bi-monthly research notes

A selection of notes on current research conducted by the Canadian Forestry Service and published under the authority of the Minister of the Department of Fisheries and the Environment. A French edition is published under the title of Revue bimestrielle de recherches.

SOILS AND FERTILIZER

Growth Response to Aerial Forest Fertilization. — In comparison with small-scale experimental forest fertilization, the direct measurement of growth responses to large-scale operational forest fertilization is costly and time-consuming because it requires the use of intensive sampling techniques. On large tracts of forest land, responses to fertilization may be masked or obscured by the natural heterogeneity of the microclimate, topography, drainage, vegetation, and soils of the area. Response to aerial forest fertilization operations has been assessed (Mitchell and Kellogg, Can. J. Forest Res. 2:95-97, 1972; Windsor and Reines, J. For. 71:659-661, 1973) but has provided poor growth estimates for the entire forest since the assessments were confined to single plots (30-100 trees per plot) in fertilized and unfertilized areas. In this study, we attempted to obtain a more precise measure of the growth response to operational aerial fertilization by using a multiple-plot sampling technique in which fertilized and unfertilized plots were paired by similar pretreatment stand criteria.

Fertilization was undertaken by Scott Paper Company Ltd. on a young stand in central Nova Scotia that contained predominantly 10- to 20-year-old red and white spruce and balsam fir at an average stocking of about 23,000 stems/ha. The area is mapped as a Shulie series, a gravelly sandy loam soil derived from glacial till overlying sandstone bedrock (Wickhard and Smith, N.S. Soil Surv. Rep. 3, 1948). The terrain is gently sloping and affords good drainage. The area was marked into four adjacent strips (340 x 2 800 m), about 97 ha each. Two nonadjacent strips were fertilized; the other two were controls. Granular urea fertilizer was broadcast at the rate of 336 kg/ha (165 kg N/ha) by two fixed-wing aircraft, on 3, 5, and 6 October, 1970. Flight paths and fertilizer dissemination were monitored and controlled by observers at swath-width positions at opposite ends of each target strip. This helped to reduce the unevenness of spread associated with aerial applications of fertilizer (Armson, Univ. Toronto For. Tech. Rep. 11, 1972).

In November 1970, eighteen 0.004-ha circular plots were established at about 120 m intervals along the center line of each strip. Plot locations were staggered within 60 m of the center line to ensure placement on different flight paths. In each plot, all trees were marked with paint at breast height and tallied with calipers in 6 mm diameter classes. Ten randomly chosen dominant and codominant trees in each plot were tagged and measured with a height pole. The number of trees ranged from 42 to 78 per plot. Totals of 3,394 and 3,465 trees were measured in the fertilized and control areas respectively; the average tree heights were 3.66 m in the fertilized and 3.97 m in the control plots. In November 1974, four growing seasons after fertilizer treatment, the plots were remeasured by the same procedure. Mortality and ingrowth were determined from tallies of painted dead trees, and from unpainted live trees that had reached breast height.

One year after fertilization, current foliage samples were clipped from balsam fir and white spruce to assess N concentrations. Composite samples were analyzed from 10 trees per strip (5 at each end) for each species, for a total of 20 trees/species per treatment. Mean N concentrations were 0.42% higher in the fertilized area (1.60% N) than in the control (1.18% N), indicating that N fertilizer was obsorbed by the trees.

After remeasurement in 1974, individual plot basal area (BA) was computed by totalling BA yield of each diameter class (i.e. class mark BA x no. of trees). Periodic BA increment was the difference in plot yields at the two measurements adjusted for ingrowth and mortality. Periodic height increment was the difference in mean height of the 10 sample trees in each plot.

