field applications of insecticidal soap on spruce budworm populations near Hearst, Ontario, 1985.

Treatment Date	Treatment (mL IS/m^3 foliage)[A]	No. of Sample Trees	Spruce budworm density[B] Pre-spray Bf	Pre-spray Sw	Post-spray Bf	Post-spray Sw	% Population reduction[C] Bf	% Population reduction[C] Sw	% Defoliation Bf	% Defoliation Sw
14 June	2.5	14	14(1)	43(9)	25(6)	39(1)			91(5)	96(1)
	control	14	11(2)	50(7)	9(2)	42(4)	0	0	70(5)	74(5)
25 June	5.8	6	16(3)	31(4)	8(4)	14(6)			87(3)	77(9)
	control	8	11(2)	31(7)	8(1)	20(3)	31[D]	30[D]	70(5)	74(5)
14 June & 25 June	2.5 & 4.2	6	15(2)	32(5)	16(2)	15(2)			55(15)	85(6)
	control	8	9(3)	46(6)	8(1)	20(3)	0	0	70(5)	74(9)
2 July	211.2	10	20(3)	25(3)	16(5)	39(6)			78(5)	80(9)
	control i)[E]	4	4(2)	17(9)	6(5)	20(10)	47[D]	0	63(3)	80(5)
	ii)	4	11(0)	31(18)	7(5)	20(2)	0	0	70(5)	68(8)

[A] IS, active ingredient of Fossil Flower's insecticidal soap; Bf, balsam fir; Sw, white spruce; m^3 foliage based on individual tree volumes.

[B] Mean number of spruce budworm per 45-cm branch tip, with standard errors indicated in parentheses; two significant digits.

[C] $\%\ \text{population reduction} = 1 - \frac{\text{post-spray density in treatment}}{\text{pre-spray density in treatment}} \times \frac{\text{pre-spray density in control}}{\text{post-spray density in control}} \times 100$ from Retnakaran (1982).

[D] Significant reduction at the $P = 0.05$ level (Student's t-test).

[E] Control trees: i) sprayed 2 July with 0.8 L water/m^3 foliage; ii) not sprayed.

TABLE IV. Proportion of spruce budworm in the larval stage, adult emergence and pupal parasitism by species of host tree following applications of insecticidal soap near Hearst, Ontario, 1985.

Host tree	Treatment Date(s)	n[A]	SBW larvae (% of total)	Adult emergence (%)	Pupal parasitism (%)
Balsam fir	14 June	15	85ab[B]	78ab	2a
	25 June	15	90a	81a	7a
	14,25 June	20	89a	66a	12a
	2 July	15	-	58a	15a
	Control	28	78b	55a	14a
White spruce	14 June	15	78a	64a	4a
	25 June	15	85a	53ab	33b
	14,25 June	20	88a	46ab	9a
	2 July	15	-	38b	13ab
	Control	28	75a	62a	4a

[A]n, number of branch samples

[B]Host trees analyzed independently; means followed by the same letter within each column are not significantly different ($P > 0.05$; Duncan's (1955) multiple range test).

Two modes of action for insecticidal soaps have been proposed: 1) as IGRs, by interfering with cellular metabolism and, thus, growth hormones during metamorphosis (McFarlane and Henneberry 1965; Andrews and Miskus 1972; Puritch 1975, 1978), or 2) as contact insecticides, by blocking spiracles and interfering with respiration (Abbasi *et al.* 1984). In the present study, although 6th instars preferred the diet incorporated with IS to that not treated, the dry weight of their frass was reduced, as was the case with the IGR. Although this reduction in feeding may result from other toxicological effects which weaken larvae, a failure to feed or a change in feeding is one the effects associated with chitin-inhibiting IGRs (Retnakaran *et al.* 1985). Further physiological studies on the mode of action of IS would be of interest, as they would better examine the possible growth regulating effects of this compound.

As with most insecticidal compounds, soaps may produce either acute mortality or sublethal debilitation. Both effects were evident in the present study. These included: larval and pupal mortality, slightly extended pupal development for 6th instars, reduced pupal weight and reduced female fecundity. The sublethal effects were more apparent when 5th and 6th instars were treated than when 3rd or 4th instars were treated. As suggested by the feeding study conducted with the 6th instars, these effects might be associated with changes in feeding or assimilation. The sublethal effects of insecticidal soaps or IGRs have been examined in a few studies (Retnakaran *et al.* 1985; Alford and Holmes 1986). Puritch (1978) observed IGR-like effects on pupae of *Tenebrio molitor* L. following treatment with fatty acid salts and Madore *et al.* (1983) found that mating success and egg production of SBW were reduced after larval treatment with a chitin-inhibiting IGR.

Parasitism of SBW appeared to be inconsistently affected by the application of IS. In the field, variable changes in parasitism were observed for IS applied during SBW pupation as opposed to treatments of earlier larval instars. This suggests that pupae treated with IS may be more available for parasitoid oviposition than nontreated pupae. In the laboratory, the larval parasitoid *A. fumiferanae* developed normally despite exposure of its host to IS. Parasitoid mortality occurred only when SBW was also killed (before the 5th instar). *B.t.* produced a similar, indirect effect on a larval parasitoid of the tobacco budworm, *Heliothis virescens* (Fabr.) (Thomas and Watson 1986) while only a direct effect of diflubenzuron on a larval parasitoid of the gypsy moth, *Lymantria dispar* (L.), was reported by Grannett and Weseloh (1975). Further studies are needed to clarify both direct and indirect effects of insecticidal soaps on beneficial insects in order to make recommendations for their use in the field.

Field applications of IS at the current label rate may lower SBW populations to a limited extent through their sublethal effects on the larvae. In the short term, however, they will not cause sufficient larval mortality to suppress SBW populations nor will they prevent defoliation. Although higher rates (4%) such as those used in the laboratory might have a greater impact, in the field, both the behaviour of young SBW larvae and the presence of tree foliage will likely reduce the chances of larvae actually contacting the spray. In Ontario, because of the high costs associated with treating extensive forested areas, IS would need to be applied aerially for the control of SBW. Aerial applications would provide considerably less coverage to individual trees than would handheld mistblowers. At the present time, therefore, neither type of IS application can compete successfully with the standard control methods of provincial agencies (i.e., *B.t.* and broad-spectrum insecticides) and IS is not currently recommended for SBW suppression.

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