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SEASONAL BEHAVIOUR OF VASCULAR CAMBIUM IN SOME HIMALYAN TREES. II. *QUERCUS LEUCOTRICHOPHORA* CAMUS EX BAHADUR

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An analysis of the influence of seasonal climatic variations on the structure, organisation and activity of vascular cambium of twigs as well as the main trunk in *Quercus leucotrichophora* showed that increased temperature and rainfall together with the burgeoing buds and young leaves augment it markedly, leading to the formation of distinct growth rings. The phloem differentiation for the new year starts from mid April onwards whereas the xylem elements whose cutting off is initiated in early April, gets completed by mid July, a few derivatives on the phloem side which had remained immature in the previous season acquired maturity in this season giving the impression that the phloem differentiation has already begun in the month of March. Cambium activity declines after August producing a few undifferentiated elements of winter pholem in November and December and these remain in this state up to the cambial initials also undergo dimensional changes coupled with the alterations in the ratio of fusiform and ray initials, throughout the year.

In the temperate regions, the tree growth is intermittent due to distinct seasonal variations in the environment. Plants growing under this type of climate adept themselves for their survival strategy. Evidently, the variation in the activity of cambium in the temperate trees is clearly demonstrated with the periodic production of wood and bark. This coincides with the climatic rhythm and results in the formation of annual rings in the wood and bark. Avila *et al.* (1975) have suggested that cambial activity or radial growth of the plant is the best parameter for such studies since it is genetically controlled and fixed through certain environmental limitations such as the climatic and soil factors and through certain internal influences such as physiological, structural and genetic factors (Fritts, 1976; Philipson *et al.* 1971). It has also been shown that two woody species of the chilean matorral, *Acacia caven* and *Proustia cuneifolia* which grew side by side, exhibited very different patterns of cambium activity (Aljaro *et al.* 1972). Almost indentical observations have been recorded by Faraggitaki *et al.* (1984) while studying the annual rhythm of cambial activity in two woody species of Greece, *Arbutus unedo* and *Quercus coccifera*, growing in close vicinity.

Keeping this in mind some temperate trees growing at Garhwal Himalayan hills were taken for study of the seasonal activity of cambium and its contiguous tissues. This paper deals with the results obtained by us on *Quercus leucotrichophora* (Fagaceae).

MATERIALS AND METHODS

Two mature specimens, growing in natural climatic and edaphic conditions at Kandolia forest (Pauri Garhwal, India) at an elevation of 1,800m MSL, Between 29°-20' and 30°-15'N latitude and 78°-10' and 79°-10'E longitude were selected for sampling. The rainy season extends here from May to September, with a few showers throughout the winter months also. October remains usually dry and from November till February, the weather is uncertain when limited rainfall occurs at higher peaks, coupled with severe cold winds which are a characteristic feature of the locality. The temperature increases slowly from February onwards, on the onset of spring season, remains more or less constant from June to October and declines again up to January (winter months). The highest sunshine is recorded in May and June and lowest in July when the rains are heavy (Fig. 4).

Monthly collections were made from the twigs of 2cm diameter in the third week of each month for two complete years. Simultaneously, after an interval of three months, small portions of the bark intact with wood, measuring 6cm^3 were also peeled off from the main trunk with the help of a chisel and hammer at the breast height. Subsequently, these larger pieces were cut into smaller ones and fixed in FAA or Craf.

Fig. 1A-D. Organization and structure of cambium (*fi* - fusiform initials: *n* - nucleus; *ric* - ray cell initials). A. Cambial zone showing non-storeyed arrangement of fusiform initials and beaded nature of its walls x 475. B-D. Same exhibiting transverse, oblique and radial divisions in the fusiform initials and nuclei of variable morphology from round, oval to elliptical. B x 470; C, D x 690.

Sections of the materials from the twigs as well as main trunk were cut in transverse and tangential longitudinal planes at 15-25 μm on Spencer's sliding microtome and stained with the tannic acid-ferric chloride- lacmoid combination (Cheadle *et al.*, 1953).

Cambial behaviour has been analysed on cross section of twigs by counting the number of undifferentiated layers, lying in between the secondary phloem and secondary xylem. The length and width of the

