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bi-monthly research notes

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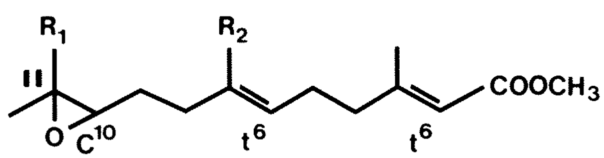
ENTOMOLOGY

Occurrence of More Than One Juvenile Hormone in *Locusta migratoria* L.—Structure elucidation of juvenile hormones (JH) in insects has revealed the existence of three different types of hormones: Methyl cis-10, 11-epoxy-3, 11-dimethyl-7-ethyl-trans, trans-2, 6-tridecadienoate (C_{18}-JH or Methyl 12, 14-dihomojuvenate or JH-I); Methyl cis-10, 11-epoxy-3, 7, 11-trimethyl-trans, trans-2, 6-tridecadienoate (C_{17}-JH or Methyl 12-homojuvenate or JH-II); and Methyl 10, 11-epoxy-3,7,11-trimethyl-trans, trans-2, 6-dodecadienoate (C_{16}-JH or Methyl juvenate or JH-III) (Table 1) (Slama *et al*, Insect hormones and bioanalogues. Springer-Verlag, New York 1974).

TABLE 1
Juvenile hormones in insects



$R_1 = R_2 = CH_2CH_3$ HJ-I
$R_1 = CH_2CH_3$; $R_2 = CH_3$ HJ-II
$R_1 = R_2 = CH_3$ HJ-III

It was reported that members of the order Orthoptera had only JH-III (Muller *et al*, Life Sci. 15:915-921, 1974; Trautmann *et al*, Z. Naturforsch. 29c:757-759, 1974). Recently, however, it has been shown by Lanzrein *et al* that in the cockroach, *Nauphoeta cinerea* all three hormones occur at different developmental stages (Life Sci. 16:1271-1284, 1975). According to these authors, the predominant component JH-III masked the JH-I and JH-II present and only the very sensitive techniques employed by them revealed existence of JH-I and JH-II. They suggest that JH-I and II may be involved in juvenilizing functions with JH-III having a gonadotropic function.

We have searched for all three hormones in another Orthopteran, *Locustra migratoria.* Locusts were reared in the gregarious phase under crowded conditions on wheat seedlings at a temperature of 30°C during day and 25°C at night, 60% R.H., and a photoperiod of 12 h light and 12 h darkness. *Corpora allata* (52 pairs) were dissected from 1-4 day old adults and extracted in benzene, concentrated to 500 μl, and stored at −20°C prior to use. The benzene extract was chromatographed on Merck F-254, 0.25 mm thick pre-coated silica gel plates using a solvent system of n-hexane, chloroform, and ethyl acetate (7:7:1 v/v/v) (Trautmann, personal communication). The plate was developed twice (approx. 50 min/run) and using tritium labelled JH-I and III as markers, the areas were located using a Berthold scanner. Under these conditions JH-I and II run together ahead of JH-III with Rf values of 0.84 and 0.79 respectively. The JH I/II and III areas on the plate were scraped separately and extracted in ether and 10 μl of peanut oil was added to each extract. After evaporating the ether under N_2, 10 mg of paraffin (MP 58°C) was added to each fraction and the JH activity was bioassayed using the *Galleria* wax test (Wilde *et al*, Proc. Kon. Ned. Akad. Wetensch. C71:321-326, 1968.)

The Rf 0.84 fraction (JH I/II) showed an activity of 20.00 *Galleria* units (G.U.) whereas the Rf 0.79 fraction (JH-III) showed an activity of 28.32 G.U. Since the 0.79 fraction is more active this precludes the possibility of tailing of the 0.84 fraction. Moreover, it is known that JH-I and II are more active than JH-III in the *Galleria* assay (Lanzrein *et al* Life Sci. 16:1271-1284, 1975). The results indicate that in addition to JH-III, JH-I or II or both is present in *Locusta migratoria*. (This work was done at the University of Louis Pasteur Strasbourg, France in 1975 with Prof. P. Joly when the author was on a Professional Development Assignment.—Arthur Retnakaran, Insect Pathology Research Institute, Sault Ste. Marie, Ont.

Aerial Application of Carbaryl for Control of Spruce Budworm in High-Value Forests.—Recent infestations of the spruce budworm [*Choristoneura fumiferona* (Clem.)] in intensively managed spruce [*Picea* spp.] and balsam fir [*Abies balsamea* L.] plantations, woodlots, parks, or recreational areas have necessitated increased use of chemical control to prevent monetary and aesthetic losses. Applications with ground spray-equipment are frequently impractical due to tree height, stand size, and limited access, leaving treatment by aircraft as the only alternative. Where large operational aerial spray programs are undertaken, as in Quebec and New Brunswick, small high-value forested areas may be incorporated for treatment. However, when these small areas are widely scattered treatment by small agricultural-spray aircraft is more appropriate.

The choice of insecticides for operations of this sort has been very limited by government restrictions and to some extent by product availability. All pest control products used in forest management in Canada must be registered as outlined in Trade Memorandum T-104 of the Control Products Section, Plant Products Division, Canada Department of Agriculture. (C.F.S. Rep. CC-X-19, 1975 lists the seven insecticides registered for control of spruce budworm by aerial application). However, for small-scale spray operations, product availability, specialized mixing equipment, insecticide dosage limitations, and unique environmental considerations often limit pesticide choice. Therefore a wider selection of insecticides and formulations to control the budworm in small high-value forested areas are needed.

