Environment Canada
Canadian Forestry Service

Environnement Canada
Service canadien des forêts

ISSN 0228-9989

canadian forestry service research notes

Vol. 4, No. 3, July-September, 1984

Canada

canadian forestry service
research notes

A selection of notes on current research conducted by the Canadian Forestry Service and published under the authority of the Minister, Environment Canada. A French edition is published under the title of Revue de recherches du Service canadien des forêts. *It is the editorial policy of this publication that the names of members of review boards considering manuscripts for publication shall not be disclosed. Manuscripts submitted for review will be accepted for consideration only on this basis.*

FOREST DESCRIPTION AND MENSURATION

A Microcomputer-Based Data Reader and Editor for the DIGIMIC Tree Ring Measuring System

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There is considerable interest in the Canadian-made DIGIMICROMETER (DIGIMIC), a tree ring measuring instrument developed in New Brunswick[1]. This instrument is designed to measure annual tree rings from cross sections of tree stems (discs) or from increment borings. The sample is placed on a motorized table and the observer records the occurrence of rings as they pass under the cross-hairs of a binocular microscope. A microprocessor unit temporarily stores the distances between rings and transfers them to a cassette recorder. Because storage and manipulation of data on a tape recorder are cumbersome and slow, direct connection of the DIGIMIC to a computer is highly desirable. Interfacing, though, requires the development of specialized software.

The DIGIMIC has been successfully interfaced to microcomputers in two cases. Jordan and Ballance (For. Chron. 59: 21-25, 1983) interfaced their DIGIMIC to a Radio Shack TRS-80 personal computer. Fayle et al. (For. Chron. 59: 291-293, 1983) connected their instrument to an Apple II Plus microcomputer and developed programs to read ring data and to produce graphic displays for comparison and editing.

Mainframe computer programs to produce tabular or graphic summaries of tree ring data gathered using the DIGIMIC have been reported by Timmer and Verch (For. Chron. 59: 298-303, 1983) and, for data gathered using conventional means, by Herman et al. (USDA, For. Serv. Res. Pap. PNW 194, 1975).

This report describes a powerful editor computer program written on an Apple II Plus microcomputer to read and manipulate ring data obtained with the DIGIMIC. This program would be of value to anyone using an APPLE computer to read DIGIMIC data.

The system consists of:

1) A 48K Apple II Plus computer.
2) Two diskette drives connected to the Apple slot 6 (Only one drive is essential).
3) One RS232 interface card in slot 3 with baud rate set to 300, connected to the DIGIMIC.
4) One 32K RAM memory card (MACROMEM) in slot 0 (not essential).
5) One printer (Epson MX-100) and interface in slot 1.
6) Television monitor.

The computer program, written in Applesoft BASIC, is divided into three sections. The first reads DIGIMIC data for each disc radius and creates data files on one diskette drive. The second section edits and prints out the tree ring data. The third section creates a data file that can be sent to a mainframe computer for further processing. Options allow the user to catalog or initialize diskettes, to load, rename or delete files, to average two or more disc radii, to list the data on the monitor or send it to the printer or to mainframe computer, and to save the data onto a diskette. The main menu is shown below:

MAIN MENU

COMMAND	NUMBER
READ DIGIMIC DATA	1
EDIT/PRINT	2
RENAME FILES	3
CATALOG	4
INITIALIZE DISKETTES	5
DELETE A FILE	6
LIST A TREE FILE	7
ADDOX	8
LOAD RAW RADII	9
AVERAGE RADII	10
QUIT	11

Choose a Number (1-11)

[1]Holman Electronic Controls Ltd., Box 1025, Fredericton, N.B. E3B 5G2

The READ option is selected after measurement of a radius has been completed in the DIGIMIC. This command creates a file called RADIUS FILE which contains the raw ring width data that is stored in diskette. A TREE FILE is also created, in diskette, that contains the average of all radii in each disc for an entire tree. After a TREE FILE is created, the raw data files are archived and subsequent analysis continues on the TREE FILE.

The EDIT/PRINT option operates on the raw data files. Upon selection of this option, the following menu appears:

EDIT/PRINT MENU

COMMAND	NUMBER
RING COUNT	1
INSERT A RING	2
DELETE A RING	3
MERGE 2 RINGS	4
SPLIT A RING	5
LIST RAW DATA	6
FILE DATA	7

Choose a number (1-7)
Press Return to exit

By selecting the appropriate option, the user can obtain the number of rings in one or all radii in a disc, display the data, or correct measurement or dating errors by inserting, deleting, merging or splitting annual ring measurements from a radius file. After editing is completed, data are automatically averaged and the TREE FILE updated. This command is an interactive editor permitting the modification of the data immediately after reading the measurements from the DIGIMIC while the wood sample is still under the microscope. Raw files that were previously saved on diskette can also be edited at this time.

The ADDOX function in the main menu prepares a data file for transmission to a mainframe computer. Transmission from the Apple to the mainframe is accomplished using the computer program VISITERM[2]. In the mainframe, data is then reformatted to be compatible with existing programs for analyzing data obtained with an older ADDO-X tree ring measuring instrument.

This program can deal with a maximum of 400 rings in up to 4 radii per disc. Information from up to 20 discs can be stored in one TREE FILE.

[2]VISICORP Ltd., 2895 Zanker Rd., San Jose, CA. 95134.

Table 1

DIGIMIC/APPLE measuring, processing time, and productivity to measure data from 11 disks in each of two Douglas-fir trees

Tree	Age (Years)	Total No. Rings[1]	Measuring Time[2] (min)	Processing Time[3] (min)	Total Time (min)	Productivity (min/Ring)
1	66	798	61	52	113	0.14
2	70	654	50	54	104	0.16
TOTAL	—	1452	111	106	217	0.15

[1] Sum of all rings along 2 radii in each of 11 tree discs.

[2] Time taken from the moment the sample disc is aligned on the measuring table under binocular microscope until the last ring in radii is measured.

[3] Includes saving and averaging raw data, saving averaged data and printing raw and averaged files.

Programs in Applesoft BASIC are being developed for graphic displays of ring increment sequences for any radius or disc in a TREE FILE as well as for the calculation of annual volume increment from stem analysis and mean annual increment calculations.

Productivity of the system was assessed by determining the time required per ring to measure, process, and save (raw and average files) ring data for two radii in each of 11 discs obtained from two Douglas-fir trees. Stump age for the trees were 66 and 70 years. Total time spent from the time the first sample disc from the first tree was aligned on the measuring table, under the binocular microscope, until all rings for all discs in both trees were measured, and files created and saved, was 217 min (Table 1). Since the total number of rings measured was 1452, the system productivity was calculated as 0.15 min per ring.

Software for this program is available free of charge from the Pacific Forest Research Centre, in diskette form, with a 32-page instruction manual if a blank diskette accompanies the request.

