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# bi-monthly research notes

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#### ENTOMOLOGY

Relative Toxicity of Aminocarb to Wandering and Mining Second-instar Larvae of the Spruce Budworm.—Second-instar (L<sub>2</sub>) larvae of the spruce budworm, Choristoneura fumiferana (Clem.), usually settle on old foliage as needle-miners after emerging from hibernacula in the spring. Upon reaching the third instar, the larvae migrate to buds and expanding shoots, where they feed until pupation. It was the practice in Quebec from 1974 to 1977 to time first spray applications against  $L_2$  at 25-50% emergence. The rationale for this advanced timing was the reduction of population densities to theoretically manageable numbers so that subsequent applications vs. L<sub>3</sub>-L<sub>4</sub> would provide more reasonable levels of foliage protection. However, the overall results of such a spray regime were variable and foliage protection was often marginal. This has led to considerable speculation as to cause, a major question being the advisability of spraying L<sub>2</sub> emerging from hibernacula (Blais, Can. J. Forest Res. 9:354-361, 1979).

As part of a cooperative study on the timing of aerial sprays against larvae of the spruce budworm during 1978 (Blais et al. Can. J. Forest Res., in press), a concomitant experiment was conducted to determine (1) the residual effectiveness of an application of insecticide vs. L2 larvae and (2) the susceptibility of L2 wandering over, or mining in, foliage. The experiment was conducted from field laboratory facilities near Rimouski, Que. Tests of residual toxicity of insecticide applied to operational spray block 101, located south of La Pocatière, were performed with healthy L2 larvae that were supplied by the Quebec Department of Lands and Forests from collections in unsprayed areas. Insecticide treatment was aminocarb (Matacil®) emitted at 53 g AI in 1.2 L spray mix/ha (0.75 oz AI in 16 fl. oz/acre) via Douglas DC-6 aircraft. The weather during most of the application (1748-1945 h) was less than ideal: temperature 13-17°C with a strong lapse (air rising) ground condition, relative humidity 32-44%, northerly crosswind 0 to 5 km/h, and cloud cover 0%. Accordingly, droplet recovery on Kromekote® sample cards averaged less than 7/cm<sup>2</sup> over the 22760 ha block. Only a few preselected sampling stations near the northest corner of the block received more than 10 small (ca. 90 µm VMD) droplets/cm<sup>2</sup>. Stations P01 and P03, where x deposit was ca. 15/cm<sup>2</sup>, were selected as representative of a good spray deposit for comparison with a near-average deposit (5 droplets/cm<sup>2</sup>) as represented by station P05.

Five 45 cm branch tips were collected from the midcrown of balsam fir (*Abies balsamea* [L.] Mill.) at sampling stations P01 and P03 at intervals of 1, 3, and 5 days after treatment. Branch tips from untreated fir near Rimouski were used for check purposes. The samples of clipped foliage from sprayed and unsprayed trees were placed in 1L ice-cream cartons; five replicates were used for each collection from the sprayed block and five for each check. Twenty

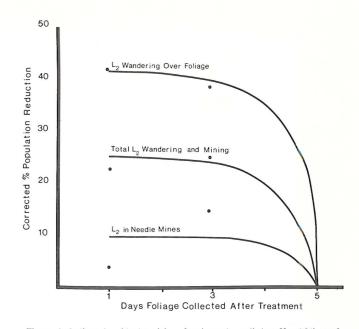


Figure 1. Indicated residual toxicity of aminocarb applied at 53 g A1/ha to L<sub>2</sub> larvae of the spruce budworm.

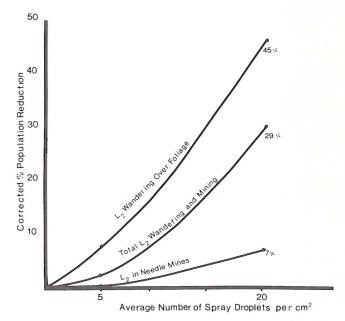


Figure 2. Trends in mortality of 50%-emerged  $L_2$  larvae within 24 h of application of aminocarb at 53 g Al in 1.2 L/ha.

healthy  $L_2$  larvae were then placed on the foliage in each carton. After 2 days the foliage and the containers were carefully examined for evidence of living or dead larvae.