In 1972, the authors found that carbaryl (1-naphthyl methyl-carbamate) applied in the commercial formulations Sevin 80S® and Sevin 50WP® by mistblower at 0.56 kg. a.i./ha (8 oz a.i./acre) and 1.68 kg a.i./ha (24 oz a.i./acre), provided excellent protection of white spruce [*P. glauca* (Moench) Voss] (C.F.S. Inf. Rep. CC-X-21, 1972). Subsequent studies conducted in Manitoba by Hildahl and DeBoo (Man. Ent. 7:6-14, 1973) showed that low-volume aerial applications of carbaryl in the formulation Sevin-4-Oil® at 1.12 kg a.i./ha (16 oz a.i./acre in 2.9 *l* total volume per hectare (40 U.S. oz/acre) by a boom-and-nozzle equipped Piper Pawnee effectively reduced budworm population density.

In 1974, a follow-up study was undertaken to determine the efficacy of carbaryl applied at 0.56 kg a.i./ha (8 oz a.i./acre). Two white spruce plantation blocks totalling 13.8 ha (34 acres) located near Shawville, Quebec, were selected for treatment. Two smaller blocks nearby, comprising approximately 4 ha (10 acres) in total area, served as untreated check areas. The trees were healthy with full crowns and averaged 8 m (25 ft) in height. Previous damage to these stands was minimal despite 3 years successive defoliation by light to moderate populations of the budworm (i.e. <20 larvae/45 cm (18-inch) branch tip/year).

The Chemical Control Research Institute's Cessna 185 Skywagon equipped with four AU3000 Micronair rotary atomizers calibrated to emit 1.45 *l*/ha (20 fl oz/acre) at a calculated effective swath width of 15 m (50 ft) was used.

Figure 1. Aerial application of carbaryl by Cessna 185 equipped with Sorensen spraying system and Microair rotary atomizers.

A spray rate of 0.55 kg a.i./ha (7.8 oz a.i./acre) was obtained by diluting 473 ml (16 fl oz) of Sevin-4-Oil with 118 ml (4 fl oz) of No. 2 fuel oil. Spray treatments were applied from 0645 to 0710 h, June 3, when conditions were favorable: crosswind speed 0-3.2 km/h (0-2 mph), temperature 12°C, relative humidity 94%, and budworm larvae 35% 3rd and 57% 4th instar. The aircraft was guided across each spray block by a flagman with red and white helium balloons tethered to aluminum pruning pole sections. Spray deposit patterns were obtained by placing 10 cm x 10 cm (4 in. x 4 in.) Kromekote® cards at regular intervals along transects oriented at right angles to the flight path in the middle of each block. The cards were later processed for estimates of droplet size (Volume Median Diameter) and density (number per square centimeter) using the National Aeronautical Establishment's Flying Spot Scanner-Analyser developed by Slack (Division of Mechanical Engineering, National Aeronautical Establishment, Quar. Bul. No. 1972 (73)). Estimates of spruce budworm population density over time in both treated and untreated blocks were based on counts from 45 cm branch tips collected twice before treatment and three times afterwards. Defoliation was appraised in September by binocular estimation of overall crown condition of representative trees.

Spray deposit analysis indicated an average of 0.56 *l* (approximately 40% of the emitted spray volume) reached ground level as uniformly fine droplets averaging 100 um VMD. Droplet density averaged 18 drops/cm^2, range 2-41 drops/cm^2. The treatment reduced the larval population to 61%. Defoliation was most evident in the lower portions of tree crowns, indicating significant interception of droplets by foliage higher up on the windward side of the trees. It was concluded that for high-value stands with budworm population densities exceeding 20 larvae per 45 cm branch tip either two applications as described or a single application at up to 1.12 kg A.I./ha (1 lb. a.i./acre) at an emitted volume to 9.27 1/ha (1 U.S. gal/acre may be necessary to protect foliage throughout the tree crown.

In 1975 carbaryl was registered in Canada on a temporary basis for control of the spruce budworm, and was used by Parks Canada to treat severe infestations in intensive-use areas

TABLE 1

Results of experimental applications of carbaryl at 0.56 kg a.i./ha

Treatment	Larval Population Density (Avg No./45 cm branch tip) Pre-spray (−1 day)	Post-spray (+9 day)	Corrected % Population Reduction[1]	Tree Crown Defoliation (%) Top	Mid	Lower
Carbaryl	21	5	61	6	11	24
Untreated Check	23	14	—	15	34	59

[1] Corrected by Abbott's Formula (J. Econ. Ent., 1925).

of La Mauricie and Forillon National Parks (Foisy *et al.*, Special Report, Parks Canada Quebec Region, 1975). Sevin-4-Oil formulation was used to spray approximately 202,000 ha (500,000 acres) of pulpwood forest during 1975 at the rate of 1.12 kg a.i./acre (16 oz. a.i./acre) (J. B. Dimond, Dept. Entomology, Univ. of Maine, Orono, personal communication). Preliminary information received by the authors indicates that carbaryl will be used in Maine to treatment of several million acres of infested forest during 1976.

Carbaryl is a broad-spectrum insecticide which is particularly effective against hymenopterous insects. Good judgement and planning are prerequisites to its use where pollinators (including honey bees) and beneficial insect parasites may be abundant.—L. M. Campbell and R. F. DeBoo, Chemical Control Research Institute, Ottawa, Ont.

Use of Radio-controlled Model Aircraft for ULV Insecticide Application in Christmas Tree Stands.—A promising new method of crop protection in small-scale operations has been tested in New Brunswick. A radio-controlled model aircraft was used successfully to spray a 2.4-ha Christmas tree farm infested with spruce budworm [*Choristoneura fumiferana* Clem.]. Such aircraft have been used experimentally to study ultra-low-volume (ULV) spray patterns, but they have never been used commercially (Murphey, Ninth Northeast Aerial Appl. Conf., pp 11-15, 1973). The purpose of this study was to test the commercial practicability of these machines for any small-area crop, usually under 20 ha, for which conventional methods of control are difficult or impractical because of rough or inaccessible terrain.

Insect control in such areas is most often carried out with backpack sprayers or conventional spray aircraft. These are efficient and practical but have some drawbacks. Mist blowers are slow, (requiring about 0.5 man-days per ha), unpleasant and tiresome to operate, and continually expose the operator to insecticide. The use of convential spray aircraft is expensive if the areas to be treated are small; large-scale operations, which can reduce costs, often depend on the cooperation of several land owners.