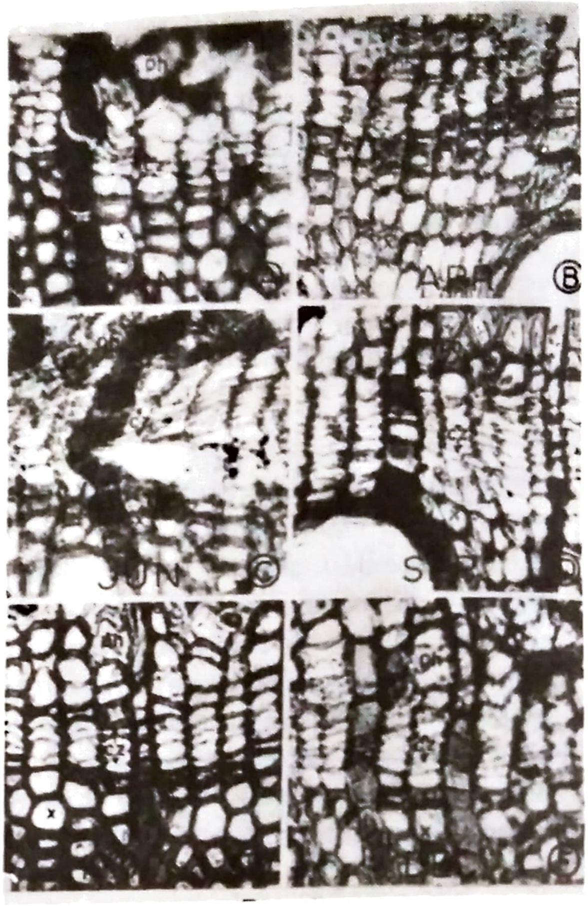

Fig. 2 A-F. Photomicrographs of t.s. of the twigs of different months of the year of bring out the seasonal behaviour of cambium through variations in the number of cambial layers (*cz* - cambial zone; *ph* - phloem; *x* - xylem). All x 540.

fusiform initials as well as height (vertical width) of ray initial strand and a ray cell initial were taken from the tangential longitudinal sections. The length and width of ray cell initial were measured randomly from the cross sections only. In order to estimate the area of fusiform initials in the cambium and number of ray initial strands/mm², camera lucida drawings have been made and plotted in the form of a graph. It was possible to group the ray initial strands into three categories, *viz.* short (1-6 cells), medium (7-12 cells) and tall ones (13-above cells high). Their frequencies are represented in the form of histograms, for different months of the year.

With a view to correlate the seasonal behaviour of the cambium with tree phenology and climatic changes, the morphological changes in the tree were recorded at the site while climate variations such as temperature, relative humidity, sunshine, rainfall were taken daily at the laboratory and at the collection site and average worked out.

OBSERVATIONS

Q. leucotrichophora is an evergreen species, commonly known as 'Banj'. It occurs rather gregariously, forming extensive, nearly pure stands or may be associated with *Rhododendron arboreum* and a variety of conifers. The leaves are alternate, lanceolate, dark green glabrous above and densely white or grey below. The leaves and flowers appear almost simultaneously in March or the latter process is somewhat delayed. The fruit formation and ripening require comparatively longer durations, exceeding even a year. Fruit sheding occurs in the month of December and extends up to February, which proved to be the months of maximum defoliation and bud formation. The defoliation of older leaves is also observed in September and October followed by the emergence of the flush of leaves.

Q. leucotrichophora has non-storeyed type of cambium, made up of the usual two types of cells - the fusiform and ray cell initials. The former are vertically elongated with overlapping, tapered ends, their wall have distinct primary pit fields (Fig. 1A-C), and possess dence cytoplasmic contents and prominent nuclei. The radial walls are thicker than tangential walls particularly during the dormant period and these become thin during active phase of cambium. The ray cell initials are short and more or less isodiametric or their length is slightly more than the width. The apical and basal ray cells of the strands are of different shapes and sizes. The ray initials are mostly organized into uniseriate rays but a few biseriate and triseriate strands were also observed in the cambium of the main trunks. Nuclei in the fusiform initials show variable shapes such as elliptical, oval and round (Fig. 1B,D). Both anticlinal and periclinal divisions have been observed in the fusiform initials. The former lead to an increment in the circumference of the cambium by oblique/pseudotransverse divisions occurring throughout the active period and the latter, periclinal division, is responsible for increase in girth through phloem and xylem differentiation.

The cambial zone between secondary phloem and xylem is not uniform throughout the periphery of the stem. In January it is composed of 1-4 cell layers whereas in the subsequent months cambial cells experience some stretching and gain an increase in the radial diameter. In April the cambial zone becomes broader (4-7 cells wide) and remains so up to July. Later these layers become fewer, up to November (3-5), followed by a constant zone of 2-4 layers in the winter months (December to February, (Fig. 2A-F).

The vessel expansion is initiated during early March, nearly two weeks before bud sprouting. In this span, a narrow band of large, early wood vessels are formed at the time of extension growth (leaf emergence - flower formation phase) in April and although these become functional prior to leaf expansion, the xylem differentiation continues till July. Phloem differentiation starts one or two weeks later (April) than xylem differentiation and continues up to early June. A second short flush of extension growth is witnessed in October and November also when a few vascular elements are cut off both on the phloem and xylem side, once again. A small fraction of these phloem elements are retained in the undifferentiated state which reach maturity in the beginning of the next year's spring (Fig. 3B).