INSECT AND DISEASE SURVEYS

Method for Evaluating Defoliation of Grey Birch Caused by Birch Leafminers

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Several species of leafminers can attack grey birch (*Betula populifolia* Marsh.). The lesser birch leafminer (*Fenusa pusilla* [Lep.]) is by far the most common species. Three other species of leafminers also attack this host – the ambermarked birch leafminer (*Profenusa thomsoni* [Konow]), *the early birch leaf edgeminer*

(*Messa nana* [Klug]) and the greater birch leafminer (*Heterarthrus nemoratus* [Fall.]). During severe infestations, the normal development of the trees attacked may be seriously affected. Also, because these trees are generally weakened, they are more susceptible to attack by other insects and diseases (Guèvremont, Can. For. Serv., Dept. Environ., Laurentian Forest Research Cent., Feuil. Inf. LFRC 13, 1973). This report deals with a study conducted in 1983 in the Province of Quebec to develop a method for evaluating the percentage of grey birch leaves mined in a stand attacked by birch leafminers.

Two methods for evaluating the percentage of mined leaves were tested: (1) ocular estimating from the ground using binoculars, (2) estimating by counting the mines on leaves removed by pole pruning shears. Twenty-five young stands attacked to varying degrees by leafminers were studied. They were located between Quebec City, Trois-Rivières, and Sherbrooke. The stands selected were deciduous (22) or mixed (3); grey birch made up 20 to 90% of the total volume of the stands. In all cases, the density of stands was low (<60%). Five grey birch less than 10 m high were chosen at random from each stand.

Using the first method, a visual evaluation of the percentage of mined leaves (classes of 10%) was done independently by two technicians on the upper, middle, and lower thirds of each selected stem. Then the two technicians consulted one another to reach an agreement on their estimates of the percentage of leaves mined. When a consensus was not reached, the ocular estimates were repeated. In the second method, branches approximately one metre long were removed from the center of the upper, middle, and lower thirds of each stem that had already been the subject of the ocular estimates. The percentage of mined leaves was obtained by on-site counting of 50 leaves taken from each branch removed. In this way, 150 leaves per tree, 750 leaves per stand, were studied. The mined leaves were then taken to the laboratory to identify the leafminers.

The standard error in the percentage of mined leaves obtained by counting was calculated, assuming that we were dealing with a cluster sampling for proportion (Cochran, *Sampling Techniques*, Wiley and Sons, Toronto, Ont. 1977):

$$\sigma p = \sqrt{\frac{\sum_{i}^{5}\sum_{j}^{3}(p_{ij} - P)^2}{n(n-1)}}$$

σ_P = standard error in the percentage of mined leaves in the plot obtained by counting

i = 1, 2, 3, 4, and 5 corresponding to each tree in the plot

Table 1
Percentage of mined leaves and confidence interval obtained by counting

Plot	P*	D(90%)**	D(95%)***
19-037	7	± 4	± 5
19-038	10	± 4	± 5
19-039	4	± 3	± 4
19-040	27	± 8	± 9
19-041	36	±10	±13
17-066	12	± 6	± 7
17-068	6	± 2	± 3
17-070	7	± 3	± 4
17-072	5	± 2	± 2
17-074	2	± 1	± 1
17-076	6	± 2	± 3
18-100	11	± 5	± 6
18-101	59	± 7	± 8
18-102	41	± 9	±11
18-103	13	± 6	± 7
18-104	43	± 7	± 9
18-105	34	± 7	± 9
18-106	17	± 3	± 4
18-107	53	± 8	± 9
18-108	8	± 3	± 4
18-109	30	± 4	± 5
18-110	36	± 5	± 6
18-111	20	± 6	± 8
18-112	32	± 5	± 6
18-113	16	± 4	± 5

* Percentage of mined leaves in the plot
** Confidence interval of 90%
*** Confidence interval of 95%

j = 1, 2, and 3 corresponding to each third of the top (lower, middle, upper)

n = 15 representing the number of clusters considered in calculating P

$P = \frac{1}{n}\sum_{i}^{5}\sum_{j}^{3} p_{ij}$ representing the percentage of mined leaves obtained by counting for each plot

P_{ij} = percentage of mined leaves in each third j of the top for each of the i trees in the plot

The confidence interval of P (90 and 95% was calculated as follows:

$P \pm (t\alpha/2, n\text{-}1)\ ^{\sigma}P$ where $t\alpha/2, n\text{-}1$ is the critical value of the Student distribution.

Table 1 shows the percentage of mined leaves obtained by counting as well as their respective confidence intervals for the 25 sample plots. It should be noted that the confidence interval of P is always less than or equal to 10% when the probability threshold is set at 90%. In other words, the sampling of 15 clusters of 50 leaves gives an accuracy of about ± 10% at 90% probability in evaluating the percentage of mined leaves in a stand. Table 2 summarizes the results obtained by means of the ocular estimate and the counting estimate. The percentages obtained by counting are grouped by classes of 10%. A T test for paired data shows that there was no significant difference ($P < 0.05$) between the

Table 2
Percentage of mined leaves obtained through binocular estimation and counting

Plot	Binocular estimation*	Counting*	Difference
19-037	10	10	0
19-038	20	10	10
19-039	10	10	0
19-040	30	30	0
19-041	40	40	0
17-066	20	20	0
17-068	20	10	10
17-070	10	10	0
17-072	10	10	0
17-074	10	10	0
17-076	10	10	0
18-100	10	20	−10
18-101	70	60	10
18-102	50	50	0
18-103	10	20	−10
18-104	50	50	0
18-105	50	40	10
18-106	20	20	0
18-107	60	60	0
18-108	10	10	0
18-109	30	30	0
18-110	50	40	10
18-111	20	30	−10
18-112	40	40	0
18-113	20	20	0

* Classes of 10%

Table 3
Percentage of mined leaves obtained by counting, according to the levels of the stems versus the percentage of the entire plot

Plot	Upper third	Middle third	Lower third	Entire plot
19-037	10	7	5	7
19-038	16	9	6	10
19-039	6	3	3	4
19-040	31	24	26	27
19-041	44	36	28	36
17-066	24	6	4	12
17-068	8	6	6	6
17-070	8	6	6	7
17-072	6	8	2	5
17-074	2	2	1	2
17-076	5	6	6	6
18-100	14	12	8	11
18-101	65	48	64	59
18-102	50	38	36	41
18-103	15	10	14	13
18-104	46	48	36	43
18-105	49	21	32	34
18-106	17	15	18	17
18-107	52	53	51	53
18-108	6	9	10	8
18-109	30	26	33	30
18-110	40	34	34	36
18-111	26	17	16	20
18-112	30	37	30	32
18-113	19	14	14	16

two methods used. The analysis of the variance of the results obtained by counting was done using a split-plot design (Kirk, pages 245-318 *in Experimental Design: Procedures for Behavioral Sciences,* Wadsworth Publishing Company, Belmont, Calif., 1968). This analysis showed that the percentage of mined leaves differed significantly ($P > 0.05$) between the upper, middle, and lower thirds of the stems. The Tukey test showed that the percentage of mined leaves was greater in the upper third than in the middle and lower thirds; these last two thirds were affected to the same extent (Table 3).