Model aircraft are small and portable and, in a few minutes, can deliver potent dosages of concentrated insecticide at low altitudes with relatively little risk to the operator. The major disadvantages are their fragility and the special skills required to operate them. Although reliable radio-controlled model aircraft have been in existence for less than 10 years, they are becoming increasingly popular. Flying clubs are being organized in most large centers, and it is from these clubs that future spray operators could be recruited if a dependable spray plane were developed.

For this trial a senior Telemaster model aircraft manufactured by Alexander Engel Co., West Germany (Fig. 1) with a 240-cm wing span, a 1.2-hp engine (at 12,000 rpm), and a capability of lifting a 4.5-kg load was fitted with a nozzle from an "ULVA" hand-sprayer (Micron Sprayers Ltd., Bromyard, England). The sprayer was powered by a 7-watt electric motor that drove two stacked corrugated plastic atomizer disks with serrated edges at a speed of 7,000 rpm (Fig. 2); the motor ran continuously when the aircraft was in flight. Fenitrothion

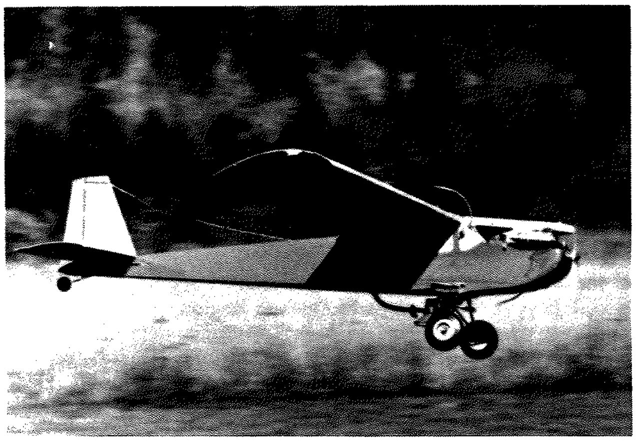


Figure 1. Model aircraft in flight.

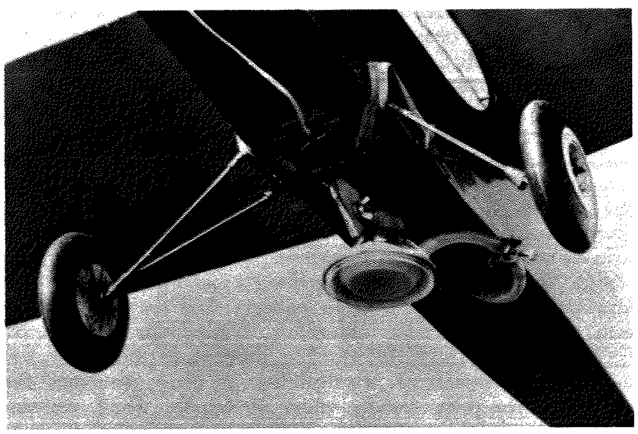


Figure 2. Location of spray-nozzle attachment.

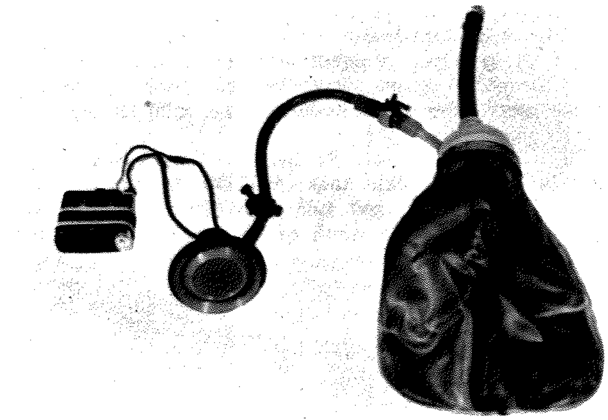


Figure 3. Spray assembly showing, left to right, the battery pack, "ULVA" spray head, hose-clamp valve, and collapsible spray bag with filling tube and attachment for aircraft exhaust.

E.C. (74% a.i.) in a 50% emulsion with water was emitted through a valve that could be opened or closed from the ground by a radio-controlled servo (Fig. 3). The spray tank was a collapsible 2.3-liter plastic bag fitted inside the fuselage. Part of the engine exhaust was piped into the bag to supplement normal air pressure in forcing out all the insecticide. The rate of flow was regulated at 63 ml/min, while the aircraft engine was running, by a hose clamp above the spray nozzle.

The area sprayed was an exprimental Christmas tree farm at St. Laurent, in northeastern New Brunswick, operated jointly by the Canadian Forestry Service, the Northeast Christmas Tree Producers Council, the Community Improvement Corporation, and the New Brunswick Department of Natural Resources. The farm was bounded by trees 12 m high, and telephone and power lines, and was beside a main highway. The choice of site was deliberate to assure a rigorous test of the aircraft. Landing had to be accomplished by bringing

TABLE 1

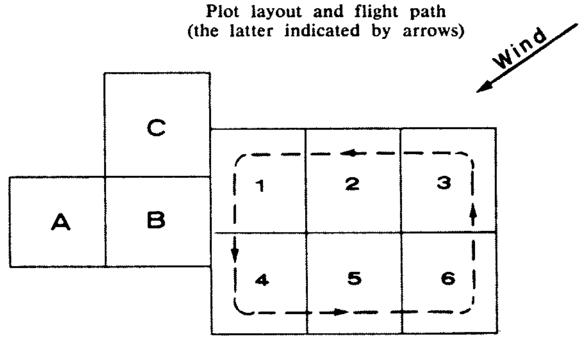
Pre- and postspray spruce budworm population indices and diagram of spray pattern.