In addition, visual observations were made by a four-man crew at P01, P03, and P05 within 24 h of treatment to assess the impact of aminocarb treatment on exposed (wandering) and hidden (mining) larvae. Since the wind was nil to only very light during this period, it was assumed that most affected larvae would not yet have fallen off and that the number of dead and living  $L_2$  found on the foliage reflected the effect of the treatment. Twenty 45 cm midcrown branches from balsam fir were carefully examined for living and dead larvae. Six branches from nearby untreated check station 100-02 were likewise examined.

The laboratory study with L<sub>2</sub> placed on sprayed and unsprayed

foliage indicated that about 25% of all emerged larvae were affected by the treatment during the first 3 days after treatment (Fig. 1). Larvae that wandered over the foliage were apparently about 4 times as susceptible as those that quickly entered needles: about 40% of all unestablished larvae were dead vs. an average of 10% of those established in needles. The fact that larvae placed on foliage collected 5 days after spraying were not affected indicated rapid loss of the residual toxicity of the insecticide.

Companion observations of toxicological effect in the field within 24 h after treatment indicated a very similar impact on  $L_2$ : about 29% of all larvae found on branches from P01 and P03 were dead; nearly half (45%) of the exposed larvae found were dead while less than 10% of the  $L_2$  population in needle mines were dead (Fig. 2). Only about 5% of larvae from branches in P05, which received only light spray deposit, and of those in the untreated check block were dead.

These observations indicate obvious limitations and difficulties associated with early spray application: (1) Larvae in mines (and most likely those in hibernacula) are not greatly affected by an aerial spray treatment as described. The greatest impact was on that portion of the L<sub>2</sub> population that wandered upon emergence from overwintering sites. However, not more than half of these were killed, even at spraydroplet densities to 20/cm<sup>2</sup>. The overall corrected population reduction of emerged larvae was calculated at less than 30%, or approximately 15% of the whole population (emerged + unemerged). At lower spray deposits, the effect of treatment was apparently close to nil. (2) The treatment of aminocarb, as applied, indicated short residual toxicity, the greatest impact on L<sub>2</sub> occurring within the first 3 davs. Thus a considerable part of the unemerged larvae (approx. 50% of the population) would be unaffected because of the timespan necessary for the bulk of L<sub>2</sub> emergence. (3) Under conditions of application, the study indicated that spray impact is critically related to coverage and the residual toxicity of the insecticidal treatment: inferior droplet distribution and/or the rapid degradation of the chemical insecticide can greatly inhibit the success of early spray application against larvae of the spruce budworm.-R.F. DeBoo, Forest Pest Management Institute, Sault Ste. Marie, Ont. and J.R. Blais, Laurentian Forest Research Centre, Ste. Foy, Que.

#### **INSECT PATHOLOGY**

Development of the Enzyme-linked Immunosorbent Assay to Detect a Nuclear Polyhedrosis Virus in European Pine Sawfly Larvae.—Periodically, the European pine sawfly, *Neodiprion sertifer* (Geoff.), severely defoliates young pines in North America and Europe. A nuclear polyhedrosis virus (NPV) is an effective agent for the control of this insect. Studies are being conducted to investigate this baculovirus as a regulatory agent, and this involves a great deal of diagnostic microscopy either when NPV is artificially introduced into insect populations or when it is studied in natural situations.

Two enzyme-linked immunosorbent assay (ELISA) methods, double-antibody and indirect (Voller et al., Bull. World Health Organ. 53:55-65, 1976), have been investigated as possible techniques for the diagnosis of NPV in sawfly larvae. The ELISA system has already been employed to detect numerous plant and insect viruses, the double-antibody technique being the one more widely applied. Briefly, the ELISA system is an immunological technique performed in plastic microtiter wells in which the presence of a specific antigen results in a color change in reagents. Tests were conducted with both ELISA techniques to monitor polyhedral inclusion bodies (PIB), virus particles (VP), or polyhedral-inclusion-body protein, polyhedrin (PN), in purified suspensions. Results from these experiments were used to develop a procedure to detect the presence of NPV in larval extracts.