The laboratory study showed that 99.3% of the mines discovered were caused by the lesser birch leafminer. The early birch leaf edgeminer, the amber-marked birch leafminer, and the birch casebearer (*Coleophora serratella* [L.]) caused 0.5, 0.1, and 0.1% of the other mines respectively.

This study has shown that sampling of 15 clusters of 50 leaves (750 leaves) taken from the upper, middle, and lower thirds of 5 grey birch gave, for a stand, an estimate of the percentage of mined leaves with an accuracy better than or equal to 10% at 90% probability. Furthermore, the results obtained by counting were comparable to the ones obtained with binoculars.

It can be concluded that ocular estimating, using binoculars, of the upper, middle, and lower thirds of five stems chosen at random from a stand gives, for that stand, an estimate of the mined leaves that has an accuracy of ± 10% at 90% probability.

INSECT PATHOLOGY

Observations of a Microsporidian Parasite in the White Pine Weevil *Pissodes strobi* (Peck) (Coleoptera: Curculionidae)

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The white pine weevil *Pissodes strobi* (Peck) is a common pest of white pine, but all pines and spruces may be attacked. Trees are not killed, but serious deformity may occur due to destruction of the terminal leader. The weevil was first examined at the Forest Pest Management Institute in 1970 for microorganisms that might be used as control agents, and a microsporidium was isolated from both adults and larvae. Streett et al. (Chesapeak Sci. 16:32-38, 1975) reported a microsporidian parasite of the white pine weevil and tentatively assigned it to the genus *Nosema*. This report presents

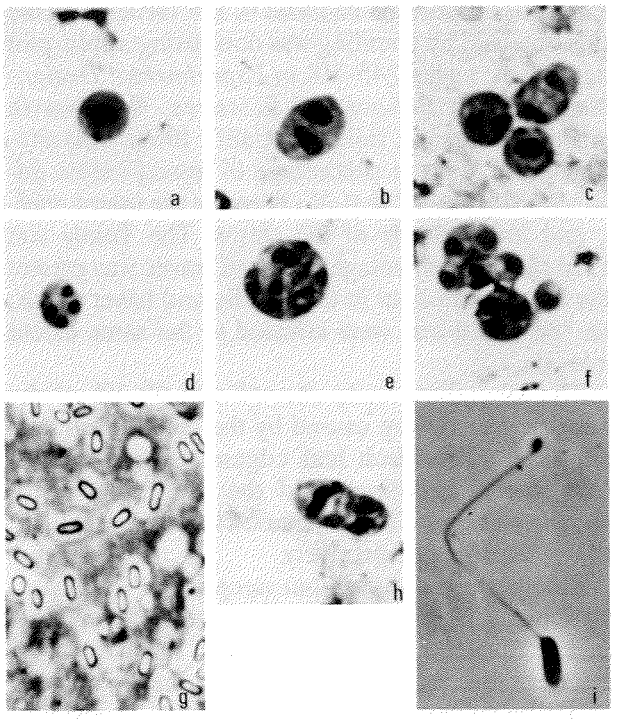

Figure 1. Various stages in the life cycle of a microsporidian parasite of the white pine weevil. All are Giemsa-stained, X3000 except h which is phase contrast, X1700. a – Uninucleate meront (maybe diplokaryon); b – binucleate meront; c – group of meronts, one indicating a diplokaryon; d – quadrinucleate meront; e – multinucleated meront, f – meront indicating budding; g – mature spores in gut tissue; h – sporont, cleaving into two sporoblasts; i – spore with extruded polar filament.

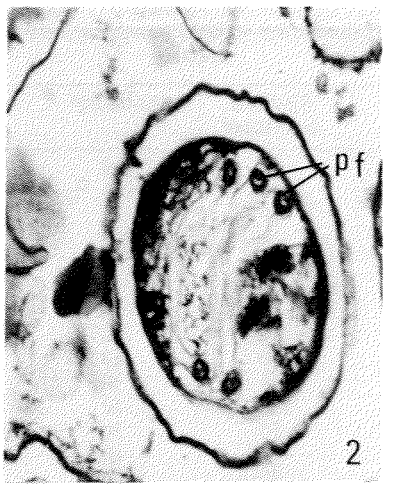

Figure 2. Developing polar filaments (pf) within the sporoblast. X22,500.

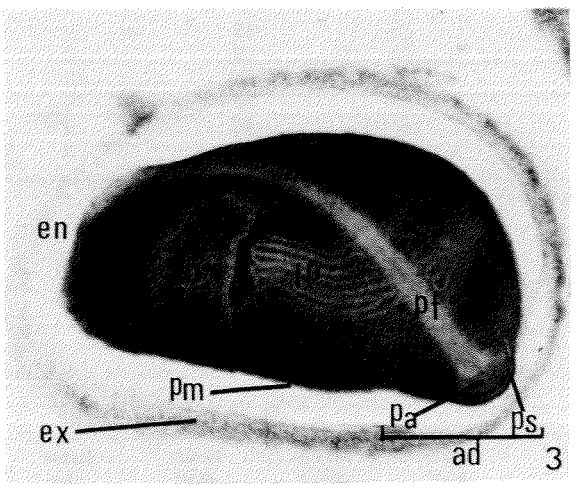

Figure 3. Mature spore showing the plasma membrane (pm), thick cell wall consisting of endospore (en) and exospore (ex). The anchoring disc (ad) is composed of the polar aperture (pa) and the polar sac (ps) which narrows to the polar filament (pf). Also shown in the spore is the lamellar polarplast (lp) and the nucleus (n). X 22,500.

further observations on the life stages of a microsporidian parasite as well as some indication of its levels of occurrence in nature .

Adult weevils were collected in the spring of a year, crushed and examined for the presence of microsporidia with phase contrast optics. Later in the summer, larvae were removed from leaders of Scots pine and examined for microsporidian infection.

Tissue smears of the infected insects were fixed in absolute methanol and stained in Giemsa solution for light microscope examination of parasite life stages. A small drop of a concentrated spore suspension was placed in 10% methocel (methyl cellulose) on a microscope slide and covered with a cover glass. This allowed measurement of spores without Brownian movement. Polar filaments were extruded by applying mechanical pressure. All measurements were made using an ocular micrometer on phase or bright-field microscope optics. Tissues were prepared for ultrastructural electron microscope examination using a method previously reported (Percy et al., J. Invert. Pathol. 39:49-59, 1982).

