RELATIONS BETWEEN SOME PHYSIOLOGICAL FUNCTIONS IN LODGEPOLE PINE AND RESISTANCE TO THE MOUNTAIN PINE BEETLE

by

R. W. REID and H. S. GATES

NORTHERN FOREST RESEARCH CENTRE
INFORMATION REPORT NOR-X-15
MARCH, 1972

CANADIAN FORESTRY SERVICE
DEPARTMENT OF THE ENVIRONMENT
5320 - 122 STREET
EDMONTON, ALBERTA, CANADA
T6H 3S5

TABLE OF CONTENTS

Page

RELATIONS BETWEEN SOME PHYSIOLOGICAL FUNCTIONS IN LODGEPOLE PINE AND RESISTANCE TO THE MOUNTAIN PINE BEETLE

by

R. W. Reid* and H. S. Gates**

ABSTRACT

Stands of lodgepole pine exhibited varying degrees of resistance to attack by the mountain pine beetle. Variations in water tension within individual trees were reflected directly in circumference expansion and contraction, particularly following periods of stress, and this characteristic was related to some extent with the incidence of successful insect attack. Growth rhythms in the tree stems were not detected and degree of starch accumulation in the bole did not appear related to the degree of tree resistance.

INTRODUCTION

Studies are reported herein into relations concerning several physiological functions in lodgepole pine (*Pinus contorta* var. *latifolia* Engelm.) and response by that tree species to attack by the mountain pine beetle (*Dendroctonus ponderosae* Hopk.) and subsequent infection by blue stain fungi.

The response of lodgepole pine to wounding by bark beetles and their associated blue stain fungi is complex. Nevertheless, that response exhibits a seasonal cycle as shown by earlier studies (Reid,

* Program Manager and formerly Research Scientist;

** Research Technician, Northern Forest Research Centre, Canadian Forestry Service, Environment Canada, 5320 - 122 Street, Edmonton, Alberta. T6H 3S5

Whitney, Watson, 1967) which demonstrated a seasonal change in response generally minimal in early spring, increasing to a maximum by mid-summer, and followed by a decline in late summer. There were differences, sometimes significant, between individual trees as to when in the season their peak resistant response occurred. Many normal physiological functions in the tree also exhibited a seasonal cycle and were undoubtedly important to the resistance phenomenon.

In the study reported here efforts were made to describe and monitor several easily measurable physiological functions and relate those measurements with resistance by the tree to attack by bark beetles and infection by blue stain. The objective was development of a simple, accurate and diagnostic test for determining degrees of resistance sufficiently sensitive to separate individual trees. That objective was only partially realized and while many of the relations were not clearly defined, neither were all found unrelated. The factors investigated were radial growth, bole expansion, water transport, and starch accumulation.

METHODS

Temperature

Temperature for the summer seasons 1964 through 1969 was recorded near Radium, B. C. by use of a Fuess Hygrothermograph in a Stevenson weather screen and checked against Canada Department of Transport records from a nearby weather station (Brisco). Monthly precipitation data for the same period were also obtained from this weather station. All test plots were located within a five mile radius of the point at which temperature and precipitation were measured.

Growth

Radial xylem growth was measured by several methods. In 1966, two hundred trees were numbered and every two weeks, beginning in June, ½ inch square chips containing approximately 5 years of growth were removed from each of 20 randomly selected trees within a contiguous stand of 70 year old lodgepole pine. All chips were stored in a formaldehyde solution in sealed containers until sampling was completed in September. A final chip was taken in September from each tree previously sampled in order to determine total seasonal growth on that particular region of the bole. That chip was removed adjacent and radial to the earlier one. Cell counts of the current seasonal growth on individual chips were made and calculated as a percentage of total growth for the year as determined by cell counts from the final chip collected in September.

In 1968, another method, was used for measuring radial xylem growth and provided more precise data. This method (Walter, 1968) required marking the cambial zone and differentiating xylem at regular intervals throughout the growing season with a micro-needle. Injury by the micro-needle caused a minute irregularity in the cambial zone and this irregularity acted as permanent marker indicating position of the cambial zone at time of marking. Nineteen trees were selected and the bark pierced to the cambium at regular intervals on a grid previously marked on the boles, at breast height. The space between adjacent piercing sites was 2 cm, sufficient to permit formation of a buffer of uninjured cells. That portion of the stem having the most uniform growth pattern was selected for location of the grid. In the fall, these marked sites were removed in chip form. Sections were then made from immediately above the injury and mounted on slides for microscopic examination. The number of cells formed prior to marking were counted and expressed as a percentage of total current growth as determined from counts made adjacent to the marked zone.

Radial growth during the preceding six years was compared on increment cores taken from 10 randomly selected trees in October, 1969. The cores were cut longitudinally, sanded, and annual growth for the past 6 years was measured in mm.

Radial Expansion - Contraction of Stems

Dendrometer tree bands (Hall, 1944) constructed of 1.5"-wide and 0.01 inch thick aluminum, were used for recording bole expansion and contraction, an index of water tension within the bole of the tree moisture which in turn was a response to varying regional climatic influence. All components used in construction were standardized to reduce error and verniers and scales were reproduced from a single photographic negative to ensure precision. The rough bark was partially removed and smoothed to provide as much band-to-tree contact as possible. Bands were installed with tension controlled by two identical springs and several days were allowed for settling before the first readings were taken. All measurements were recorded to 0.01 inches.

Two plots of 10 trees each were located in a wet and a dry site to record the influence of moisture availability on bole expansion and contraction. Three more plots of approximately 25 trees each were located in dissimilar sites with respect to stand density and availability of soil moisture, i. e. dry sites were located on exposed sandy benches while wet sites were adjacent to rivers or ponds.

Water Tension

The water tension pressure bomb, as described by Waring and Cleary (1967) was used as a measure of the relative water tensions within individual trees. By that method, two to three year-old tips from lateral branches were sealed in an air-tight pressurized container with their cut stems protruding. Nitrogen was then introduced into the pressure bomb chamber until pressure was built up sufficiently to force water from the exposed stem. Pressure was recorded at the first appearance of free water on the cut surface.

Before extensive inter-tree sampling was undertaken, tests were made to determine intra-tree variability. Two groups of 10 trees each were located: (1) in a hot, dry valley bottom; and (2) on a cool, moist, north facing slope. The trees were approximately 70 years of age, averaged 8 and 12 inches in diameter in each plot and near 40 feet in height. These trees were sampled over a twenty-four hour period at 6-hour intervals to determine the diurnal variation in moisture tension.

Six trees, 50 feet in height, were sampled for moisture tension variation within the crown. Three of these trees were open-grown with crowns extending to ground level, while the other 3 trees were in a closed stand and the lower level of their live crowns commenced at 30-feet. Each of the trees was sampled at 3 levels: (1) lower crown, (2) mid-crown, and (3) upper crown (with the lower crown being the level of the lowest living branches). Samples were also taken to determine moisture tension variation around the circumference of the crown.

Trees in the 5 plots chosen for bole expansion-contraction studies were also sampled regularly for water tension.

Starch

The xylem starch content during periods of growth has been sampled regularly by us in this region since 1964. During that year the starch content of lodgepole pines was sampled at monthly intervals, from May to September, at a number of points in the Columbia River Valley (East Kootenays) between Golden and Canal Flats. Since no difference in starch content of trees could be detected between the sampling points by our methods, in subsequent years sampling was confined to a single stand. Each spring, 10 trees were selected from an extensive stand of 60 or 70 year old trees and increment cores were removed at 2 week intervals throughout the summer. After removal, the cores were cut lengthwise and quickly dried. Upon completion of sampling in the fall, all cores were treated with IKI starch indicator solution (Reid and Watson, 1966) and the resulting starch-ioding blue colouration visually assessed. Starch content was rated within a range from 0 to 5; absent 0, very low 1, low 2, medium 3, high 4, very high 5. Cores were stored in a dry atmosphere and succeeding years cores compared with those of the preceding years to reduce error in year to year assessment.