Block	Prespray: Total larvae on 30 shoots per block	Postspray: % population drop
Control		
A	8	0
		(0)
Borderline		
B	21	67
C	8	37
Average		(57)
Sprayed		
1	24	83
2	23	83
3	15	73
4	15	73
5	11	73
6	21	100
Average		(83)

Postspray sampling of trees near spray assessment cards

Spray drops per cm²*	No. of buds	Total larvae	% dead larvae
.59	146	33	70
.70	196	45	67
.41	179	48	45
.40	73	22	65

Plot layout and flight path (the latter indicated by arrows)



* Per 10 cards at 10-ft (3-m) intervals between blocks 1 and 2; and 4 and 5.

the aircraft down over these obstacles onto a rough 60-m landing area. The farm included several 61-m square blocks of culivated wild balsam fir in the early stages of Christmas tree production. The area was understocked with trees 2 m high at about 2,000 stems per hectare, which had been only slightly defoliated in 1974. Six blocks (Table 1) were selected for treatment and three blocks (two bordering on the spray area) for controls. At the time of spraying the spruce budworm larvae were at the peak of the fourth instar.

The aircraft was flown in an oblong flight pattern (Table 1) above the six blocks at an altitude of 15 m. The test consisted in making several circuits lasting a total of 6 min, just at dusk when wind velocity was less than 1.6 km/hr. A total of 384 ml (a.i.) of fenitrothion plus rhodamine W.T. fluorescent dye was evenly distributed over the 2.4 ha. Forty spray detection cards were distributed at 3-m intervals along a transect through the center of the area, roughly at right angles to the flight path, between blocks 1 and 2, and 4 and 5 (Table 1). Droplets of spray were detected, with a black light, on all cards to within 90 m from the windward edge of the spray area. Stain size averaged 400 microns in diameter and density averaged 0.52/cm^2.

The area was sampled for budworms immediately before and 1 week after the spray operation. Larvae were counted on one randomly selected shoot at eye level from each of 10 trees along three lines traversing each block. The decrease in budworm population, as shown by reduced counts and dead larvae, averaged 83% in the sprayed blocks and 57% in the adjacent blocks. No decrease was detected in the one true control block (Table 1) and no dead larvae were found. In addition, 594 shoots showing severe budworm damage were collected from trees along the line of the spray cards. The shoots were divided into four subsamples, each representing a 30-m distance along the lone. Larval mortality in the subsamples ranged between 45 and 70% (Table 1).

Two modifications that would improve the aircraft's performance were indicated by the tests. The aircraft should be equipped with landing flaps to allow a steeper and slower descent, and with plastic skids instead of wheels to permit landings on rough ground. The existing spray equipment appears satisfactory, but almost any lightweight ULV system could be used. Ideally the insecticide should be highly concentrated because flight time is short, and of low viscosity to insure small droplets.

The results are surprising because of the low density and size of droplets detected on the spray cards. However, the final indication of success came at the end of the growing period. The sprayed blocks were virtually free of budworm damage, while in the surrounding areas damage was severe.— D. C. Embree, C. M. B. Dobson, E. G. Kettela, Maritimes Forest Research Centre, Fredericton, N.B.

A New Insect Trap for Use with Lepidopteran Sex Pheromones.—The use of sex-pheromone baited traps in the monitoring of management of populations of lepidopteran agricultural or forest pests, is affected by several factors including availability of the synthetic pheromone, type of pheromone dispenser, release rate and trap design.

Among the various components of trap design which led to the development of a new trap, the Astrotrap (Canadian Patent, 964,059 and U.S. Patent, 3,863,384), were (a) ease of handling (b) area of trapping surface and (c) durability. Three types of traps are discussed to illustrate these points. **Type 1** — A flat plywood board (60x60x20 cm), with a small hole drilled through the center for the lure dispenser. To both sides of the board an adhesive such as Stickhum™ or Tanglefoot™ is applied and the trap may either be suspended from a tree limb or be affixed to a stake. This type, although having a large trapping surface, is not easy to handle and presents problems

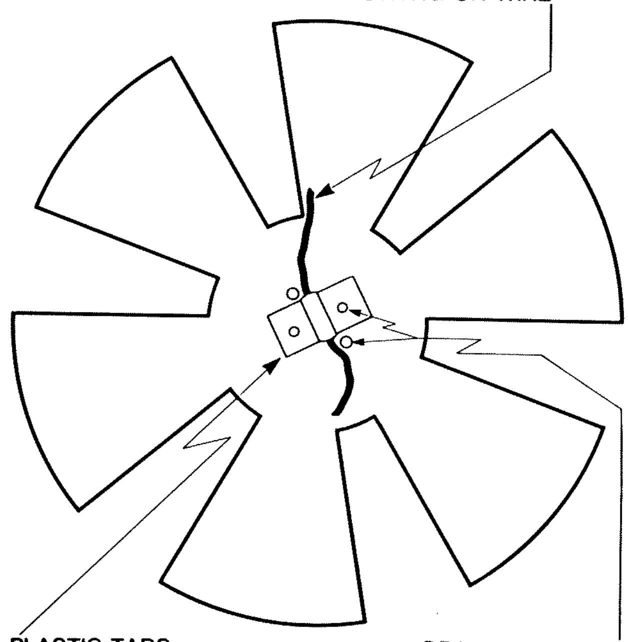


Figure 1. Sketch of panel from which the insect trap was constructed.

in storage, transportation, and assembly. It will withstand adverse weather conditions, but windborne debris, such as leaves, needles, bark and even an occasional small bird, stick to the surfaces thereby considerably reducing the trapping effectiveness. **Type 2** — Wing trap, made of plasticized cardboard, collapsible, with usually three adhesive coated vanes. Type 2 is superior to Type 1 in handling because large numbers are already transported and assembly is easy. The trapping area is large, but unprotected from rain, wind and debris. **Type 3** — Ice cream carton or food container (Sealright™) made of waxed carboard, cylindrical in shape with the adhesive applied to the inside of the cylinder. The trapping surface of type 3 is protected from the elements, and despite being usually smaller in area than types 1 and 2, is more effective. The trap is durable in that it holds its shape in inclement weather. Although the cartons are light, they are not collapsible and ease of handling is inferior to type 2. The commercially available Sectar 1™ or Pherocon-2™ traps are modified type 3 traps which are collapsible. One hundred Pherocon-2 traps measure approximately 30x15x9.5 cm.