The life stages of the microsporidium described here are based on observations of the parasite in Giemsa-stained tissue smears and electron microscope photographs. The main site of infection was the midgut cells of the host. Figure 1 depicts various stages in the life cycle of this parasite. Meronts containing various numbers of nuclei were present and in some cases appeared in a diplokaryon arrangement. Confirmation of the diplokaryon by electron microscopy was not successful because we were unable to obtain sections with the vegetative stages present. It appears that the parasite may multiply in several ways – binary fission of the binucleate meront, fragmentation of multinucleate stages, and as suggested by Figure 1, budding. In most cases the sporont gives rise to binucleate sporoblasts (Fig. 1, g) and eventually to two binucleate spores.

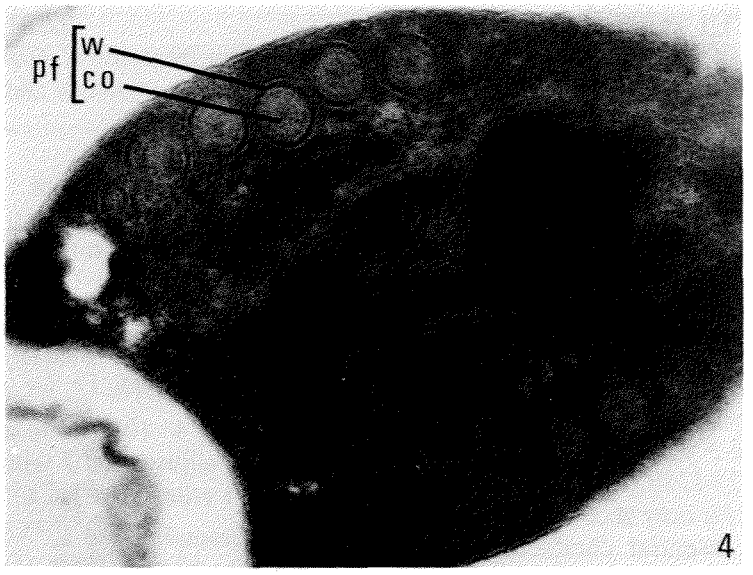

Figure 4. Mature spore with cross section of the polar filament (pf) showing the core (co) and filament wall (w). X30,000.

Fresh spores (Fig. 1, h) measured 3.4 ± 0.8 × 1.6 ± 0.3 μm; stained spores measured 2.9 ± 0.5 × 1.5 ± 0.3 μm. The average length of the extruded polar filament (Fig. 1, i) was 32.0 μm. All measurements were based on the examination of at least 50 specimens.

Sporoblasts depicted in electron photomicrographs were observed to lie singly in the host cytoplasm and did not appear to be surrounded by a pansporoblastic membrane. The developing polar filament was visible in the sporoblast (Fig. 2), and a cross-section (Fig. 4) shows the core and filament wall. Many spores contained up to 9 polar filament coils. Each spore is enclosed in a thick wall consisting of the endospore and exospore. (Fig. 3). Within the wall the spore is limited by a plasma membrane. A large portion of the spore is occupied by the extrusion apparatus, made up of the apical anchoring disc, which consists of the polar aperture and the polar sac. The polar sac narrows to the polar filament. The laminar polarplast extending from the lateral edge of the anchoring disc is also shown in Figure 3.

Larvae and adults of the white pine weevil were collected over a 10-year period in areas near Sault Ste. Marie, Ontario. (Data supplied by J.M. Burke [retired] of the Insect Disease Survey at the Forest Pest Management Institute). During this time an average of 15.8% of the 1,382 larvae examined were infected with microsporidia. Adults from the same area were examined for 8 years out of this 10-year period, and an average of 9.0% of the 524 adults examined were infected with the parasite.

This microsporidium is similar to one reported from the white pine weevil by Streett et al. (Chesapeak Sci. 16:32-38, 1975). They reported spores of two size groups; microspores that were ovoidal and 3.5-4.2 × 1.8-2.2 μm, and ovoidal or reniform macrospores, 5.0-6.0 × 1.8-2.5 μm. This finding led them to suggest that two species of microsporidia may be present. They also reported that the average length of the polar filament was 80 μm. The parasite that was observed had ovoidal microspores of similar size to those reported by the above authors. A microsporidium, *Nosema gasti*, with similar spore sizes, 4.3 ± 0.3 × 2.3 ± 0.2 μm has also been reported from boll weevil, *Anthonumus grandis* (McLaughlin, J. Protozool. 16:84-92, 1969). Based on the photographs of McLaughlin (J. Protozool. 16:84-92, 1969), many of the life stages are similar to the microsporidium described here. A *Nosema* species has been reported from the alfalfa weevil *Hypera postica* (Maddox and Luckmann, J. Invert. Pathol. 8:543-544, 1966, Drea et al., J. Invert. Pathol. 13:303-304, 1969), however, not enough information is available to make a comparison, but spore size and polar filament length are not the same as the microsporidium from the white pine weevil. Based on the forgoing observations, the microsporidium of the white pine weevil probably belongs to the genus *Nosema*, although assignment to this genus should await electron microscope studies.

SILVICULTURE

Cambial Activity in Foliated *Abies balsamea* (L.) Mill. Stem Segments Cultured *in vitro*

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Cambial activity can be induced in dormant and debudded 1-year-old foliated cuttings of balsam fir by placing them in favorable environmental conditions and supplying water and indol-3-ylacetic acid, (Little and Bonga, Can. J. Bot. 52: 1723-1730, 1974). However, the cambium produces xylem for only about 6 weeks, presumably because some essential chemical factors become depleted. Characterization of these factors requires a procedure for culturing the segments on a chemically-defined nutrient medium. Brown and Wodzicki (For. Sci. 15: 26-29, 1969) described a procedure which involves feeding of liquid medium through sterile latex tubing aseptically fitted over the ends of each segment. However, their procedure is tedious to assemble and maintain, and cannot readily be used in controlled environment cabinets with limited space; moreover, they reported that it routinely resulted in as much as 50% contamination. The purpose of the present work was to determine the feasibility of an alternative procedure, involving the growing of surface sterilized, relatively short segments on solid medium in test tubes. This approach would have the advantage of rapidly producing cultures of large numbers of segments in a minimum growing space.

One-year-old shoots were collected from the upper crown of field-grown trees during the September-May period of cambial dormancy. They were used immediately, or after storage at 4°C in a plastic bag for periods up to several weeks. Surface sterilization was attempted in the initial experiments using 0.6 – 6.0% sodium hypochlorite (NaOCl) for 10 min to 24 h, with or without subsequent treatment with 70% ethanol + hydrochloric acid (1 drop per 100 mL ethanol) for 2 to 4 min or 0.1% mercuric chloride ($HgCl_2$) for 10 to 60 sec. Other sterilizing agents tested in subsequent experiments included hydrogen peroxide, boric acid, polyvinylpyrrolidone-iodine, benzethonium chloride, hexamethonium chloride, benzalkonium chloride, hexachlorophene, calcium hypochlorite, three antibiotics (chloramphenicol, nystatin, grisovin), the mercury containing compound thimerosal, and the germicide, wavicide.