It is indicated in Fig. 4, the fusiform and ray initials show dimensional changes in the year. The average length of the fusiform initials range between 369.40±46.43mm (June) and 503.84±63.36mm (September). It seems that both at the beginning and end of the cambial activity, these become elongated due to intrusive growth and become shorter during the active phase of cambium (June), evidently due to repeated divisions in them. On the other hand, the dimensions of the ray cell initials are higher at the time of augmented cambial activity. The width of the fusiform initials ranges between 10.64±1.92mm

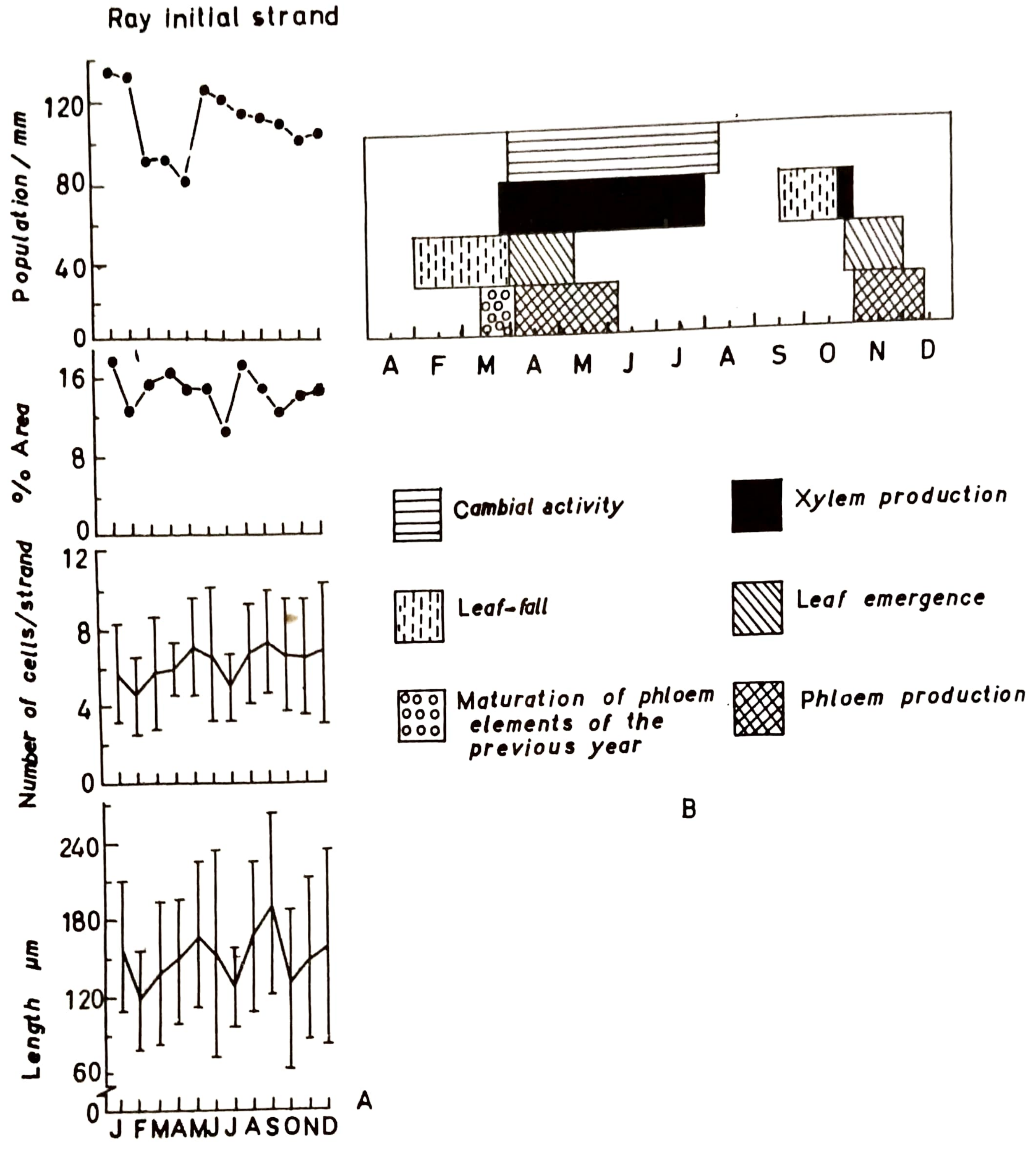

Fig. 3A, B.
A. Variations in the size, number of cells per strand, percentage area and population per mm2 of ray initial strands for different months of a year.
B. Relationship between the seasonal behaviour of cambium and its derivaties and the phenological features of the tree during a year.