Tree Resistance and Susceptibility

Inoculation of selected trees with a blue staining fungi, *Europhium* sp., was undertaken in conjunction with several of the afore-mentioned studies. Tree response to *Europhium* fungi (Reid, Whitney, Watson, 1967) was used as an indicator of the resistance of the tree to attack by the mountain pine beetle (*Dendroctonus ponderosae* Hopkins). During the 1967 summer season, nineteen trees were selected from a total of 200 which had been inoculated with Europhium fungi. These trees had exhibited the greatest and least degrees of reaction to the inoculations and these differences were readily apparent. Ten of these trees with extensive resinous response were rated, utilizing earlier experience, to be resistant, and 9 trees with little to no resinous response were rated as non-resistant. These trees were subsequently fitted with aluminum tree bands to measure their seasonal contraction and expansion. In 1968, seasonal growth rates for these 19 trees were determined by the cambial marking method described earlier. Some water-stress measurements were also made.

RESULTS

Temperature

Mean monthly maximum temperatures during May, June, July, August, and September for the years 1964 to 1969 conformed to a typical pattern. Temperatures gradually increased to a maximum in July and then declined through August and September. Although precipitation was more variable between years, there was a general trend towards heavier precipitation in spring and fall than in mid-summer.

During 1967, temperature and precipitation for May and June remained normal, however, in July and August there was less than one half the normal amount of rainfall. Maximum seasonal temperatures were recorded in August rather than in July, and temperatures for September remained 10°F above normal. During July and August temperature and humidity inversions were common and intensified a developing drought condition. The five-year average monthly maximum temperatures, the average monthly maximums for 1967, and average precipitation are summarized in Figure 1.

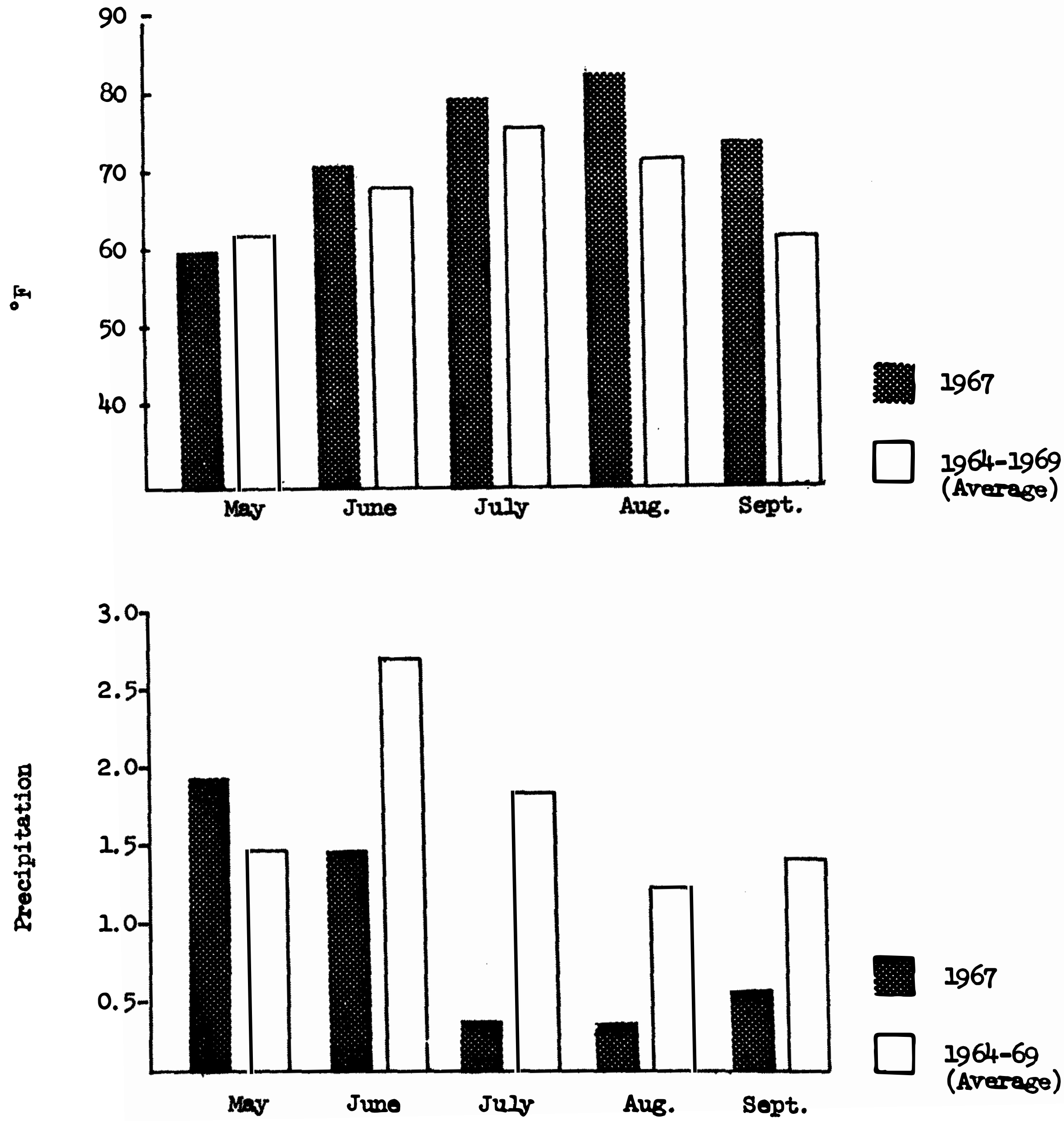

Fig. 1. Monthly maximum temperatures, and precipitation, near Radium, British Columbia May-September 1967; and 1964-1969 (minus 1967)

<u>Growth</u>

Rates of growth during a single season as determined from chips removed at intervals from the stems of lodgepole pine are summarized in Figure 2. Growth was well advanced when sampling commenced, and progressed after mid-June at a near linear rate.

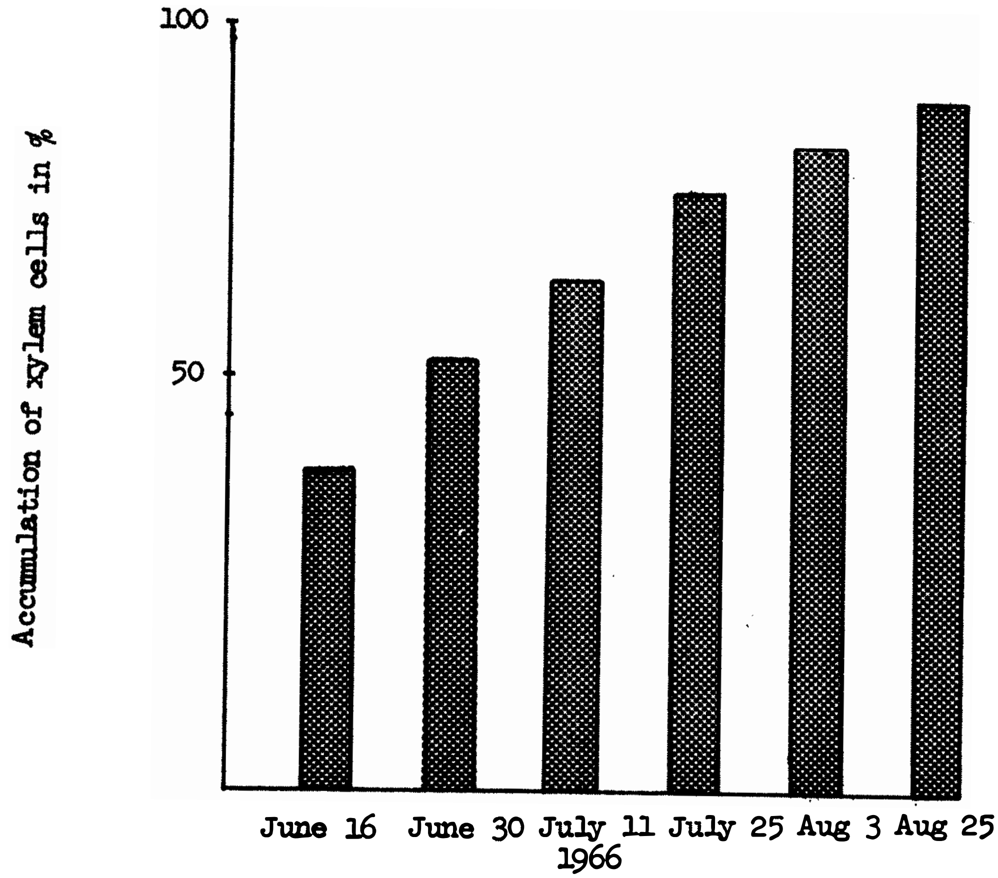

Fig. 2. Cumulative radial growth of lodgepole pine in 1966 near Radium, British Columbia

The cambial marking method of assessing seasonal growth, used in 1968, provided more sensitive radial growth data and required less time to implement than other methods tested. Again, growth progressed in a near linear fashion throughout the growing period (Fig. 3). Variations were found to occur between trees in the cessation of their growth periods. Xylem cell accumulation in the fastest growing tree, slowest growing tree, and the average tree are compared in Fig. 3. In both 1966 and 1968, some trees completed their growth up to 40 days sooner than others. Approximately 20% of total growth had occurred by 16 June 1968 as compared with 50% by the same date in 1966 (Fig. 2, 3).