Improvements in trap design that lead to the patenting of the Astrotrop were increased access to the trap by flying insects while still maintaining a large trapping surface, increased service life by making the trap reversible because the majority of moths are trapped on the lower half of type 3 traps. These improvements were accomplished by making the trap from flexible sheets of plastic laminated paper. The traps were constructed of upper and lower, 25.4 cm dia, panels coated on one side with adhesive, and joined with plastic tape along the whole width of the edge of six arms (Figure 1). Both panels have drainage holes, plastic tabs for opening the trap and wire or string loops for suspending the traps.

The Astrotraps were evaluated against Sectar 1 and Sectar XC-26™ traps obtained from 3 M Co., St. Paul, Minn. Dr. T. I. Bradshaw of 3 M Co., also supplied several of the Astrotraps used. The synthetic pheromone, (E)-11-tetradecenal (Weatherston, *et al.*, Can. Ent. 103:1741-1747, 1971) was kindly supplied by Ayerst Research Laboratories, Montreal and dispensed in 1, 5 and 10 mg amounts into polyethylene stoppers (Weatherston, et al. *loc cit.*). The trapping experiments were carried out in 1972 and 1973 at Parkinson Township in Algoma District.

In 1972 Astrotraps (3 replicates) and Sectar 1 (2 replicates) were randomly placed at a height of 1.5-2.0 m in a mixed timber stand. The experiment started on July 4 and concluded on July 20, the peak flight period, with the traps inspected on July 6, 8, 12, 16 and 20. On July 8 and 16 all traps were reversed; while on July 12 all traps, but not the lures were replaced. The virgin female spruce budworm used as baited were replaced every 4 days with newly emerged insects. The 1973 tests involved Astrotraps (3 replicates), Sectar 1 (3 replicates) and Sectar XC-26 (3 replicates) charged with 5 mg amounts of (E)-11-tetradecenal and randomly placed at a height of 1.5-2.0 m in a mixed stand on July 17, and inspected every 2 days until the test concluded on July 31. On July 21 and 29 all traps were reversed; while the traps but not the lures were changed on July 25. Virgin females used as bait were changed every 4 days. The 1972 tests were performed in a high budworm population while the 1973 tests were carried out in a low population by trapping only towards the end of the flight period.

TABLE 1

1972 trapping results. Total numbers of male spruce budworm caught with various amounts of (E)-11-tetradecenal, virgin female and empty control traps.

Day	(E)-11-tetradecenal 10 mg	5 mg	1 mg	Control	Virgin ♀*	(E)-11-tetradecenal 10 mg	5 mg	1 mg	Control	Virgin ♀
2	108	131	135	46	27	43	57	53	15	39
4	40	19	32	7	21	40	42	36	17	17
8	181	162	132	70	40	82	82	68	60	44
12	140	152	111	2	31	72	87	80	0	95
16	24	13	31	4	29	3	0	0	0	0
Total	493	477	441	129	148	240	268	237	92	195
Av./trap/night	10.27	9.94	9.19	4.03	4.63	7.5	8.38	7.41	2.88	6.09

* Two replicates

TABLE 2

1973 trapping results. Total numbers of male spruce budworm caught with (E)-11-tetradecenal (5 mg), virgin females and empty control traps.

Day	Sectar 1 pheromone	♀	control	Astrotrap pheromone	♀	control	Sectar XC-26 pheromone	♀	control
2	45	6	0	30	1	1	14	1	0
4	17	0	4	2	0	0	7	0	0
6	8	0	0	4	0	0	7	0	0
8	3	0	0	3	0	0	5	0	0
10	9	0	0	5	0	0	4	0	0
12	2	0	0	0	0	0	0	0	0
14	1	1	0	0	0	0	0	0	0
Total	85	7	4	44	1	1	37	1	0
Av./trap/night	2.02	0.17	0.1	1.05	0.02	0.02	0.58	0.02	0

Results (Tables 1 and 2) were subject to a "t" test. The 1972 data are given in Table 1. When traps were baited with synthetic pheromone there was no significant differences between the catches of the Astrotrap and the Sectar 1. The same treatment of 1973 data showed the Astrotrap to be good as the Sectar 1 and the Sectar XC-26 (Table 2).—J. Weatherston, Insect Pathology Research Institute, Sault Ste. Marie, Ont.

Preliminary Observations on the Vulnerability of White Spruce to Spruce Budworm Defoliation in Western Quebec.— Although balsam fir [*Abies balsamea*, (L.) Mill.] is the most vulnerable tree species to spruce budworm attack, it has been known for some time that white spruce [*Picea glauca* (Moench) Voss] and red spruce [*Picea rubens* Sarg.] can also succumb to attacks by this insect. The amount of white spruce destroyed through budworm defoliation has been estimated for the Algoma region of Ontario (Turner Can. Dep. Agric. Publ. 875, 1952), for northwestern Ontario (Elliott. For. Chron. 36:61-82, 1960), and for certain areas in New Brunswick (Swaine, Craighead, and Bailey. Can. Dep. Agric. Tech. Bull. 37 (n.s.), 1924), but there are no records of losses for this species in Quebec.

A large part of Quebec's white spruce occurs in the western part of the province (37% on the Ottawa River watershed) where the current spruce budworm outbreak started in 1967. The insect population is still at outbreak level in this area although some decrease is expected in 1976.

In the summer of 1975, a study was initiated to determine the degree of vulnerability of white spruce to budworm attack, rate of mortality of this tree species with respect to defoliation, stand age and composition, geographical location, and in relation to mortality of balsam fir, sequence of attack by various species of secondary insects, and rate of deterioration of killed trees. It may take two or three years to cover all aspects of this problem.