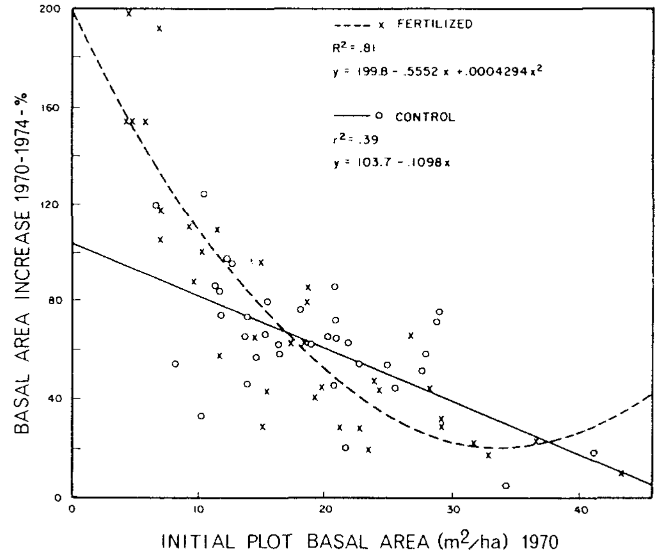

Figure 1. Relationship between periodic basal area increment and initial plot basal area.

The systematic placement of plots in this area yielded a series of sample plots with a wide range in BA (Fig. 1). To ensure response comparisons between plots of similar BA, the initial plot basal areas in each strip were systematically ranked and then paired according to rank for each treatment. Both the absolute and the relative BA increment between paired plots were tested by ANOVA, and showed no significant differences between fertilized and unfertilized plots. However, noticeably larger BA increments were evident in fertilized plots of low BA (Fig. 1). Polynomial curves of degrees 1 to 3 showed no significant deviations from linearity in the control plots, whereas the fertilized plots had a significant quadratic component. Height response assessment involved similar pairing procedures, initial plot height being used instead of BA. Although ANOVA failed to detect significant height increment in absolute terms, relative height growth was significantly greater ($P>.05$), increasing from an average of 25.4% (control) to 28.6% (fertilized).

Despite the relatively large number of plots used in this operational assessment, the number, combined with the level of precision of measurement used, was insuffucient to detect small differences in radial growth, given the stand variability encountered. To detect a BA increase of 10% with a 90% probability (at 0.05 significance level), it is estimated that 110 plots would be needed (Snedecor and Cochran, page 111 *in* Statistical Methods, Iowa State Univ. Press, 1972). Under these heterogeneous operational conditions, the assessment of height and diameter responses is apparently more sensitive in relative than in absolute terms. This may be the result of differing relative responses of plots at varying pretreatment stand conditions. — V.R. Timmer and R.A. Fisher, Maritimes Forest Research Centre, Fredericton, N.B.

TREE BIOLOGY

Anatomical Modifications in Xylem of Lodgepole Pine Container Seedlings Induced by TOK-E-25 (Nitrofen). — Auxin herbicide TOK-E-25 is a 2, 4-D derivative that affects a wide range of plant species during their active growing stage. Since nurserymen find handweeding almost prohibitive in closely spaced container seedlings, TOK-E-25 is commonly used for eradicating various broad-leaved weeds such as dandelions and Salicaceous germinants. However, actively growing conifer seedlings can also be very susceptible to such herbicides. This paper deals with anatomical effects of TOK-E-25 on the xylem of actively growing lodgepole pine container seedlings *(Pinus contorta* var. *latifolia* Engelm.) in the nursery.

The seedlings were developed to 5 weeks of age in the greenhouse and maintained outside for a full growing season. TOK-E-25 was applied at

Figure 1. Five sets of daughter tracheids of lodgepole pine affected by TOK-E-25 show approximately the same stage of development without any sign of fiber tracheids.