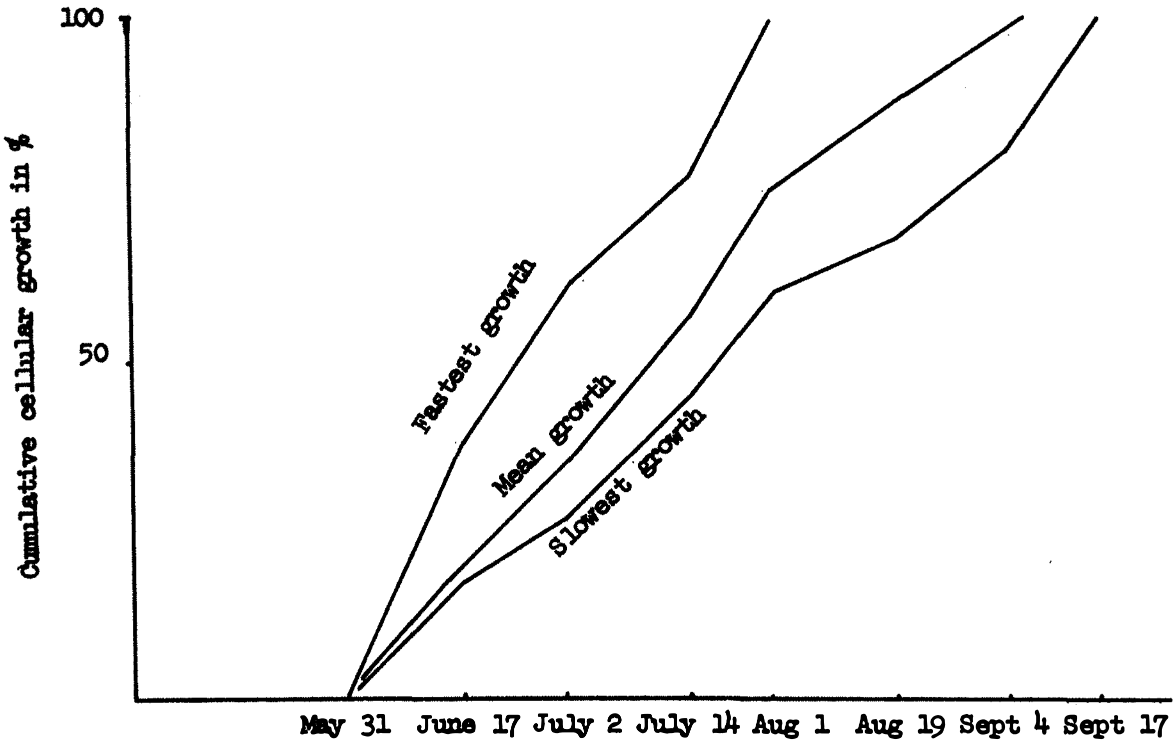

Fig. 3. Slowest growing tree, fastest growing tree, and mean growth for 19 lodgepole pine trees in the Radium, British Columbia area, as determined by the cambial marking method (Walter, 1968).

A comparison of annual radial growth from 1964 to 1969, as shown in Figure 4, indicates the sharp decline between 1966 and 1968. It was noted that although poor growing conditions prevailed during 1967, the greatest decline in annual increment did not occur until the following year, which itself was a good growing year.

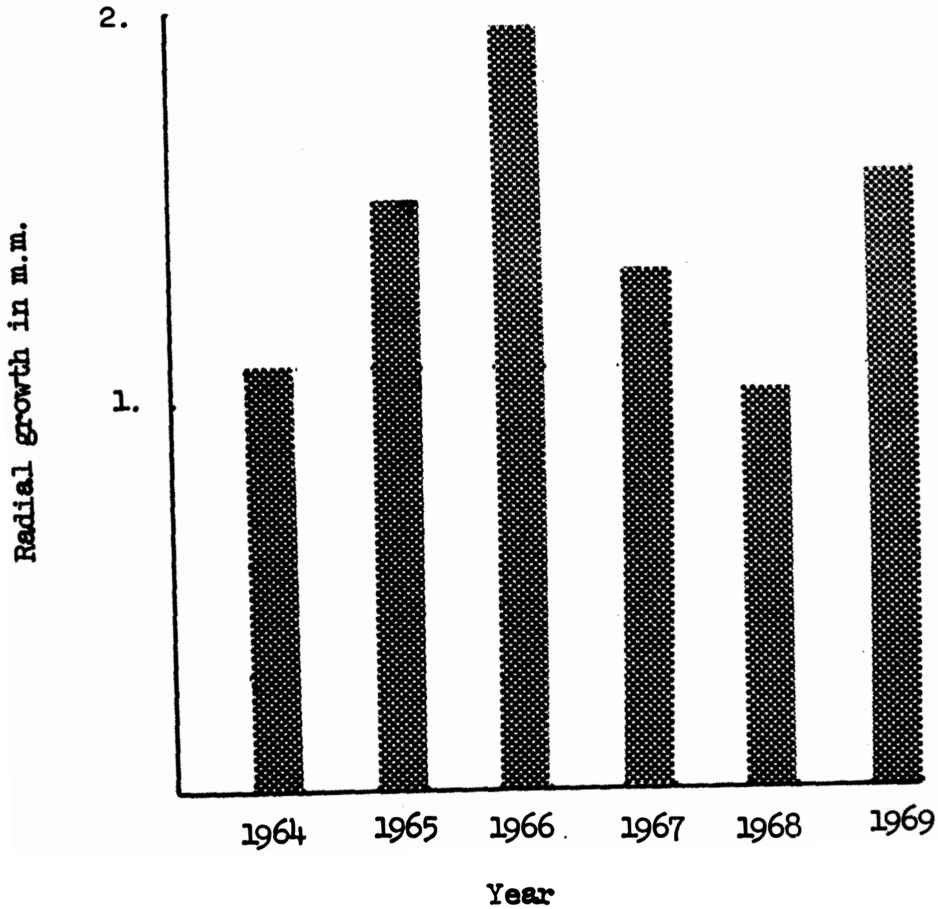

Fig. 4. Radial growth in lodgepole pine near Radium, British Columbia 1964-1969.

Radial Expansion-Contraction of Stems

In the 3 plots, which were located specifically for the examination of the relation between site and bole expansion, all trees separated out into one of two distinct groups. One group showed a gain and the other a seasonal net loss in diameter at breast height. The seasonal bole expansion and contraction curves within each of the 3 plots are shown in Figure 5. The number of trees in each category are given in Table 1.

Table 1. Number of lodgepole pine trees expanding and contracting in 3 plots and their mean D.B.H.