Ten line plots 60 m wide and varying in length, but averaging 0.5 km, were established in mature stands approximately 14 km apart along bush roads in the Dumoine and Coulonge river watersheds. These are the first in a series of plots to be established over a larger area. All living and dead white spruce trees within each plot were marked for continued observation. The number of trees varied from a minimum of 25 to a maximum of 60 per plot for a total of 341 trees.

Data obtained in 1975 by plot and for all plots including average diameter, number of living and number of dead trees, number and percentage of living trees by defoliation class (Table 1). Tree mortality varied among plots; in three plots there were no dead white spruce, while in one 55% of the white spruce was dead. For all plots, 20% of the trees were dead. Of those still living, 32% had lost from 50 to 99% of all foliage. Due to the lowered vigor of the severely defoliated trees many are expected to die in the course of the next few years. Less severely defoliated survivors of the current outbreak will suffer a loss in annual increment.

TABLE 1

Preliminary data form deterioration studies of white spruce in western Quebec, including average diameter, number of living and dead trees, number and percentage of living trees by defoliation class for each of ten plots, 1975

Plot No.	No. of trees	No. of living trees	No. of dead trees	Average DBH in cm	Number and percentage of living trees by defoliation class: Severe[1]	Moderate	Light
1	25	25	0	28.1	25 100%	0	0
2	27	25	2	37.1	6 24%	8 32%	11 44%
3	38	25	13	41.2	0	4 16%	21 84%
4	27	25	2	34.4	9 36%	5 20%	11 44%
5	31	25	6	36.8	2 8%	6 24%	17 68%
6	25	25	0	33.6	8 32%	9 36%	8 32%
7	60	28	32	29.0	2 8%	5 20%	18 72%
8	38	26	12	28.7	5 20%	7 28%	13 52%
9	37	35	2	30.4	6 24%	13 52%	6 24%
10	33	33	0	34.0	18 72%	6 27%	1 1%
Total	341	272	69	33.3	81 32%	63 25%	106 43%

[1] Severe 50-99% loss of total foliage.
Moderate 49-25% loss of total foliage.
Light 24-0% loss of total foliage.

Growth-ring studies, and records from limit holders in the study area indicate little, if any, white spruce mortality in this region at the time of the last budworm outbreak in the 1940's. White spruce in the region is cut mostly for the sawmill industry. Since dead trees become heavily infested by wood borers almost immediately upon dying, their value is considerably reduced the second year after attack. Pre-salvage and salvage operations should be a major concern of limit holders in the region. Black spruce is fairly abundant in this area and it is also cut for lumber. However, it is relatively immune to budworm damage so pre-salvage and salvage cutting of white spruce should take precedence over the harvesting of black spruce. More complete and detailed information covering a larger area will be published when it is available.—J. R. Blais, Laurentian Forest Research Centre, Sainte-Foy, Quebec.

Tests of an Introduced Parasite against the Native Elm Bark Beetle.—At the northerly limit of its range in North America, Dutch elm disease is spread almost solely by the native elm bark beetle, *Hylurgopinus rufipes* (Eichh.). Indigenous parasites do not seem to be effective in controlling populations of this beetle, in spite of a report by Kaston (Conn. Agric. Exp. Stn. Bull. 420, 1939) of up to 75% parasitism (average: 5-10%) by a native braconid, *Spathius benefactor* Matthews. For example, a 1974 mass rearing of *H. rufipes*-infested elm near Sault Ste. Marie yielded 19,911 beetles but only 191 *S. benefactor*. This suggests a parasitism rate of less than 1% in a full-blown beetle infestation.

A European braconid, *Dendrosoter protuberans* Wesm., has been introduced into the United States in recent years and is reported to be established in Wayne County, Michigan and possibly in Missouri (Peacock, J. W. 1975. Proc. IUFRO Conf., Minneapolis-St. Paul, Sept. 1973. USDA For Serv., Northeastern For. Exp. Stn., Upper Darby, Pa.). This is the most common parasite of the smaller European elm bark beetle, *Scolytus multistriatus* (Marsh.), in Europe and was introduced in an attempt to control this beetle in the United States. It seemed worthwhile to test *D. protuberans* against *H. rufipes* in central Ontario where the beetle parasite niche seems to be inefficiently filled.

On request, E. Rohringer of the Commonwealth Institute of Biological Control provided 788 *D. protuberans* from the vicinity of Tulln, Austria in the spring of 1972. This stock was reared continuously on *H. rufipes* larvae for over 2 years with no problems; generations could even be switched back and forth between *H. rufipes* and *S. multistriatus*. Although the first rearings employed beetle-infested sticks of dead elm, subsequently only infested bark was used, in plastic box cages. The female parasites oviposited through both inner and outer bark surfaces, and emergence occurred through both as well. Parasites were reared successfully on larvae in elm bark collected in the field in winter and stored in a freezer for up to 9 months.

Outdoor overwintering tests were carried out at Sault Ste. Marie and Angus, Ont. during 1972-1973 and at Sault Ste. Marie during 1973-1974. In the first test, short bolts of elm, infested with either *H. rufipes* or *S. multistriatus*, were kept on the ground under snow, and on racks above snow. In the second test, elm logs 2.5 m long and infested with *H. rufipes* only, were stood vertically outdoors, with the snow line being marked periodically during the winter. In both cases, parasite attack was conducted in cages outdoors in the fall to acclimatize the progeny before the onset of cold weather. The results of the tests are given in Table 1.

Examination of sticks and logs showed that mortality above snow occurred in the parasite cocoons when the insects were in the pupal and adult stages. In the Angus test, parasitism was very heavy; in some galleries almost all beetle larvae were parasitized, both above and below the snow. In both tests, unparasitized beetles above and below snow completed development normally. Both winters were relatively mild, the temperature at both sites never dropping below −31°C.