The shoots were submerged in the sterilizing solution at room temperature in beakers placed on a shaker or stirrer. Vacuum, ultrasound, and detergents were also tested in an attempt to increase penetration of the sterilizing solution. At the end of the sterilizing treatment, the shoots were washed 2 to 4 times in sterile distilled water. After removing both ends, each shoot was subdivided into foliated segments, about 2.5 cm long. These segments were placed in nutrient medium (Romberger, Varnell, and Tabor, USDA For. Serv. Tech. Bull. 1409, 1970) to which 1 mg indol-3-ylbutyric acid L^{-1} and either 7 g Difco bacto-agar L^{-1} or 1.5 g Kelco gelrite L^{-1} was added. The segment's morphological apical end was put into the medium to facilitate auxin uptake and translocation (Little, Can. J. Bot. 53: 3041-3050, 1975). In some experiments, thimerosal, chloramphenicol, nystatin or grisovin were incorporated into the medium. The cultures were kept either at 21°C in 50 $\mu Em^{-2}s^{-1}$ of GRO-LUX fluorescent light for 16 h daily or at 20-25°C in indirect outdoor lighting for 10 to 14 h daily. Contamination was monitored at about weekly intervals during an approximate 12-week culture period. At the end of this period, cambial activity was assessed in transverse handcut sections made at the midpoint of noncontaminated viable segments and stained in phloroglucinol-HCl (Little and Bonga, *op. cit.*).

Cambial reactivation occurred, typically resulting in a wide band of new tracheids around the circumference and along the segment length, except in the apical portion embedded in the agar. In addition, callus

TABLE 1
Sterilizing treatments resulting in noncontaminated segments with an active cambium

Treatment	Number of segments*
6% NaOCl for 60 min, 70% ethanol + HCl⁺ for 2 min	2
6% NaOCl + 2 g sodium bicarbonate L^{-1} + (pH adjusted to 7 with HCl) for 10 min, 70% ethanol + HCl⁺ for 2 min	3
6% NaOCl + 10 g boric acid in 10 mL H_2O (pH adjusted to 7 with HCl) for 10 min, 70% ethanol + HCl⁺ for 2 min	4
1 g chloramphenicol L^{-1} for 16 h, 6% NaOCl for 10 min, 0.1% $HgCl_2$ for 1 min	2
6% NaOCl for 10 min, 0.1% $HgCL_2$ for 1 min, 0.1 mg thimerosal L^{-1} incorporated in medium	4

*20 segments per treatment.
⁺One drop HCl per 100 mL ethanol.

frequently was produced from both cut surfaces, although most obviously at the apical end. The segments remained foliated, the needles retaining their original green, healthy appearance.

Although a comprehensive sample of sterilizing agents and conditions was tested (details available upon request), only five treatments produced segments that were both sterile and viable at the end of the culture period; none of these was more than 20% effective (Table 1).

The appearance of contaminants occasionally was delayed for as long as 4 weeks. This suggests that surface sterilization actually was achieved and that subsurface microorganisms caused the subsequent contamination, which is not unexpected because internal microbes occur in needles (Bernstein and Carroll, Can. J. Bot. 55: 644-653, 1977). A method to control this internal contamination must be found before this *in vitro* procedure can be used routinely as a cambial activity assay.

TABLE 1
Species planted, origin, survival after 2 and 8 growing seasons, and height after 8 and 10 years from seed.*

Species planted	Origin	Percent survival		Average total height (m)	
		2	8	8	10
L. laricina	Petawawa N.F.I., Ontario	97.8a	90.0a	1.21c	1.75bc
L. eurolepis	Newton Seed Orchard, Scotland	77.8b	66.7b	1.56a	2.21a
L. eurolepis	Mabie Seed Orchard, Scotland	84.4b	78.9b	1.49ab	1.96ab
L. kaempferi	High Meadow For., England	63.3c	43.3c	1.32bc	1.71bcd
L. sibirica(1)	Krasnojarsk, USSR	50.0d	4.4d	.92d	1.50cd
L. sibirica(2)	Krasnojarsk, USSR	68.9bc	3.3d	.62e	1.07e

*Means followed by the same letter are not significantly different from each other at probability level 0.95.

Field Test of Exotic Larches in Western Newfoundland

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Indications of success in planting trials using Japanese larch (*Larix kaempferi* (Lam.) Carr.) and European larch (*L. decidua* Mill.) prompted the establishment of a species trial of other larch species thought to be equally well adapted to local conditions. Local tamarack (*L. laricina* (Du Roi) K. Koch) was tested, as was Japanese larch and two seed sources of Dunkeld larch (*L. eurolepis* Henry) a hybrid between Japanese and European larch. Siberian larch (*L. sibirica* Ledeb.) was also included in the test because it was believed to be well adapted to boreal forest conditions.

The test area is in western Newfoundland at 48°40′N, 58°30′W, and at an elevation of 365 m. The site is exposed to the prevailing onshore westerly winds. The forests are part of Forest Section B.28(b) in an area characterized by balsam fir (*Abies balsamae* (L.) Mill.) stands, with white spruce (*Picea glauca* (Moench) Voss) and white birch (*Betula papyrifera* Marsh.) as minor species (Rowe, J.S. 1972. Forest Regions of Canada, Can. For. Serv. Publ. 1300. 172 p.).

The site was cleared of slash in October 1975 and planted on 27-28 April 1976. The tamarack and one seedlot of Siberian larch (2) were aged 2 + 1 when planted; the others were aged 2 + 2 (Table 1). The experimental design consists of a 10-replicated, randomized complete block with 9-tree square plots and trees planted at 2 × 2 metre-spacing. The tamarack seedlings were about 35-40 cm tall when planted; the other species were 15-25 cm tall. Both sources of Siberian larch had already flushed by the time they were planted.

Survival in the first 8 years after planting was best for tamarack and hybrid larch and poorest for Siberian larch, which failed almost completely, and Japanese larch, of which less than half the seedlings survived. The planted stock was under severe competition from the dense vegetation on the site, which consisted of natural regeneration of balsam fir and white birch; shrubs and herbs, including *Sorbus, Ribes, Rubus, Aster, Epilobium augustifolium,* and dense patches of *Pteridium aquilinum*. Based on survival, only the tamarack and hybrids are suitable for planting. The Siberian larch originated from a boreal environment (ca. 55°N), but it has not survived at this site. This is because it flushes early and is therefore subject to spring frosts which are common in the area.