Plot No.	No. Expanding	Mean D.B.H. (in.)	No. Contracting	Mean D.B.H. (in.)	Total No. Trees
1	18	8.8	4	10.2	22
2	19	9.5	6	8.1	25
3	4	10.7	21	10.6	25
Grand Totals	41		31		72
Means		9.7		9.6	

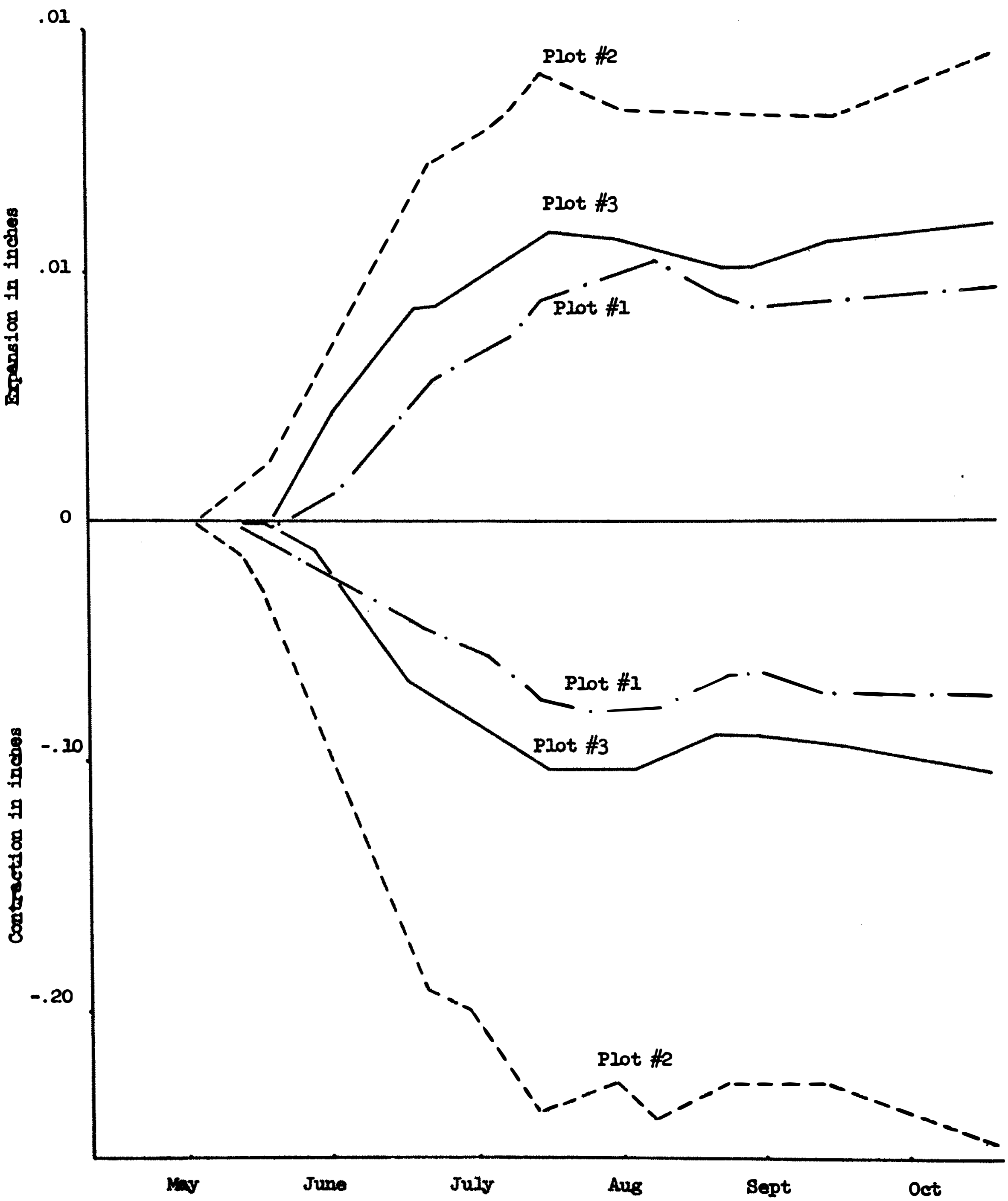

Fig. 5. Average bole expansion and contraction of lodgepole pine in three sample plots near Radium, British Columbia, 1969.

A curious and unexplained phenomenon occurred in over 40% of the trees monitored for bole expansion. Those individuals contracted. Data from the two groups of trees exhibited almost "mirror images" of each other. Root grafting was not believed a factor, as the affected trees were often too far apart.

Frequently, both expanding and contracting trees of similar age and diameter, growing side by side had put on identical radial growth as measured from increment cores taken at the end of the season. It appears therefore, while trees put on comparable radial growth, cell division occurred in both trees at a near equal rate; such trees were experiencing unequal degrees of dehydration during the growing season. There was no relationship between D.B.H. and expansion or contraction.

The results of a two-year study (1968-69) of bole expansion in trees, located in wet and dry sites, indicated boles of most trees from both sites continued to contract throughout the growing season in 1968 (Fig. 6). This radial shrinkage of the boles was attributed to water stress related to adverse climatic conditions which occurred in 1967 (Fig. 1). By 1969, all trees had apparently recovered from the 1967 drought and showed a net gain in bole circumference; however, trees on wet sites exhibited a greater increase than trees on dry sites.

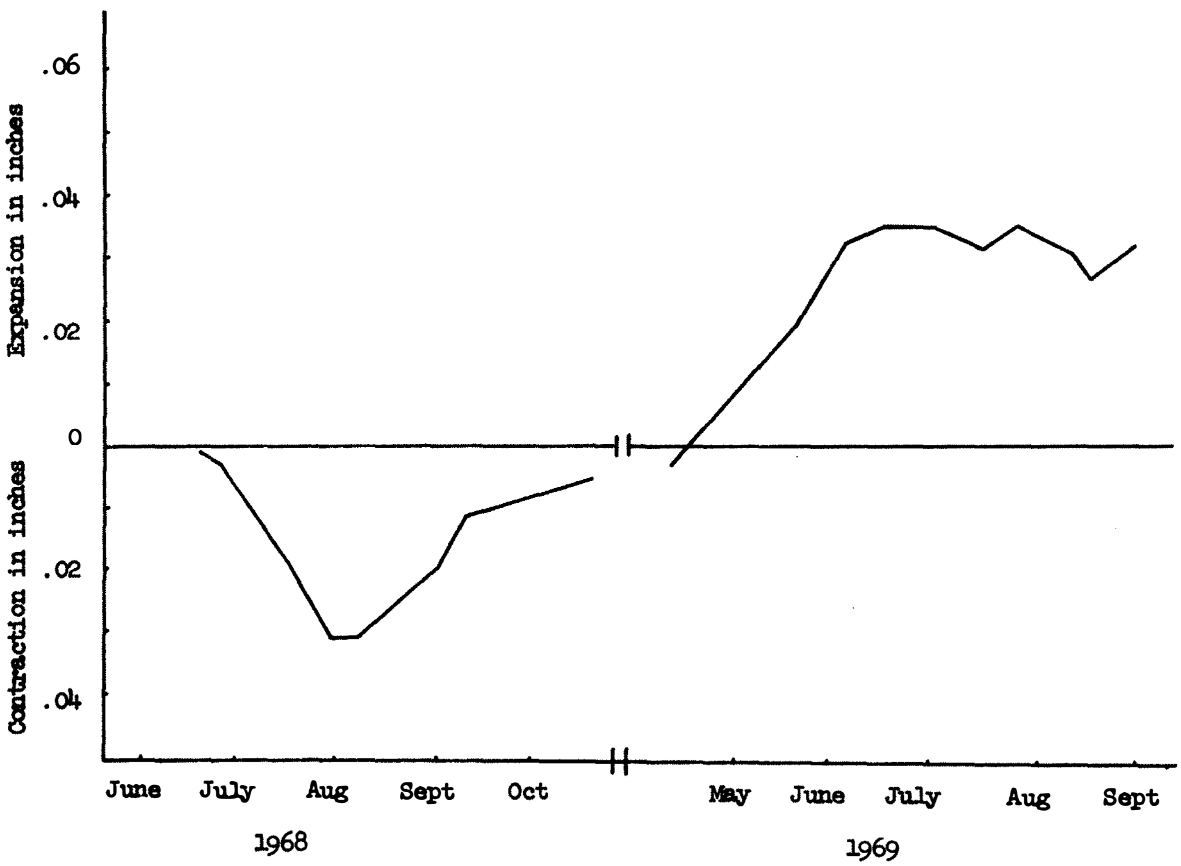

Fig. 6. Average bole expansion and contraction of 5 lodgepole pine stems during 1968-69.