PIB's and VP's were purified according to procedures described by Harrap et al. (Virology 79:4-31, 1977) and Brown et al. (Virology 81:317-327, 1977) respectively. Dissolved PIB's were used to provide a crude suspension of PN. Since PIB's are alkaline labile, crude PN was produced by dissolving suspensions of PIB's in a known volume of 0.1M Na<sub>2</sub>CO<sub>3</sub> for 7 min and then adding an equal volume of 0.1M HCl. The concentrations of all PIB suspensions were estimated by a microscopic dry counting method that involves counting PIB's in a known area across the radius of a standard circular stained smear (Wigley, D. Philos. Thesis, University of Oxford, 1976). Protein

TABLE 1 Limits of detection by two ELISA methods (ng protein mL)

Antigen	Indirect method	Double-antibody method		
VP	10	100		
PN	10	100		
PIB	560	560		

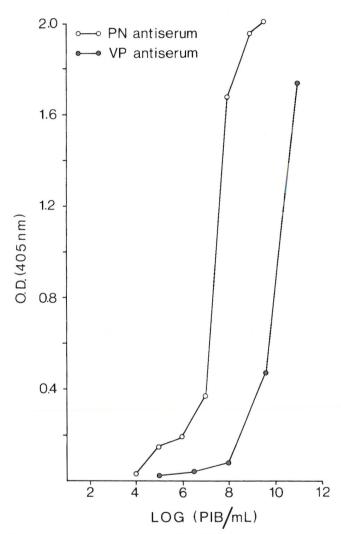
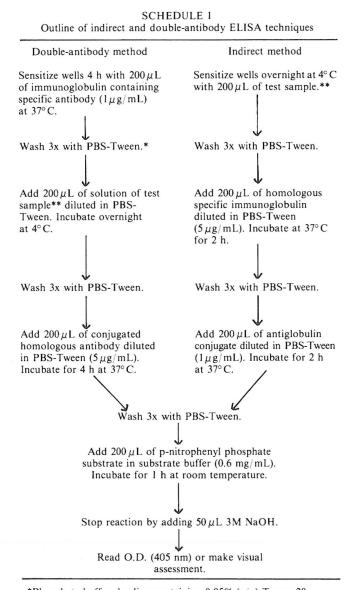


Figure 1. Relative levels of detection of VP and PN in suspensions of dissolved PIB's by means of homologous antisera.

estimates were made on all PIB and VP suspension by the method of Lowry et al. (J. Biol. Chem. 193:265-275, 1951), in which bovine serum albumin was used as a standard.

Antisera against PIB's, VP's and PN were prepared by the methods of Kelly et al. (J. Gen. Virol. 40:465-469, 1978). Partial purification and standardization of the Y gobulins and conjugation of both the homologous and antiglobulin-Y globulin with alkaline phosphatase were carried out by the methods outlined by Kelly et al. (Ann. Appl. Biol. 90:369-374, 1978), and the substrate enzyme was pnitrophenyl phosphate. Initially, a range of coating globulins (10-0.1  $\mu$ g/mL) and conjugate (10-0.1  $\mu$ g/mL) were employed to determine the most efficient combinations of antisera used in each method. An outline of the respective ELISA techniques developed for these tests is shown in Schedule 1. All tests were performed in wells in polystyrene M29AR microtiter plates (Dynatech Ltd., Billinghurst, Sussex,



\*Phosphate-buffered saline containing 0.05% (v/v) Tween 20.
\*\*Samples can be aliquots of alkali-treated larval extracts or alkali-treated purified PIB's.

England). Optical densities of each sample well in the plates were determined on a Pye-Unicam 1800 spectrophotometer at 405 nm wavelength and the values used were the mean of four replicates.

Results indicated that in homologous reactions the indirect ELISA method was more sensitive than the double-antibody method in detecting VP and PN (Table 1). The reactions involving PIB's were poor, perhaps because the inclusion-body surface "membrane" interfered with antibody production (Kelly et al., 1978). Generally, PIB's do not elicit the production of high-titer antisera, and this has been demonstrated in complement-fixation tests for N. sertifer NPV PIB's. Thus intact PIB's were eliminated as an antigen in these investigations. It was also decided to employ the indirect ELISA method because of its sensitive range of detection, ease of application, and greater versatility. Here, the antigen solution was introduced first into the wells, and not onto a sensitive layer of antibody-coated plastic. The indirect ELISA method also has the advantage that a single antiglobulin conjugate can be employed against antisera to numerous NPV's providing the viral antisera are produced in the same species of animal.