At 8 and 10 years from seed, the tallest species on average was hybrid larch, followed by Japanese larch which was about 15 percent shorter. The tamarack appears to be gradually outgrowing the Japanese larch, as it is slightly taller after the 10-year remeasurement. The results suggest that both the native tamarack and hybrid larch should be considered for reforestation of sites as productive and exposed as the test site. The tamarack reached a height of only 1.75 m, 10 years from seed, whereas on a low elevation site at the same age, it was 2.18 m tall (Hall, J. Peter. 1973. Survival and growth of exotic tree species in Newfoundland. Env. Can. For. Serv. Inform. Rep. N-X-85. 23 p.). The difference probably reflects the differences in site quality. At the lower elevation, tamarack was outgrown by a wide margin by Japanese larch at age 15 but, at the higher elevation, the Japanese larch did comparatively poorly. This illustrates some of the difficulties associated with the introduction of exotic species. They must be field tested on a variety of sites before conclusions can be drawn as to their suitability for reforestation.

Preliminary Investigation of the Field Performance of Black Spruce Rooted Cuttings in Boreal Ontario

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Approximately 450 000 black spruce (*Picea mariana* [Mill.] B.S.P.) rooted cuttings were planted in reforestation projects across Ontario in 1983. Currently (1984), two provincial forest stations, Swastika (48°N, 80°W) and Orono (44°N, 78°N), are producing rooted cuttings on an operational scale. Cuttings offer several advantages over traditional seeded planting stock: more seedlings from fewer seed, faster improvements in the genetic quality of stock, and the potential for avoiding diseases and insects which attack only juvenile trees (Brix and van den Driessche, B.C. For. Serv./Can. For. Serv. Joint. Rep. No. 6, 1977). In July 1979, 800 black spruce cuttings and 800 seeded paperpots were outplanted in northeastern Ontario to assess field performance. This note compares fifth-year survival, growth, and form of these two stock types.

Cuttings and seeded paperpots were planted together on two sites in Webster (49°19′ to 49°27′N, 80°28′ to 81°40′W) and Raven (49°10′ to 49°19′N, 80°17′ to 80°29′W) townships. Four hundred trees of each stock type were planted at each location for a total of 1600 trees. The Webster Township planting site is flat, moist to moderately wet with a 20- to 60-cm-deep organic mat covering a silt-loam parent material. The Raven Township planting is on a gently sloping (2-3%) fresh site with a 16- to 20-cm-deep organic mat covering silty-clay/loam parent material. Both sites were clearcut from 1977 to 1978 and winter shearbladed in the spring of 1979.

The rooted cuttings planted in these trials were produced at Orono Forest Station and their production has been previously described (Armson et al., J. For. 78:341-343, 1980). The cuttings were taken in late March, 1979, from stock grown from general collection seed, Site Region 3E (Hills, Ont. Dep. Lands For., Maple, Ont., 1959), and were rooted in 308 Japanese paperpots. The seeded 308 Japanese paperpots had been over-wintered, having been sown in a greenhouse at Swastika Forest Station in late June, 1978.

Morphological characteristics of the rooted cuttings and seeded paperpots at outplanting are given in Table 1. The cuttings were grown to test the feasibility of producing cuttings on an operational scale and were not originally intended for field outplanting. The stock was

TABLE 1

Morphological characteristics of rooted cuttings and seeded paperpots at time of planting.

Stock type	No. in sample	Total oven-dry weight (g)	Shoot length (cm)	Root collar diameter (mm)	Root area index* (cm^2)	Shoot:root ratio (oven-dry weight)
Rooted cuttings	25	0.3a (±0.1)†	15.5a (±2.2)	1.8a (±0.2	4.4a (±1.5)	6.0a (±1.7)
Seeded paperpots	25	0.2b (±0.1)	9.7b (±1.7)	1.4b (±0.2)	4.9a (±1.8)	3.9b (±1,1)

*After Morrison and Armson, For. Chron. 44:21-23, 1968.
†Data within parentheses are standard deviations.
Differing letters within each column indicate a significant difference at the P.05 level.
Data were subjected to analysis of variance.

TABLE 2

Mean survival, total height, fifth-year height increment, stem diameter, and form of rooted cuttings and seeded paperpots after five growing seasons (1983), Webster Township.

Stock type	No. of trees assessed	Survival (%)	Total height (cm)	Height increment (cm)	Stem diameter (mm)	Trees with single, straight, and upright main shoots (%)
Rooted cuttings	399	82b	36.4a	12.6a	6.6a	31.4a
Seeded paperpots	399	88a	36.1a	13.2a	7.6a	33.7a

Differing letters within each column indicate a significant difference at the P.05 level.
Data were subjected to analysis of variance.

TABLE 3
Mean survival (%), total height (cm), fifth-year height increment (cm), stem diameter (mm), and form (%) of rooted cuttings and seeded paperpots after five growing seasons (1982), Raven Township.

Stock type	No. of trees assessed	Survival	Total height	Height increment	Stem diameter	Trees with single, straight, and upright main shoots
Rooted cuttings	400	69a	35.9a	8.5a	6.4b	30.5a
Seeded paperpots	400	79a	35.2a	9.2a	7.5a	24.5a

Differing letters within each column indicate a significant difference at the P.05 level.
Data were subjected to analysis of variance.

not of particularly good quality; however, the oven-dry weight, shoot length, and root collar diameter of the seeded paperpots were significantly less than those of the cuttings at outplanting.

Five years after scarification and planting, vegetative competition was observed to be heavier on the Raven Township than on the Webster Township planting site.

In Webster Township, fifth-year survival of seeded paperpots (88%) was greater than that of cuttings (82%) (Table 2). However, neither fifth-year total height nor height increment differed significantly between cuttings and seeded paperpots. Likewise, there were no significant differences in stem diameter or main shoot form. In the Raven Township planting, none of fifth-year survival, total height, height increment, and main shoot form differed significantly between cuttings and seeded paperpots (Table 3). The only significant difference between these two stock types was in stem diameter, which was larger (7.5 mm) for the seeded paperpots than for the rooted cuttings (6.4 mm).

Although the results of these outplantings are preliminary, black spruce rooted cuttings on the sites tested performed as well as seeded paperpots, and show good potential for future use in Ontario.

The author is indebted to staff of the Cochrane District and to K.A. Armson of the Forest Resources Group, Ontario Ministry of Natural Resources, for establishing and plantings and for providing the planting stock.

The Seasonal Foliar Moisture Trend of Black Spruce at Kapuskasing, Ontario

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Variations in the moisture content of conifer foliage, if large enough, could affect the incidence of crown fires. A fair amount of evidence for a distinct seasonal trend in over-wintered (i.e., old) conifer foliage now exists, both in Canada and elsewhere. The main feature of this trend is a pronounced dip in the moisture content during a few weeks in late spring or early summer before the new foliage has developed. This note reports specifically on the foliar moisture content (FMC) of black spruce in the vicinity of Kapuskasing at 49.5°N in northeastern Ontario. (Moisture content here means percent moisture based on dry weight.)