Water Tension

Water tension, as determined by the pressure bomb (Waring and Cleary, 1967), showed considerable variation vertically within the crown (Fig. 7) and diurnally (Fig. 8) on both dry and wet sites.

Water tension in the crown increased proportionate to height above the base of the crown (Fig. 7). Although all trees exhibited increasing water-tension with height in the crown, open grown trees had less variation vertically within the crown than those individuals in closed stands. Allowing for these variables, trees located in wet sites exhibited lower water tensions than trees on dry sites. However, the diurnal range, or difference between high and low, is about twice that of trees growing on dry sites. On the average, periods of highest water tension occurred between 12 noon and 3 p.m., and lowest tension during the morning hours from 6 to 8 a.m. The time during the day at which maximum or minimum water tension occurred was weather dependent and was influenced by changes in climatic factors such as cloud cover, precipitation, or temperature. Trees growing in dry sites reached their maximum water tension somewhat earlier than those in moist sites. Moreover, trees in the dry sites maintained a maximum, over longer periods, than those in the moist sites.

Small differences in water tension were recorded between sunlit and shaded aspects of the same crown; those differences were inconsistent and their magnitude was within the range of instrument error. There was, however, wide difference between trees where one crown was entirely shaded and the other entirely exposed to sunshine. Water tension measurements from suppressed, co-dominant, and dominant trees were about equal during periods of normal precipitation.

The average seasonal water tension curves for trees growing in wet and dry sites are shown in Figure 9. During 1968, trees in wet sites had lower seasonal water tensions than trees growing in dry sites. In 1969, with good precipitation and two years after the drought, there was no difference in moisture tension between the two types of sites in one area but the differences were maintained in another.

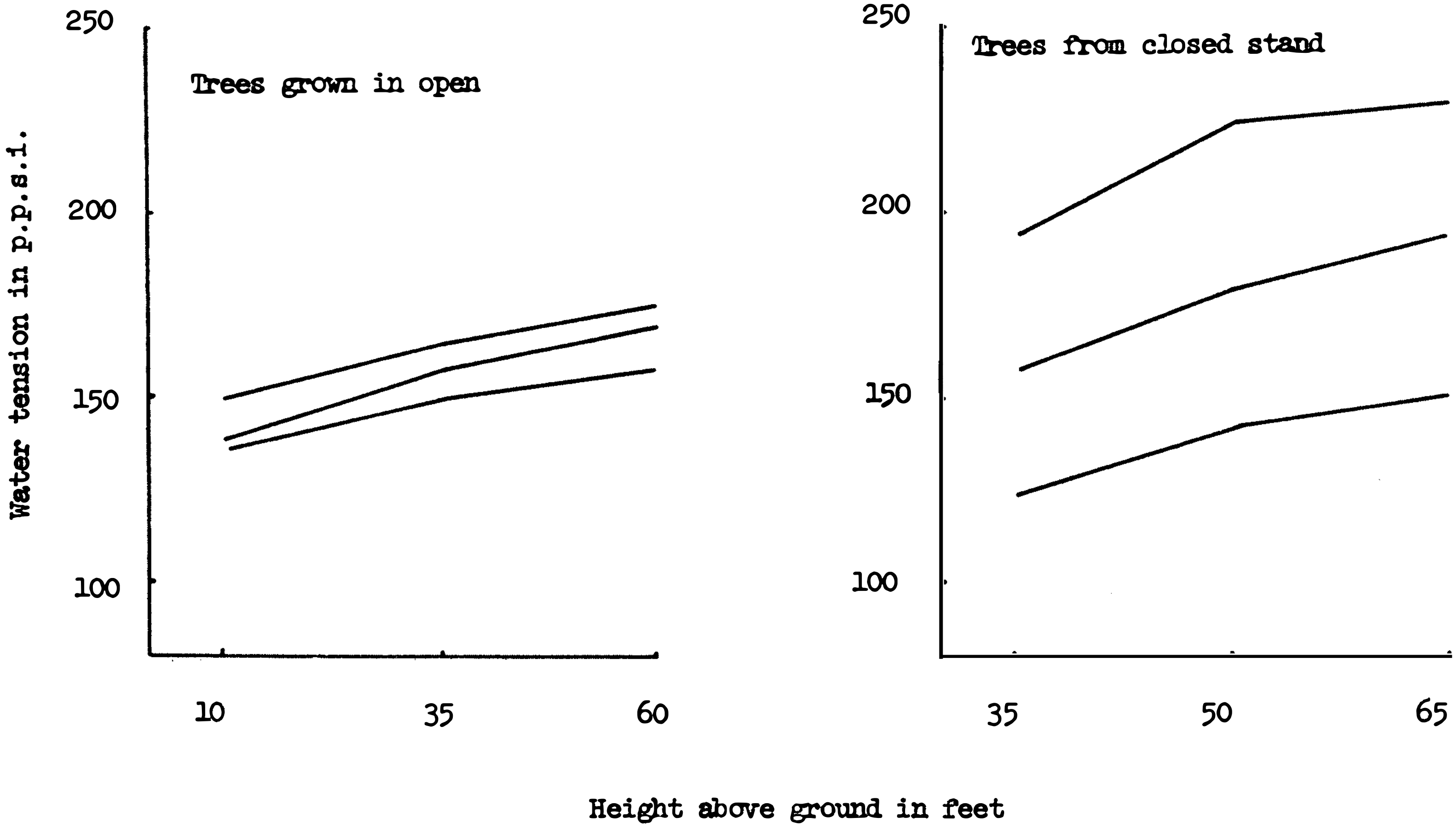

Fig. 7. Water tension gradients within the crowns of 6 lodgepole pine trees in 1968 near Radium, British Columbia. Individual trees graphed.

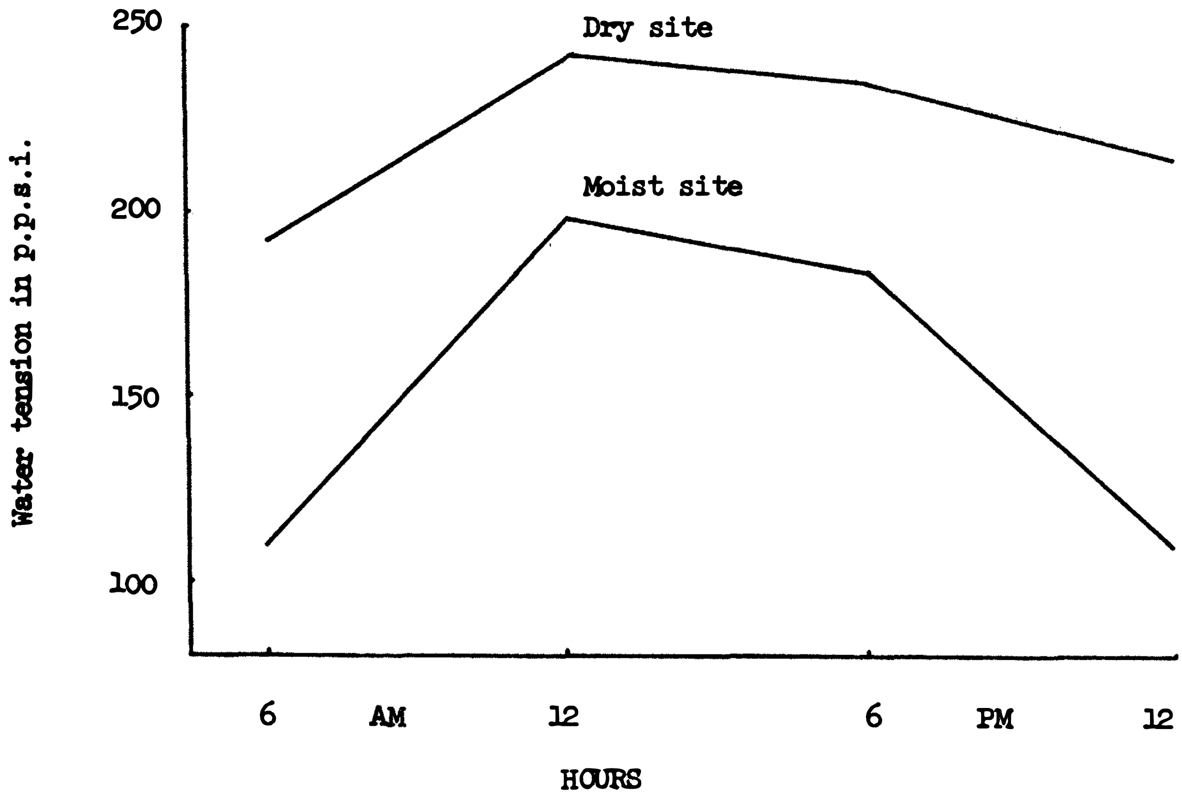

Fig. 8. Average diurnal water tension curves for lodgepole pine occurring on wet and dry sites near Radium,British Columbia, 1968. Measurements were made in the lower crown on each curve represents 5 trees.