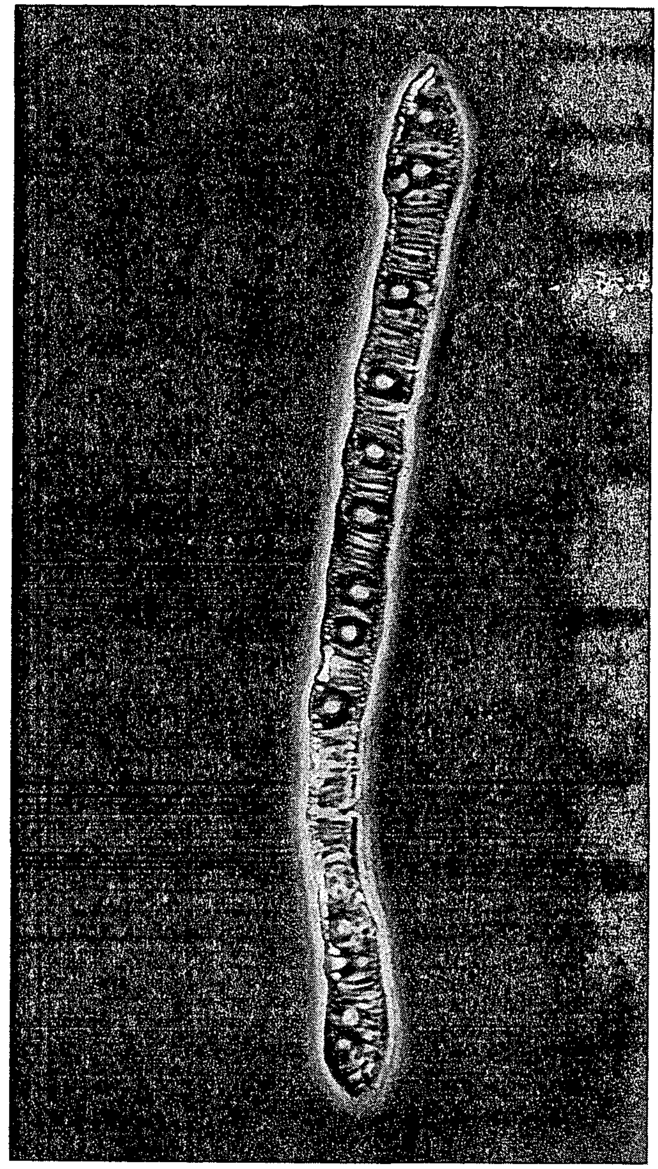

Figure 2. Spiral tracheid of lodgepole pine affected by TOK-E-25 has broad fibrils and bordered pits that are not normally found in primary tracheids.

weekly intervals during May, June, and July to control new weed germinants. First and second year's wood of lodgepole pine seedlings collected after July was prepared for anatomical study by macerating in a 1:1 mixture of glacial acetic acid and 15% hydrogen peroxide.

TOK-E-25 internal effects in seedlings can be readily verified by retarded development of affiliated daughter tracheids. The normal set of daughter tracheids matures as the new set is cut off by the mother cambium. In these seedlings, the first set in the rank of daughter tracheids was indistinguishable from the last set in the series. As many as five sets in the rank of daughter tracheids showed the same stage of development (Fig. 1); there were no mature tracheids or fiber tracheids in these sets. Other daughter tracheids matured to resemble vascular primary tracheids but were short with blunt ends and had double-bordered pits and broad spiral fibrils (Fig. 2) not unlike those of Douglas-fir. Others matured into fiber tracheids with fine spiral fibrils (Fig. 3). Fibril modifications in tracheids caused by TOK-E-25 are diagnostically significant of external agents. Similar modifications were observed in frost rib tissues of Rosaceous species (Zalasky, Can. J. Plant Sci. 56:501-504, 1976) but not in fungus-induced swellings of pine (Zalasky, Can. J. Bot. 54:1586-1590, 1976). The fiber tracheids and tracheids in these pine seedlings were branched and curvate as in Fig. 3. They had a variety of shapes and sizes, as reported for frost-induced chimeras in cambial tissues (Zalasky, Can. J. Bot. 53:1888-1898, 1975).

The main difference between xylem affected by TOK-E-25 and frost- and fungus-affected xylem appear to be delayed maturation of daughter tracheids. The delay in xylem development means that diameter and height increment and phenology of the seedlings can be severely affected. Plants are slower-growing and have less chance of developing a normal bud (author's observations). Pine must develop secondary needles to produce a bud that yields a long shoot rather than just short shoots bearing needles. The presence of fibrils in tracheids is evidence that cambium affected by TOK-E-25 produced a higher proportion of primary than of secondary xylem.