By the indirect method, various standard curves were produced illustrating the relationship between optical density and quantities of VP protein and crude PN utilized in homologous reactions. In conjunction with respective PIB concentrations and protein estimates, these standards were employed to develop the regression curve log x =  $4.65 + 1.58 \log y$  (r<sup>2</sup> = .99), which describes the relationship between concentration of PIB's/mL (x) and ng VP protein/mL (y) for N. sertifer NPV. This mathematical relationship was used to transform the standard curve for VP protein detection into equivalent PIB's/mL (Fig. 1). Hence, by monitoring dissolved suspensions of PIB's for PN, more-intense color reaction and lower detection level of PIB's can be obtained than if VP antiserum is employed. This is not surprising, since sawfly NPV's are small, singly embedded baculoviruses yielding few virus particles per polyhedron by comparison with the multiply embedded NPV's of some Lepidoptera. Sawflies also produce fewer PIB's per larva than many other insect species because the virus replicates only in midgut cells. Dissolving the PIB's increases the amount of detectable antigen available when anti-PN antiserum is used.

The indirect ELISA method was therefore selected to monitor PN in alkali-dissolved larval extracts for the detection of NPV. Aliquots of treated larvae were transferred to microtiter wells and monitored for PN (Schedule 1). A detection level of  $4 \times 10^5$  PIB's per larva (background O.D.=0.12) was obtained. It is quite probable that an even lower detection level can be obtained once dilution factors obtained during trituration are reduced. By comparison with microscopic examination of infected larvae, whose detection limit is about 10<sup>7</sup> PIB/larvae, the high level of sensitivity of the ELISA technique can be appreciated.

This application of the indirect ELISA method indicates its potential as an efficient, rapid, and reliable immunological technique for the detection of N. sertifer NPV. It is estimated that, for large numbers of larvae, this technique is three times faster than microscopic diagnosis. With low detection levels and high specificity, it permits much earlier and more confident diagnosis of NPV infection in sawfly larvae, thus providing new information on virus infection and epizootiology.—W.J. Kaupp, Forest Pest Management Institute, Sault Ste. Marie, Ont.

#### PATHOLOGY

Assessing Nectria macrospora as a Biological Control Agent for Hemlock Dwarf Mistletoe.-The fungus Nectria macrospora (Wr.) Ouellette parasitizes hemlock dwarf mistletoe (Arceuthobium tsugense [Rosendahl] G.N. Jones) in coastal British Columbia (Funk et al., Can. J. Forest Res. 3:71-74, 1973). Because it reduces aerial shoot production of the mistletoe without damaging the host tree, western hemlock (Tsuga heterophylla [Raf.] Sarg.), the fungus might be used for biological control. Techniques for culturing N. macrospora and inoculating mistletoe swellings have been developed (Funk et al., 1973). However, since the fungus has not been tested under field conditions, we made a series of studies to investigate its usefulness in nature. In 1973, three experiments, each with up to 30 replicates of treated and untreated swellings, were carried out in the Jordan River area near the coast of southern Vancouver Island, in March, August, and November. The results of these tests were inconclusive. In October 1975, however, an experiment was made at Cowichan Lake (inland, southern Vancouver Island), in which the results were encouraging. That experiment is reported here.

Fungus inoculum was grown on 100 mL of 2% malt extract broth in 500 mL Erlenmeyer flasks, and one 15-day-old mycelial mat, with conidia, was blended in 200 mL of water for 15 sec. About 10 mL of spore suspension were sprayed on each of 20 labelled mistletoe swellings and their aerial shoots. Twenty swellings, about 1 km away, were sprayed with distilled water. Before treatment in 1975 and again in 1978, data on the following were collected on each mistletoe swelling: length and diameter, maximum distance between aerial shoots as measured along the swelling, number of aerial shoots, and height of the tallest shoot. The age of each swelling was determined at the end of the experiment by counting the annual rings in the affected host-tree branch. Successful establishment of *N. macrospora* was based on field identification of its conidial state produced on cankered tissue and confirmed by laboratory examination at the conclusion of the experiment.

In terms of measured mistletoe plant and swelling characteristics,

#### TABLE 1

Comparison of dwarf mistletoe swelling characteristics (Nectria colonized and noncolonized) for initial (1975) and final (1978) observations\*

		Swelli	ng size	Average maximum distance between	Average number	Average maximum
	Average age (yr)	Average length (mm)	Average diameter (mm)	aerial shoots (mm)	of aerial shoots	shoot height (mm)
1975						
Noncolonized	5.7 a	91.5 a	10.7 a	42.2 a	13.5 a	48.5 a
Colonized	4.6 a	98.4 a	11.8 a	43.5 a	19.5 a	53.2 a
1978						
Noncolonized	8.7 a	148.5 a	15.2 a	92.5 a	14.1 a	19.6 a
Colonized	7.6 a	135.4 a	15.6 a	69.6 b	1.4 b	11.5 b
1975-1978 (difference)						
Noncolonized		57.0 a	4.5 a	50.3 a		
Colonized		37.0 b	3.8 a	26.1 b		