TABLE 1

Emergence of overwintered *D. protuberans*

Place	Year	Under Snow ♂	Under Snow ♀	Under Snow Total	Bark area (m^2)	Above Snow ♂	Above Snow ♀	Above Snow Total	Bark area (m^2)
Angus	1973	652	1,181	1,833	1.72	0	0	0	1.53
S.S. Marie	1973	102	70	172	1.09	0	0	0	1.15
S.S. Marie	1974	34	71	105	0.52	0	0	0	0.57

In 1972, free releases of 304 and 475 *D. protuberans* were made at two points near Sault Ste. Marie. Repeated cagings of infested elm wood from these points in 1973 and 1974 failed to produce any parasites.

Only a very small proportion of larval elm bark beetles overwinters below the snow. It must therefore be concluded that *D. protuberans* could not contribute effectively to the control of *H. rufipes* (or *S. multistriatus*) in central Ontario.—L. M. Gardiner, Great Lakes Forest Research Centre, Sault Ste. Marie, Ont.

PATHOLOGY

Some Fungi Isolated from the Seeds of *Kalmia angustifolia*.—*Kalmia angustifolia* L., commonly known as sheep laurel, is an ericaceous shrub which frequently invades productive forest sites in Newfoundland following logging and/or fire. The development of this shrub interferes with the natural regeneration of commercially important species, and causes site deterioration. In order to develop a control for this species, it is imperative to understand its biology, ecology and physiology, including germination of its seeds. As a part of these investigations, a study was undertaken to determine the microfungi associated with the seeds of the species.

Seeds of *K. angustifolia* were collected in October from a location southwest of Gander in central Newfoundland (48°48'N, 54°55'W). The seeds were stored in a clean, wide-mouthed glass vial for a few days in the laboratory at a temperature of 24°C and a relative humidity of 42%.

To isolate the externally seed-borne fungi, 100 unwashed and 100 washed (three successive washings in sterile distilled water, 1 to 2 minutes each) seeds were placed in 40 sterilized petri dishes (5 seeds per dish) containing blotting paper moistened with 0.2% solution of the sodium salt of 2,4-dichlorophenoxy-acetic acid ('Blotter method' — Intern. Rules for Seed Testing, Proc. Int. Seed Test. Assoc., 31: 1-152, 1966). For

TABLE 1

List and frequency percentage of fungi isolated from the unwashed, washed and surface sterilized seeds of *Kalmia angustifolia*

Fungal species	Frequency %* Unwashed	Frequency %* Washed
Phycomycetes		
Cunninghamella echinulata Thaxter	34	2
Mucor hiemalis Wehmer	33	—
Rhizopus nigricans Ehrenberg	52	4
Basidiomycetes		
Rhizoctonia sp.	8	—
Fungi Imperfecti		
Alternaria tenuis Nees	76	8
Aspergillus flavus Link	36	—
Aspergillus fumigatus Fresenius	26	2
Aspergillus nidulans (Eidam) Winter	22	7
Aspergillus niger van Tieghem	31	—
Aureobasidium pullulans (de Barry) Arnaud	4	—
Cladosporium herbarum (Pers.) Link	6	2
Colletotrichum sp.	14	3
Diplodia sp.	5	—
Fusarium nivale (Fr.) Ces.	23	4
Monilia sp.	10	—
Papulaspora sp.	9	—
Penicillium sp.	19	2
Trichoderma viride Pers. ex S. F. Gray	8	—

* Based on 100 seeds plated in 20 petri dishes for each treatment.

the isolation of internally seed-borne fungi, 100 seeds were surface sterilized by washing in 0.1% aqueous mercuric chloride for 1 to 2 minutes, followed by three washings in sterile distilled water, 2 to 3 minutes each. After surface sterilization, the seeds were similarly plated in petri dishes. The petri dishes were incubated in darkness at 25°C and examined every alternate day for 15 days for the growth of fungi.

The fungi were purified, subcultured and maintained on 2% malt agar slants for further examination and identification. The prevalence of a species was expressed as the percentage occurrence from 100 seeds.

A total of 18 species of fungi were isolated from the seeds of *K. angustifolia* (Table 1). All of them were obtained from unwashed seeds, nine from the seeds washed with sterile distilled water, and none from the seeds surface sterilized with mercuric chloride. The data show that the seeds did not carry any internally seed-borne fungi. On the unwashed seeds *Alternaria tenuis* was the most abundant species and *Aureobasidium pullulans* the least common. On the washed seeds *Alternaria tenuis* was also the most abundant species.

The parasitic potential of these fungal species on *Kalmia* has not been demonstrated but the importance of seed microflora, including some of these fungi, on the viability of seeds, in general, is well recognized (Malone & Muskett, Seed-borne fungi. *In* Proc. Int. Seed Testing Assoc. *29*: 179-384, 1964). Although all fungi isolated in this study were externally seed-borne, some of them may influence the germination of *Kalmia* seeds.—Pritam Singh, Newfoundland Forest Research Centre, St. John's, Nfld.

Fungicide Use in Relation to the Compatibility of Damping-off Fungi.—Damping-off and root rots in forest nurseries are caused by three very different fungi, *Rhizoctonia solani* Kuehn, *Fusarium oxysporum* Schlecht., and *Pythium* spp., as well as by other soil-borne organisms. Because two or more of these pathogens can often be detected within the same seedling, questions arise concerning primary and secondary invasion, synergism, and additive effects or antagonism among the various pathogens. It is important to know if all of these pathogens must be suppressed to control damping-off and if interactions occur among them, further complicating their control.

Earlier work with pure and mixed cultures indicated no antagonistic or synergistic effects between *Rhizoctonia solani* and *Fusarium oxysporum*. Both invaded aseptically-grown pine seedlings with slight but non-significant additive effects in pathogenicity (Lindsay, B.Sc. Thesis, University of New Brunswick, 1974).