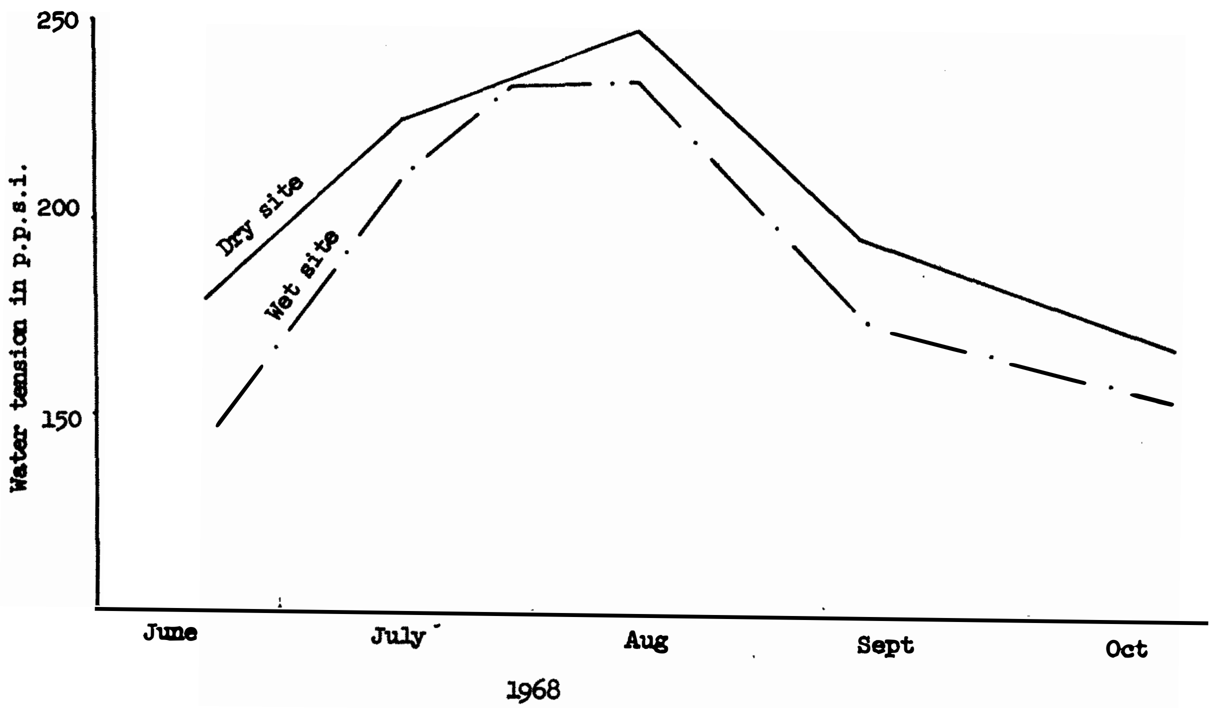

Fig. 9. Average seasonal water tension curves for lodgepole pine trees growing on wet and dry sites in 1968 near Radium, British Columbia. Each curve represents 5 trees.

Starch

Summarized data illustrating the seasonal starch cycle over a 6-year period are shown in Figure 10. The low starch content evidenced in trees during 1968 is believed related to the drought which occurred in 1967. By 1969, some recovery was noted but by no means equaled the amounts recorded in earlier years; 1968 and 1969 were generally favorable years for growth. Distinct differences in starch content, detectable by our methods, occurred between years 1968, 1969, and 1964-67.

The IKI test for starch was applied to trees identified for their resistant or non-resistant potential response to beetle attack. These trees were selected by methods previously described (Reid *et al.*, 1967).

That method involved inoculating trees with ¼-inch wood chips impregnated with a blue stain fungi. The response by the tree in formation of resin, amount and its distribution around the inoculation point are used as an indice of resistance. No differences in starch concentration were detected between these groups of resistant and non-resistant trees.

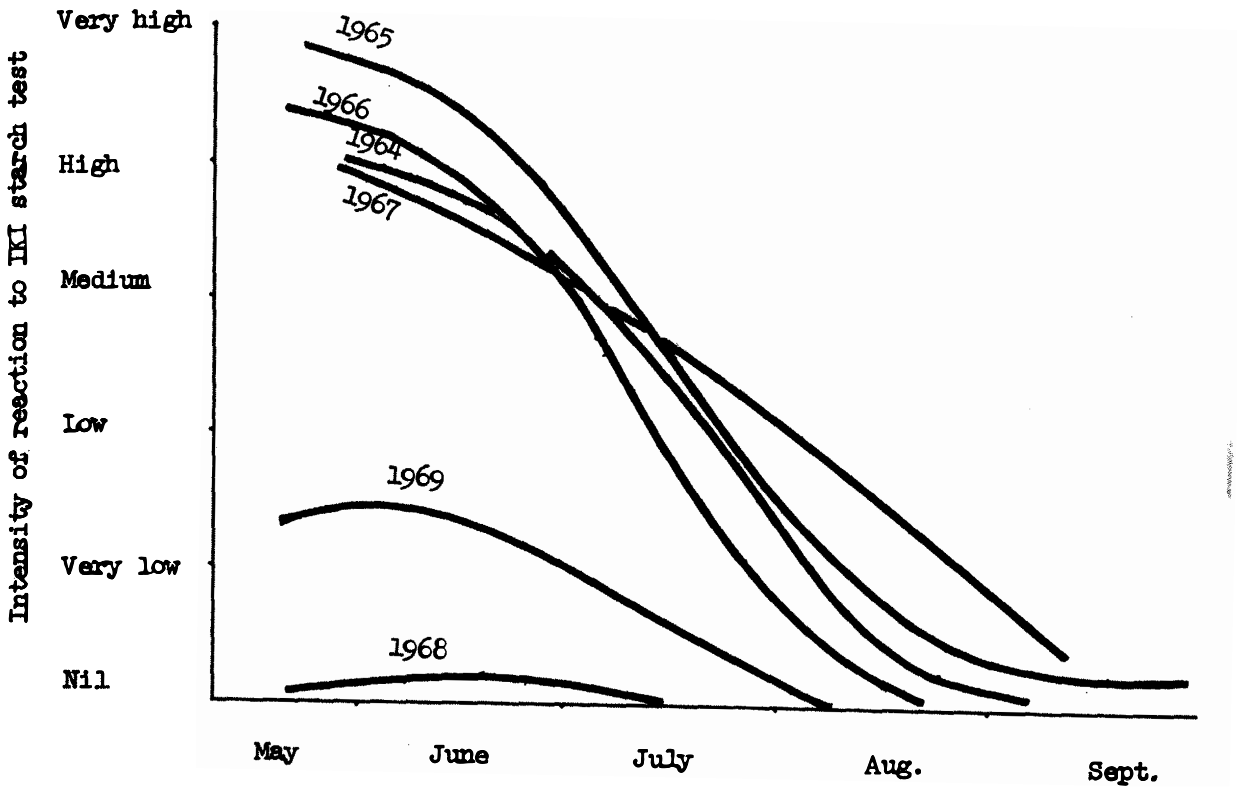

Fig. 10. Seasonal starch cycle in stems of lodgepole pine trees near Radium, British Columbia 1964-69.