•For pairs of data within each column, means followed by a common letter are not significantly different at P = 0.05. There were 28 uninfected and 10 infected swellings. One swelling colonized by *N. macrospora* and one by a *Tympanis* sp. were removed for identification in the laboratory before the end of the experiment and could not be included in these comparisons.

no significant differences (P=0.05) occurred in 1975, before spraving, between the treated and the untreated groups. Subsequently, eight of the mistletoe swellings in the treated group and three in the untreated group became colonized by N. macrospora. The difference between the proportions colonized was not statistically significant (Dixon and Massey, Introduction to statistical analysis, McGraw-Hill, 1957). By 1978, the treated swellings averaged fewer and smaller aerial shoots than the untreated, but differences were not statistically significant. However, when all colonized swellings (treated plus untreated) were compared with all those without the fungus in 1978, the colonized group had significantly fewer and smaller aerial shoots, shorter maximum distance between aerial shoots, and a slower rate of swelling elongation during the 1975-78 period (Table 1). Because of unequal variances in several group comparisons, all t-tests were based on nonpooled variances (Ryan et al., Minitab student handbook, Duxbury Press, 1976).

Spray inoculations with *N. macrospora* did not significantly increase the level of colonization of hemlock dwarf mistletoe swellings. Results confirmed, however, that, once mistletoe swellings are colonized, the fungus significantly reduces the production and growth of the aerial, reproductively active portions of the mistletoe. An improved inoculation technique is one objective that will have to be attained before *N. macrospora* can be offered as an effective biological control agent for hemlock dwarf mistletoe. Further trials are planned for the fall of 1980.—R.B. Smith and A. Funk, Pacific Forest Research Centre, Victoria, B.C.

#### SILVICULTURE

Response of Douglas-fir Seedlings and Xiphinema bakeri Nematode to Fertilizers.—The mutual interaction among tree host, phytonematodes, and fertilizers is complex. Despite a general increase in nematode population on the host following fertilization, yields may increase (Jones, pages 233-258 *in* Proc. 12th Colloq. Int. Potash Inst., 1976) or disease symptoms decrease by comparison with unfertilized checks (Johnson, Plant Dis. Rep. 55:1126-1129, 1971).

The nematode Xiphinema bakeri Williams and the fungus Cylindrocarpon destructans (Zinnsm.) Scholten are associated with corky-root disease, which can occur on important species of conifer seedlings grown in British Columbia. Major plant nutrients are generally less abundant in soils with corky-root pathogens than in soils without pathogens. Sutherland and Sluggett (Can. J. Forest. Res. 3:299-303, 1973) suggested that protection against X. bakeri not killed by present control methods of bare fallowing and disking might be achieved by increasing the fertility of pathogen-infested soil to offset the nematode feeding. A preliminary trial was therefore established to

study the relationship between different mineral fertilizers and their effect on X. bakeri and on Douglas-fir seedling growth.

At the British Columbia Forest Nursery, Campbell River ( $50^{\circ}$  01' N, 128° 18'W), Vancouver Island, soil samples were collected randomly from panel 5, where *X. bakeri* had previously caused seedling damage. As the sandy loam soil had recently been disked, 3.4 kg (fresh weight) were placed directly in plastic pots 15 cm in diameter and 18 cm deep and transported to the laboratory. Soil in 10 check pots (treatment I below) was heated in enclosed polyethylene bags in an oven at 50°C for 48 h, which killed all nematodes in these pots.

The experiment consisted of nine fertilizer treatments, including a check (treatment 1), each replicated 10 times. The treatments were: (1) at the currently recommended fertilizer rate, 374 kg/ha, combining a mixed fertilizer of 10-52-10 and a top dressing of ammonium sulphate for an N, P, K content of 179, 161, and 34 kg/ha, respectively; (2) at 0 kg/ha (i.e. with no fertilizer); (3) at 448 kg N/ha as ammonium nitrate; (4) at 74 kg P/ha as phosphoric acid; (5) at 88 kg K/ha as potassium chloride; (6) at 176 kg K/ha as potassium chloride; (7) at 896 kg Ca/ha as calcium chloride; (8) at 280 kg Mg/ha as magnesium chloride; and (9) with a complete fertilizer, at 1 786 kg/ha, combining N, P, K, Ca, and Mg at 448, 74, 88, 896, and 280 kg/ha, respectively. Two rates of K were used, because Kirkpatrick et al (Phytopathology 54:706-712, 1964) and Sivapalan (pages 73-74 *in* Tea Res. Inst. Annu. Rep., Part. 2, 1968) showed that nematode pathogenicity is inversely related to leaf potash content.