References on foliar moisture trends in northern conifers include Molchanov (1957) for Scots pine (*Pinus sylvestris* L.) in the USSR, Dieterich (1963) for red pine (*Pinus resinosa* Ait.) in the Lake States, Van Wagner (1967) for several conifers at Petawawa, Russell and Turner (1975) for several conifers at various locations in British Columbia, and Fuglem and Murphy (1980) for lodgepole pine (*Pinus contorta* Dougl. var. *latifolia* Engelm.) in Alberta. All references agree on the presence of a pronounced spring dip in the FMC of old conifer foliage. In addition, Little (1970) showed that the mechanism is primarily a temporary buildup of starch within the needle cells rather than a reduction in absolute water content; thus it is primarily a physiological phenomenon, and not directly related to soil frost and current weather. Gary (1971) supplies additional argument for this conclusion. The physical effect of variations in FMC on crown fire behavior has been analysed by Van Wagner (1967, 1974, 1977), with the conclusion that crown fires should spread more easily during the period of the spring dip.

The purpose of this study was limited to producing information for use in the prediction of crown fire behavior, not to yield a complete account of foliar moisture dynamics in black spruce. The feature of primary interest, therefore, is the trend of average foliar moisture from the fire behavior viewpoint, in the afternoon when forest fire behavior is generally at its daily peak. Variations in FMC throughout the day, up and down crown length, from tree to tree, or with tree age and site were not addressed. Specific goals were i) to identify the presence of a spring dip in FMC if any; ii) to determine its magnitude; and iii) to determine its timing.

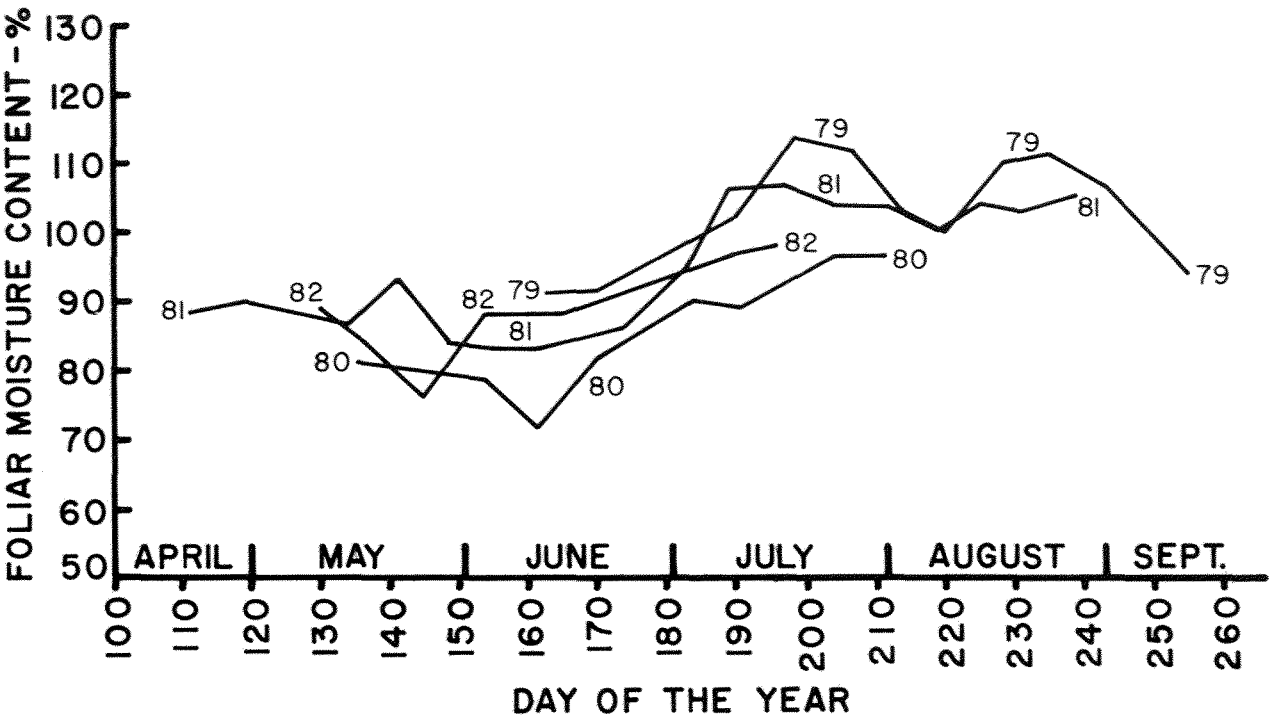

Figure 1. Annual trends in moisture content of old black spruce needles for four years at Kapuskasing, Ontario.

Black spruce foliage near Kapuskasing, Ontario, was sampled for moisture content (MC) on a dry-weight basis during the four years 1979-1982:

a) from natural stands, aged 70 to 90 yr, of height 12-13 m, on "black spruce-Sphagnum" sites of intermediate quality, stocked 50 to 80%,

b) from eight trees during each season, a different set of trees each season,

c) once a week in early afternoon avoiding rainy weather,

d) taking one sample each of old foliage (all years together) and new foliage (after flushing) from each tree,

e) from the middle third of the crown with long-handled shears. The needles were separated from the twigs, and oven-dried at 80-100°C with a precision of about ± 2% MC per sample. The ranges of sampling dates were

1979, June 12 to September 12
1980, May 16 to August 1
1981, April 22 to August 27
1982, May 10 to July 15

During the four years, samples were taken on 41 days. The average daily range in the eight individual old-foliage samples was 21.6% MC and the average standard error ± 2.7%. Special adjustments of the MC data were made on only two days, namely the first two sample days in 1979. On the first of these, two very high individual samples were judged faulty on grounds that they were more than two standard deviations higher than the mean. Also the range of the eight samples on that day was 2.6 times the average, a suspicious result. On the second day, 10 points was subtracted from the daily average on grounds that the samples were taken in early morning rather than in afternoon. Otherwise, occasional individual samples were spoiled, and seven weekly samples were missed within the data ranges shown above.

The principal analysis consisted of plotting the weekly FMCs over date in the form of trends. This was done in two ways for each class of foliage: 1) as a nest of four graphs, one for each year (Figs. 1 and 2); and 2) as a composite single graph made by averaging the annual data sampling date within 10-day intervals (Fig. 3). Because the annual date ranges were not of equal length, these composites are 4-yr averages only near the centre, and ultimately based on single years at the extremities.