Tree Resistance and Non-resistance

Of the resistant and non-resistant trees on which radial expansion was recorded over a 2-year period, resistant trees showed the greater expansion. The average expansion rate over the 2-year period for resistant and susceptible trees is shown in Figure 11. Resistant trees averaged 16% greater in D.B.H. and a correction factor for that amount was applied in Figure 11. As seen in Figure 11, when local conditions were such that little or no expansion occurred in resistant trees, non-resistant trees suffered a shrinkage in diameter.

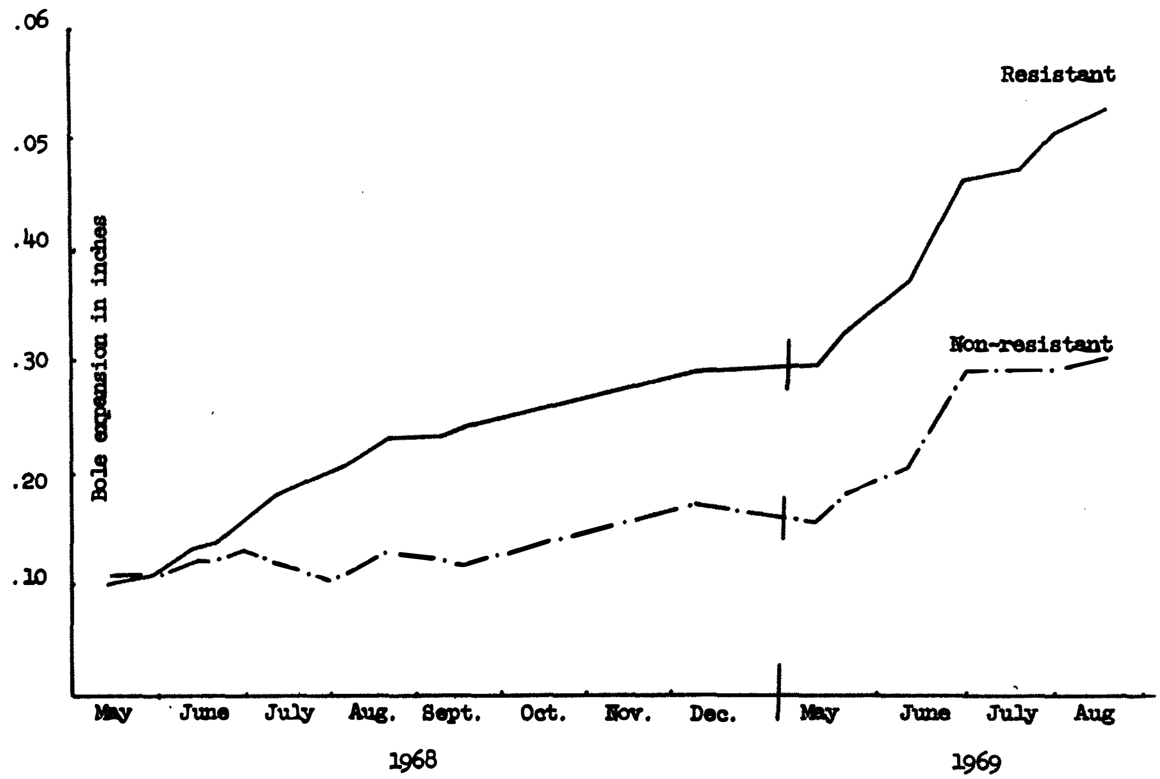

Fig. 11. Average expansion in 10 resistant and 9 non-resistant lodgepole pine trees near Radium, British Columbia, 1968-69.

Also, resistant trees appeared to grow at faster rates, although they completed their seasonal growth at approximately the same time, or slightly later, than non-resistant trees (Fig. 12).

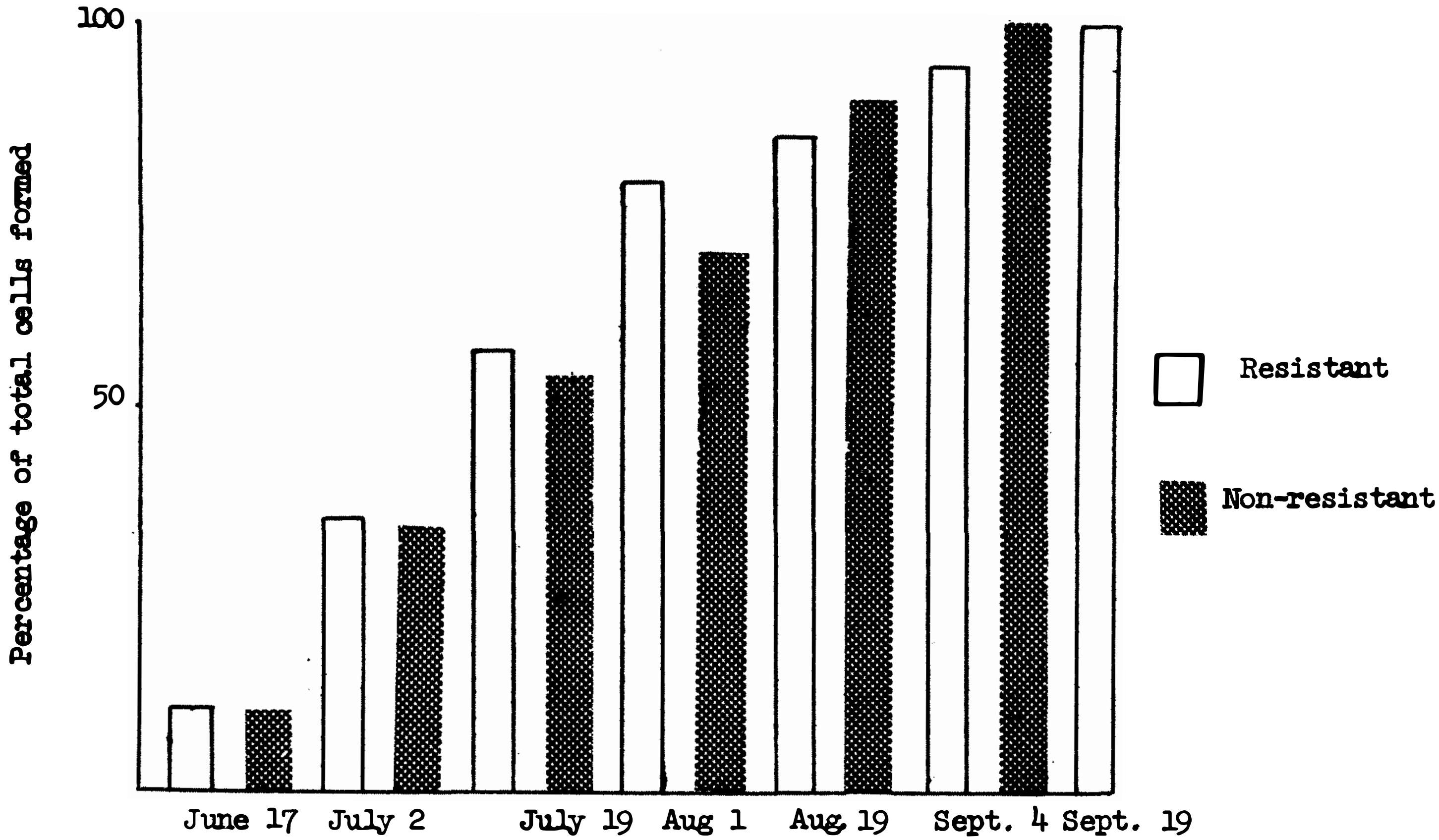

Fig. 12. Cumulative percentage of annual Radial growth for resistant and non-resistant lodgepole pine near Radium, British Columbia, 1968.

When growth was calculated in terms of number of cells per specified time periods, it was found that on the average, resistant trees produced a greater number of cells per period of time (Fig. 13).