From 1-yr-old Douglas-fir styroplug seedlings of uniform height (Vyse et al., Pac. Forest Res. Cent. Rep. BC-X-58, 1971), with ectomycorrhizae on about 10% of feeder roots, a seedling was transplanted to each pot and allowed to grow for 13 days in the greenhouse before nematode inoculation. All pots, except the check, were inoculated with X. bakeri. A 200 g soil inoculum that contained an average of 81 X. bakeri was placed on the soil of the pots to be inoculated. This inoculum was obtained by growing, during the previous 3 mo, the common chickweed (Stellaria media [L.] Vill.) in flats with Deerholm sand infested with X. bakeri to increase the nematode population (McElroy, J. Nematol. 4:16-22, 1972). The inoculum, plus the natural nursery soil population, gave an initial population of 3.8 X. bakeri per 100 g soil dry weight.

Twenty-nine days after seedling transplant, fertilizer elements for each treatment were dissolved in separate 100 mL aliquots of distilled water and added to the pots. The top dressing of ammonium sulphate used in treatment I was applied 7 days later. Levels of soil nutrients (% total N, extractable P, and exchangeable K, Ca, Mg) were determined during the week following collection, and the actual rates applied were less than those estimated as adequate for this soil (Sutherland and Sluggett, Can. J. Forest Res. 3:299-303, 1973) but more than van den Driessche (B.C. Forest Serv. Res. Notes 48, 1969) considered to be optimal. This compromise was necessary to prevent possible nutrient toxicity but still give sufficient nutrients to meet the experimental objective.

The pots were placed in a completely randomized design in the shadehouse. The few weeds that grew in the pots were pulled as they appeared. The pots were watered as required, about once weekly, until harvesting of the seedlings, 26 wk after fertilizer application.

Stem heights were measured and seedlings were removed from the pots. The plants were gently shaken to remove soil loosely adhering to the roots. Stems were separated from roots, which were placed in containers for nematode extraction. After nematodes were washed from roots, fresh and ovendry (70° C) weights of plant tops and roots were taken. Soil moisture content was estimated on 100g samples dried at  $105^{\circ}$  C.

Six random subsamples of soil from each pot were thoroughly mixed for a 200g aliquot sample, from which nematodes were extracted by the Christie and Perry method (Proc. Helminthol. Soc. Wash. 18:106-108, 1951) but with a final washing screen of  $45\mu$ m. Nematodes from roots were obtained by quickly submerging the roots in a pail half filled with water, washing the roots with a stream of water, and extracting the nematodes by the Christie and Perry method.

The nematode data were transformed to square roots, to compensate for widely varying counts and zeros for X. bakeri in the check, and analyzed as a completely randomized design. The significance ( $P \le 0.01$ ) of each mean difference was determined by

Fertilizer	kg ha	Total stem height cm	Post- treatment height increase cm	Stem dry weight g	Root dry weight g	Other nematodes <sup>1</sup> per 100 g soil	X. bakeri per 100 g soil	X. bakeri per g root	Soil pH	
Check	374	37.2 a <sup>2</sup>	17.9 a	10.0 a	12.5 a	215	0.0 c	0.0 b	5.2	
None	0	24.5 c	6.5 b	4.2 c	4.9 b	91	12.1 a	82.2 a	5.4	
N	448	36.3 a	18.2 a	9.9 a	11.3 a	131	4.2 ab	14.4 b	4.3	
Р	74	25.0 c	7.8 b	4.0 c	4.4 b	87	9.2 a	66.4 a	5.4	
К	88	24.5 c	7.0 b	3.9 c	5.1 b	79	11.0 a	75.5 a	5.2	
К	176	26.4 c	7.9 b	4.0 c	5.2 b	84	9.1 a	51.9 a	5.0	
Ca	896	30.3 b	10.8 b	3.3 c	2.9 b	35	5.7 ab	66.4 a	5.1	
Mg	280	25.4 c	7.7 b	3.6 c	5.0 b	49	8.8 a	56.2 a	5.1	
Complete	1 786	39.1 a	20.4 a	7.4 b	4.2 b	99	2.4 b	13.4 b	4.6	

TABLE I

Nematodes other than X. bakeri per 100 g soil dry weight at the end of the experiment.