Since the additive effects might be more pronounced in non-sterile soils, a forest soil (fine sandy loam, pH 4.4, 4.6% organic matter, 18.2% field capacity) that contained no detectable traces of these or other damping-off fungi, was mixed in equal proportions with cornmeal-sand cultures of *Rhizoctonia solani, Pythium irregulare* or *Fusarium oxysporum*. These inoculated soils were then mixed in equal proportions with each other or with uninoculated soil (forest soil plus uninoculated cornmeal-sand) to give eight combinations with a high inoculum level (Table 1). Each soil combination with the high inoculum level was in turn mixed with nine parts of uninoculated forest soil to give low inoculum levels. Soil pH was raised to 5.5 with saturated calcium hydroxide solution. *Fusarium* populations, monitored by dilution-plating on a modified Martin's peptone agar (Wensley and McKeen, Can. J. Microbiol. 8: 57-64, 1962), were about 20,000 propagules per gram of soil at the high inoculum level, and about 2,000/g at the low level. *Rhizoctonia* recovery after 4 days incubation with buckwheat stem segments was 28% and 18% at the high and low inoculum levels, respectively, using the method of Davey and Papavizas (Can. J. Microbiol. 8:847-853, 1962).

TABLE 1

Damping-off in soil inoculated with various combinations and levels of *Rhizoctonia solani, Pythium irregulare* and *Fusarium oxysporum*

	Damping-off, %[1]			
	High inoculum level		Low inoculum level	
Inocula	Jack pine	Black spruce	Jack pine	Black spruce
Rhizoctonia	76[b2]	60[ab]	39[abc]	45[bc]
Phythium	4[a]	13[ab]	11[ab]	45[c]
Fusarium	2[a]	28[ab]	11[ab]	14[abc]
Rhizoctonia + Pythium	80[b]	85[b]	58[bc]	21[abc]
Rhizoctonia + Fusarium	91[b]	80[b]	88[c]	22[abc]
Pythium + Fusarium	5[a]	12[ab]	1[a]	10[ab]
Rhyzoctonia + Pythium + Fusarium	95[b]	38[ab]	77[c]	41[bc]
Uninoculated control	1[a]	2[a]	1[a]	1[a]

[1] Percentage of emerged seedlings, averaged from arcsin % transformed values.

[2] Figures within a column followed by the same letter not significantly diferent at P = 0.05, using Duncan's Multiple Range Test.

TABLE 2

Survival of seedlings in grenhouse soil inoculated with *Rhizoctonia solani*, *Pythium* spp., and *Fusarium oxysporum* and drenched with fungicides at the time of seeding and after emergence, and in naturally colonized nursery plots drenched at the time of seeding

		Seedling survival[1]		
		Greenhouse		Nursery
Fungicide	Rate (g/sq m)	Jack pine	Black spruce	Red pine
Benomyl	0.1	172(Py)[2]	100	95
Captan	1.0	114(Rh)	40(Fu)	114
Chloroneb	1.0	314	190	110
Ethazole	1.0	171	70(Fu)	129
Quintozene	10.0	57(Py,Fu)	80	39
Thiabendazole	1.0	129	50	39
Benomyl + captan	0.1 + 1.0	157	170	124
Benomyl + chloroneb	0.1 + 1.0	265	240	109
Benomyl + ethazole	0.1 + 1.0	228	290	82
Captan + ethazole	1.0 + 1.0	180	160(Fu)	124
Captan + thiabendazole	1.0 + 1.0	108(Py)	20	—
Ethazole + quintozene	1.0 + 10.0	200	260	50
Ethazole + thiabendazole	1.0 + 1.0	200	190	115
Ethazole + thiophanate methyl	1.0 + 1.7	334	440	125

[1] Percentage of untreated checks; underlined values significantly greater than the untreated check of P = 0.05.

[2] Fungi isolated from 10 or more diseased seedlings, Py = *Pythium*, Fu = *Fusarium*, Rh = *Rhizoctonia*.

The soils were placed in styrofoam cups and planted with 15 conifer seeds. The containers were enclosed in plastic bags to minimize cross-contamination and were arranged in four replicate blocks on a greenhouse bench (night temperature 20°C, day length 16 hours). Emergence and mortality were recorded twice weekly until seedling numbers stabilized.

Results with two tree species, jack pine and black spruce, and the two inoculum levels indicated few significant interactions among the three fungi in causing damping-off (Table 1). In this experiment, severe mortality was associated with *Rhizoctonia* but in other experiments, using essentially the same methods, damping-off depended mainly on other fungi, including *Fusarium*.

Control is difficult when three or more compatible, but otherwise different, pathogens coexist in the same soils, because fungicides, to be effective, must have either a broad spectrum of activity or they must be combinations of specific compounds. Vaartaja (Bot. Rev. 30: 1-91, 1964) has reviewed the results of combining some of the older fungicides.

Drenches of various commercial fungicides were applied to soils after planting in both the greenhouse and a local nursery (Table 2). The soil used in the greenhouse had natural populations of *Pythium* and *Fusarium* in addition to the above three inocula. The nursery soil had been treated with vapam 3 weeks prior to seeding and had subsequently become naturally colonized with *Pythium, Rhizoctonia,* and *Fusarium.* Damping-off was severe in areas not drenched with fungicides.

Combinations of fungicides were generally more effective in controlling damping-off than were single chemicals

(Table 2). The latter reduced the recovery of certain groups of fungi but often did not prevent damping-off under the rigorous conditions of these tests. For instance, in benomyl treated soils the recovery of *Fusarium* was reduced but that of *Pythium* was greater. Quintozene suppressed *Rhizoctonia*, but not the other two major fungi; furthermore it was phytotoxic in field tests. Combinations of ethazole to suppress *Pythium* plus a systemic benzimidazole (benomyl or thiabendazole) to suppress other groups were generally most effective. Chloroneb plus a benzimidazole also was a promising combination.

Because of possible interactions among chemicals, new combinations should be tested widely before they are recommended for general use.—R. E. Wall, Maritimes Forest Research Centre, Fredericton, N.B.

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