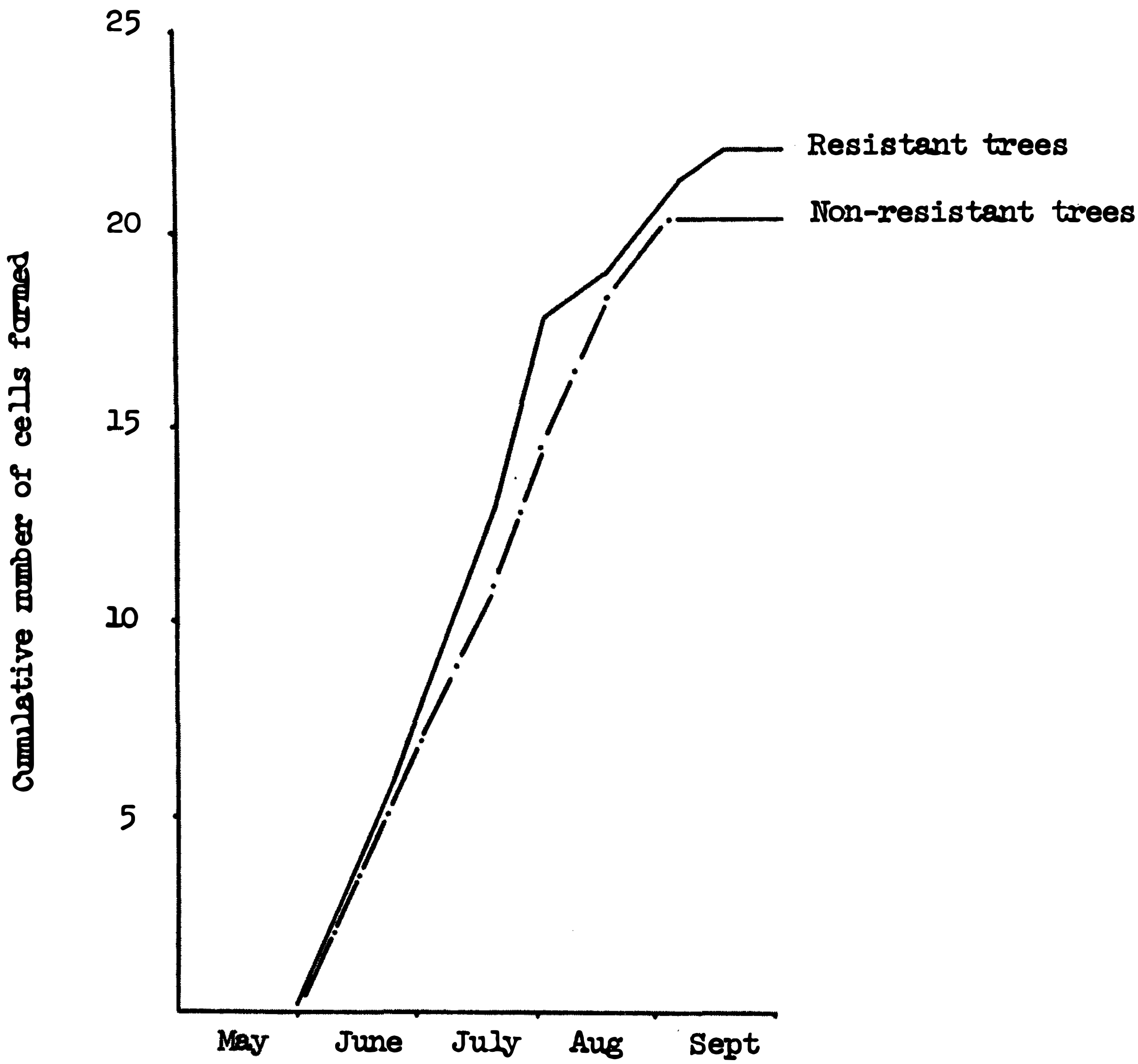

Fig. 13. Cumulative number of cells produced during the growing season by 10 resistant and a non-resistant lodgepole pine tree near Radium, British Columbia, 1968.

In 1969, water tension measurements were made on the 19 trees 3 September at 10:00 a.m. and at 4:00 p.m. on 22 September. On both occasions average tensions were lower by 5%, in resistant trees. These differences were not significant by 't' test. However, it is interesting to note the differences were consistent; and, the lower tensions were related with the thriftier trees, i. e. resistant trees.

DISCUSSION

Radial growth (cell division) was regular in both resistant and non-resistant trees throughout the season. No fluctuations or rhythm was recorded in spite of the occurrence of precipitation and temperature variations during the growing season. However, trees varied in their time of growth initiation, their rate through the season, and their time of growth cessation. It is recognized our methods of detecting those changes were imprecise and first symptoms, particularly of growth initiation, could include significant error. Nevertheless our observations did reveal real differences in these growth characteristics throughout the season between individual trees. Measurable growth in some trees, was completed in nearly half the time of others. There were also yearly differences. Resistant and non-resistant trees, however, were not distinguishable in terms of these parameters except that resistant trees tended to grow more rapidly. It appears unlikely that cell division, as it is reflected in radial growth, will be a useful diagnostic character for separating the two types of trees.

Possibilities appear good for development of a diagnostic character utilizing differences in degree of bole expansion. Wide and consistent differences were found between trees and were related directly to periods of dryness. Within a single stand on a uniform site having adequate moisture, non-resistant trees exhibited more bole contraction during dry periods than did resistant trees. It is interesting to note here that the peak beetle flight periods generally coincided with periods of maximum water stress and bole contraction (in late July and early August).

In addition, stands located in dry sites exhibited a more stable seasonal tension curve, and conversely less expansion and contraction than those stands located in wet sites. Further studies should be made of these relations.

Water tension measurements, by use of the pressure bomb, were not adequate alone to separate resistant and non-resistant trees on the same site. The two years in which those measurements were made, 1968 and 1969, were good years for growth; precipitation and temperatures being near optimum, and at no time were the trees severely stressed. Nevertheless, tensions were higher on drier exposed sites than on the more sheltered and cool sites with superior water supplies. The longer the period between rainfalls, the wider those differences became. Maximum tension during the growing season was found to coincide with the point at which expanding trees ceased to expand. We believe measurements by the pressure bomb are considered representative of the water condition in the tree. However trees must be "stressed" before separations can be made on a tree to tree basis. Hence in dry years, the pressure bomb will have the most potential usefulness in separating probable resistant and non-resistant trees.

Starch accumulation and hydrolysis is directly associated with the basic metabolism of the tree. The drought of 1967 appeared to effectively reduce the trees' ability to accumulate starch at least for the succeeding two years. It is known that starch is utilized by the tree as a source of energy, during periods of intensive respirational activity. Our methods of assessment, however, do not reveal any relation between starch and tree resistance.

LITERATURE CITED

Hall, R. C. 1944. A vernier tree-growth band. Journal of Forestry 42: 742-743.

Reid, R. W. and J. A. Watson. 1966. Sizes, distribution and numbers of vertical resin ducts in lodgepole pine. Can. J. Bot. 44: 519-525.

Reid, R. W., H. S. Whitney, and J. A. Watson. 1967. Reactions of lodgepole pine to attack by Dendroctonus ponderosae Hopkins and blue stain fungi. Can. J. Bot., 45: 1115-1126.

Walter, K. E. 1968. A new method for marking xylem. Forest Science, 14: 102-104.

Waring, R. H. and B. D. Cleary. 1967. Plant moisture stress: evaluation by pressure bomb. Science 155: 1248-54.

Reid, R. W., and H. S. Gates

1972. Relations between some physiological functions in lodgepole pine and resistance to the mountain pine beetle.

Information Report NOR-X-15;35 p.; Northern Forest Research Centre, Canadian Forestry Service, Department of the Environment, Edmonton 70, Alberta, Canada.

Reid, R. W., and H. S. Gates

1972. Relations between some physiological functions in lodgepole pine and resistance to the mountain pine beetle.

Information Report NOR-X-15; 35 p.; Northern Forest Research Centre, Canadian Forestry Service, Department of the Environment, Edmonton 70, Alberta, Canada.

Copies of this publication (if still in stock) may be obtained from:

Information Officer,
Northern Forest Research Centre,
Canadian Forestry Service,
Department of the Environment,
5320 - 122 Street,
Edmonton 70, Alberta, Canada.