<sup>2</sup>Values in a column without letters, or those having the same letter, are not significantly different (P  $\leq 0.01$ ).

Student-Newman-Keuls' multiple range test (Steel and Torrie, Principles and procedures of statistics, McGraw-Hill, New York, 1960). Analysis of plant measurements and regression analysis of X. bakeri on pH were carried out on the original data.

Table 1 shows that none of the fertilizers completely eliminated X. bakeri, but some interesting plant and nematode responses were associated with the complete-fertilizer and ammonium nitrate (N) treatments. These fertilizers gave significant increases in posttreatment height and stem dry weight. Only the ammonium nitrate treatment. however, was equal to the check for root and stem weight.

The failure of the complete fertilizer to promote better root growth than the ammonium nitrate was probably caused by nutrient toxicity and not by nematode damage, since both the completefertilizer and the ammonium nitrate treatments had similar numbers of nematodes. The complete-fertilizer requirement was estimated from the background nutrient levels of the soil and a meager knowledge of the ionic tolerance of Douglas-fir. During the first 2 mo after fertilizer application, the seedlings receiving the complete-fertilizer treatment grew well but thereafter became chlorotic and began shedding needles from the lower part of the stem; the plants were spindly and in poor form. Also, high rates of nitrogen and phosphorus would reduce ectomycorrhizal development.

Nematodes apart from X. bakeri (i.e. "other nematodes") were not significantly affected by any fertilizer treatment. The slightly higher numbers in pots receiving nitrogen (check, N, and complete) resulted from nonparasitic species. Many such species benefit from an increase in microfloral and macrofloral growth following fertilization (Marshall, Commonw. Bur. Soils, Spec. Publ. 3, 1977). The nonparasitic nematodes in the check, which was heat-treated, presumably came in with soil from the styroplug. For X. bakeri, expressed as numbers per 100g soil dry weight, the differences in fertilizer treatments were not clearly separated. However, both complete fertilizer and ammonium nitrate produced significant reduction in X. bakeri when expressed as numbers per g root dry weight.

Significant increases in posttreatment height and stem dry weight in the complete and ammonium nitrate treatments were associated with reduced numbers of X. bakeri. Ammonium, ammonia, and nitrates, all of which are breakdown products of ammonium nitrate fertilizer, are inimical to some species of plant-parasitic nematodes (Baker et. al., Nematologica 17:377-385, 1971; Walker, J. Nematol. 3:43-49, 1971). Phytonematodes can also be reduced by changes in osmotic pressure effected by fertilizers (Juhl, Tidsskr. Planteavl 79:609-624, 1975).

The fact that the reduction in total numbers of X. bakeri was not significantly correlated with pH (r = 0.518) supported the idea that pH is not a major factor affecting nematode ecology. Douglas-fir grows well at pH 4-6.

Fertilizers are being tested for their potential in reducing phytonematodes in agricultural crops, and the results from the present experiment suggest that nitrogenous fertilizers at 448 kg N/ha could be used to improve the status of X. bakeri-infected Douglas-fir seedlings without leaving higher populations in the soil for the next crop. Further research is required to determine the types of nitrogenous fertilizers that could be most useful and the optimal rates for production of seedlings suitable for outplanting.-V.G. Marshall, Pacific Forest Research Centre, Victoria, B.C.

#### **ERRATUM**

A "plus or minus" sign (±) appeared in error on line 7 of paragraph 1 of "Stand Volume Estimates for Natural Jack Pine Stands," which was carried on page 7 of the March-April issue (vol. 36, no.2). The sentence should have read as follows:

The HS expression (dominant height/ V spacing) was tested with data from published yield tables for several Canadian species.

#### **RECENT PUBLICATIONS-MAY-JUNE 1980**

- 3 Bonga, J.M. and A.H. McInnis. 1979. Sterilization of Gelman metricel GA8 membrane filters with isopropanol. TCA Man. 5(2):1037-1038.
- 7 Hart, John H., and D.M. Shrimpton. 1979. Role of stilbenes in resistance of wood to decay. Phytopathology 69:1138-1143.
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