In addition, the old-foliage annual trends were compared by linear correlations, carried out as follows. For each pair of years, the weekly mean FMC's nearest in date were plotted against each other and a least-squares best fit computed. The results are quoted below in terms of number of data pairs (n) and coefficient of

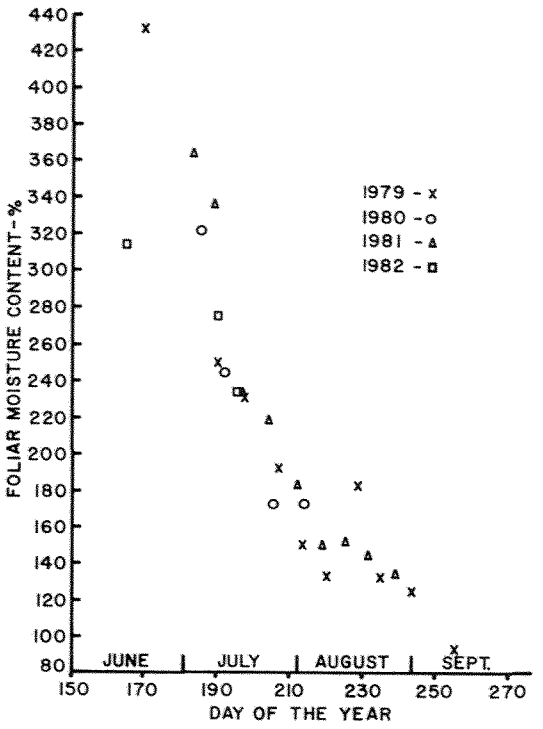

Figure 2. Annual trends in moisture content of new black spruce needles for four years at Kapuskasing, Ontario.

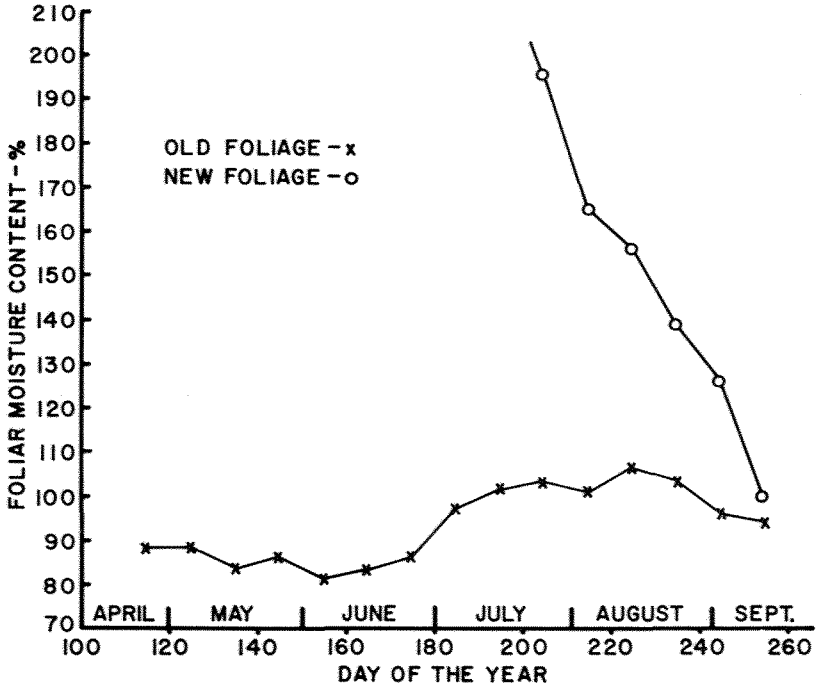

Figure 3. Average annual trends of old and new black spruce needles at Kapuskasing, Ontario.

determination (r^2).

	n	r^2
1979/1980	5	0.76
1979/1981	9	0.66
1979/1982	3	(insufficient joint data)
1980/1981	8	0.79
1980/1982	5	0.72
1981/1982	6	0.84

The r^2 values average 0.75, suggesting that about three-quarters of the differences between pairs of annual trends is mutually accounted for. This result is to be considered descriptive of the present results, but not definitive of the true picture. The regression line coefficients were also examined, but there are too many anomalies to warrant an attempt at interpretation. Sampling density was insufficient for any further analysis.

The conclusions to be drawn are fairly simple. The spring dip is the dominant feature of the old-foliage trends, in common with all other studies referenced. Its magnitude, namely a rough 20-point difference in FMC between spring and midsummer also matches other findings. The timing of the dip, about six weeks centred around June 1, is about two weeks later than for Petawawa at 46°N (Van Wagner 1967), and about five weeks later than for spruce on the west coast (Russell and Turner (1975). The new foliage, as expected, flushes at over 300% MC; its FMC gradually falls to within 5 points above the old-foliage FMC by mid-September.

With respect to timing, there is Little's (1970) evidence that the spring dip in FMC is mainly physiological. In addition, Van Wagner (1974) tried without success to link the timing of the spring dip over a period of 6 yr with some measure of cumulative daily weather. It could therefore be argued that the annual trend probably does not vary greatly from year to year. If so, then the composite graphs (Fig. 3) are a fair representation of the average annual trend of black spruce foliar MC in northeastern Ontario. The results of this study constitute one more benchmark of conifer FMC trends in Canada, and could be used in any scheme that may be developed to explain or predict potential crown fire incidence or behavior in that region.

References

Dieterich, J.H. 1963. Fire control systems. Pages 35-37 *in* Annual Report 1963 – Lake States Forest Experiment Station. U.S.D.A. For. Serv.

Fuglem, P.L.; Murphy, P.J. 1980. Foliar moisture and crown fires in Alberta. Alberta Energy and Nat. Res. Rep. No. 158. 47 p.

Gary, H.L. 1971. Seasonal and diurnal changes in moisture contents and water deficits in Engelmann spruce needles. Bot. Gaz. 132(4): 327–332.

Little, C.H.A. 1970. Seasonal changes in carbohydrate and moisture content in needles of balsam fir (*Abies balsamea*). Can. J. Bot. 48: 2021–2028.

Molchanov, V.P. 1957. Conditions for the spread of crown fires in pine forests. Lesnoe Khoziaistvo 10(8): 50–53. Translated from the Russian.

Russell, R.N.; Turner, J.A. 1975. Foliar moisture trends during bud swelling and needle flush in British Columbia. Can. For. Serv. Bi-Monthly Res. Notes. 31(4): 24–25.

Van Wagner, C.E. 1967. Seasonal variation in moisture content of eastern Canadian tree foliage and the possible effect on crown fires. Can. Dep. For. and Rural Dev. For. Br. Pub. 1204. 15 p.

Van Wagner, C.E. 1974. A spread index for crown fires in spring. Can. For. Serv. Petawawa For. Exp. Sta. Inf. Rep. PS-X-55. 13 p.

Van Wagner, C.E. 1977. Conditions for the start and spread of crown fire. Can. J. Forest Res. 7(1): 23–34.

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