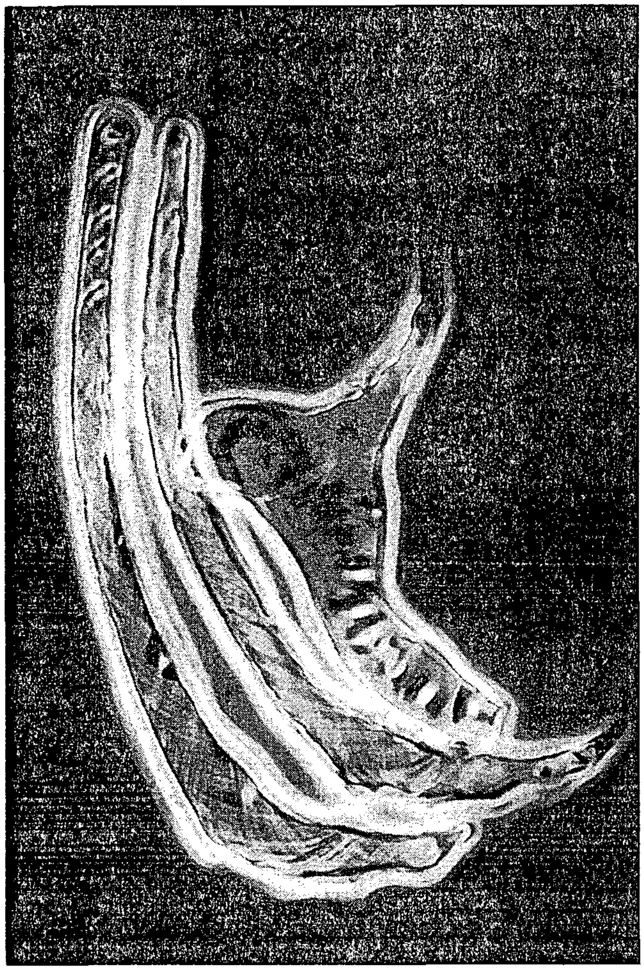

Figure 3. Short spiral fiber tracheids of lodgepole pine affected by TOK-E-25 showing fine fibrils, curvation, and branching.

The herbicide 2, 4-D, from which TOK-E-25 is derived, has been shown to proliferate parenchyma, retard development of vascular strands, and inhibit root growth and primary needle expansion in 2- to 18-day-old pine seedlings (Wu et al., Can. J. Bot. 49:1737-1741, 1977). TOK-E-25 is a highly mobile compound in conifer tissues. Although its activity is suspended over the winter months, it persists in the cambium and xylem during the second growing season, presumably as breakdown products. — H. Zalasky, Northern Forest Research Centre, Edmonton, Alta.

PHYSIOLOGY

Low Molecular Weight RNA in Jack Pine *(Pinus banksiana* Lamb.) Seedlings. — A new class of ribonucleic acid (RNA) with low molecular weight has been detected in animal (HeLa) (Weinberg and Penman, J. Mol. Biol. 38:289-304, 1968) and bacterial *(Escherichia coli)* cells (Ikemura and Dahlberg, J. Biol. Chem. 248:5024-5032, 1973). This RNA has been termed smRNA. Its occurrence adds to the already recognized RNA types such as messenger, ribosomal, and transfer RNA's. Our report provides evidence for smRNA in jack pine. The new class of low molecular weight RNA has so far not been observed in gymnosperms. In animal cells Weinberg and Penman (1968) observed that the RNA fraction contained six discrete species ranging in size from 100 to 180 nucleotides (4-6S RNA). These components represented only 0.4% of the total RNA and were metabolically stable. Bases of at least four of the components were extensively methylated. Although the authors termed this fraction "the small molecular weight monodisperse nuclear RNA's,"

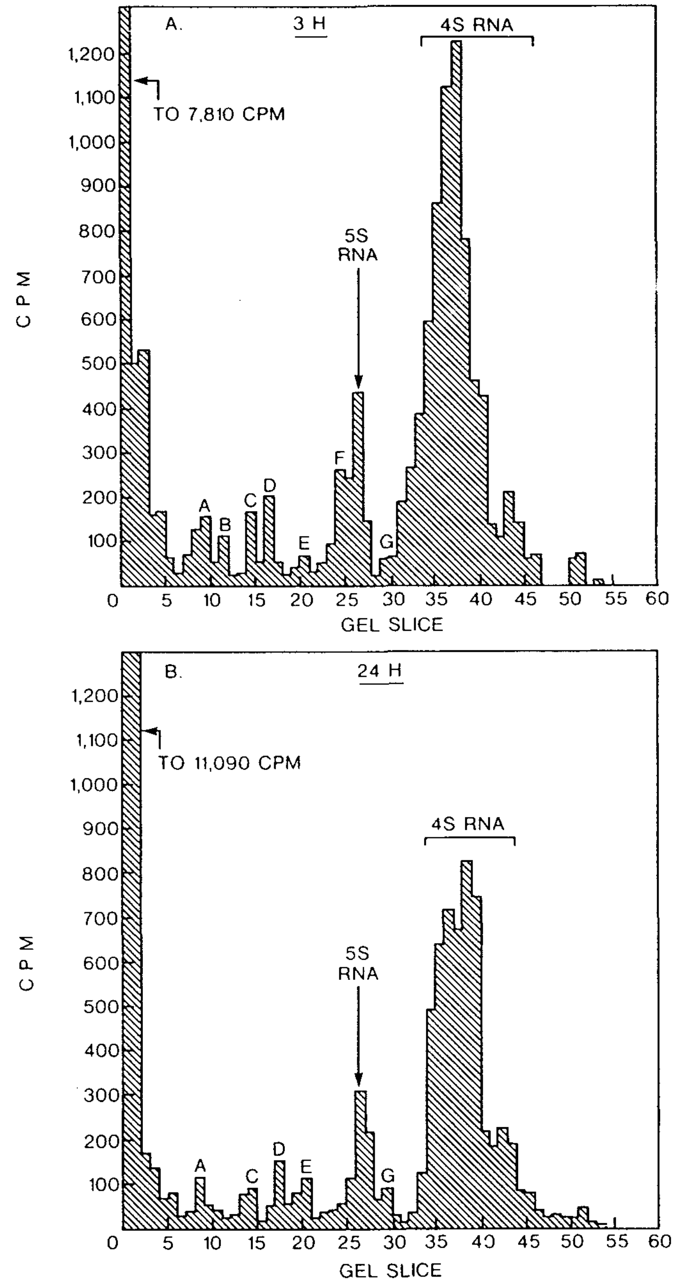

Figure 1. Distribution of radioactivity in jack pine RNA along a 10% polyacrylamide gel after a (A) 3-h and a (B) 24-h exposure to 50 μCi uracil-6-^{3}H. SmRNA's are identified by radioactive peaks A to G. Migration of RNA is toward the anode at the far right.

other studies have shown that these components are also present in the cytoplasm (Frederiksen and Hellung-Larsen, FEBS Lett. 58:374-378, 1975). In the latter study, unstable precursors to the smRNA's and four stable low molecular weight RNA's were identified.

Jack pine seeds were obtained from the Petawawa Plains, Ont. Seeds with germination greater than 95% were surface-sterlized with 5% calcium hypochlorite, washed, and germinated under constant light not exceeding 6.2, 5.5, and 2.7 μW/cm^2/nm in the blue, red, and far-red respectively. Six-day-old seedlings were exposed to 50 μCi of uracil-6-^{3}H (specific activity 8.9 Ci/m mol). Duplicate samples of seedlings were taken after 3 and 24 h of exposure. Seedlings were washed in distilled water and extracted for total RNA (Pitel and Durzan, Can. J. Bot. 53:673-686, 1975).