(February) and 15.24±1.60mm (September). Their radial diameter was found to increase at the onset of spring and decline slowly at the time of cambial activity. The height of a ray cell is also less at the time of cambial activity in June (21.71mm) as against in January (27.04mm) when it is dormant. The length of ray initials varies between 8.85±1.86mm (February) and 11.60±2.64mm (June). The average height of ray

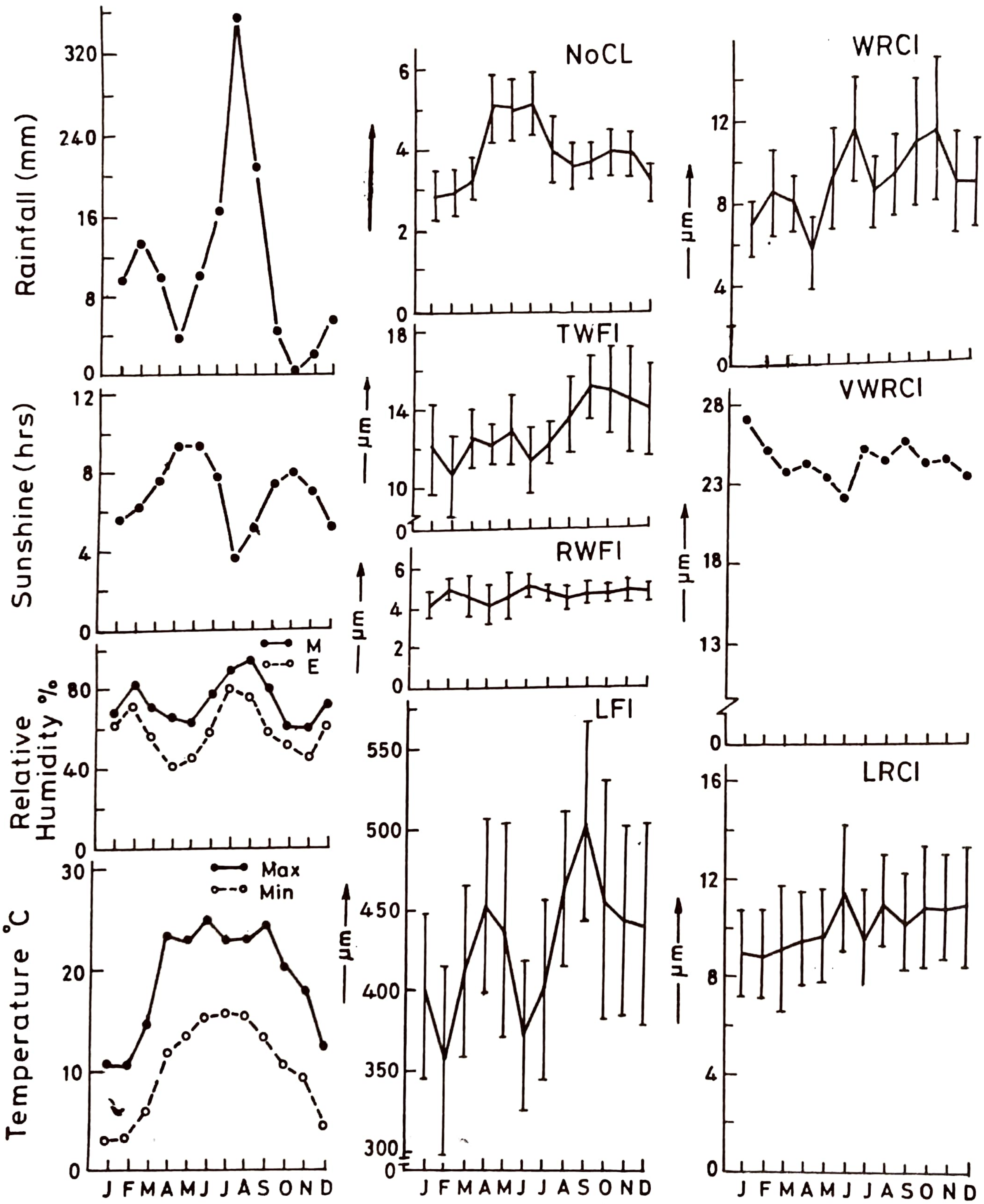

Fig. 4. A comparative analsysis of the meteorological data for Pauri (Garhwal) alongwith the number of cambial layers and dimensions of fusiform and ray cell initials for different months of the year. (*E* - evening; *LFI* - length of fusiform initials; *LRCI* - length of ray cell initials; *M* - morning; *Max* - maximum; *Min* - minimum No. *CL* - member of cambial layers; *RWFI* - radial width of fusiform initials; *TWFI* - tangential width of fusiform initials; *VWRCI* - vertical width of ray cell initials; *WRCI* - width of the ray cell initals).

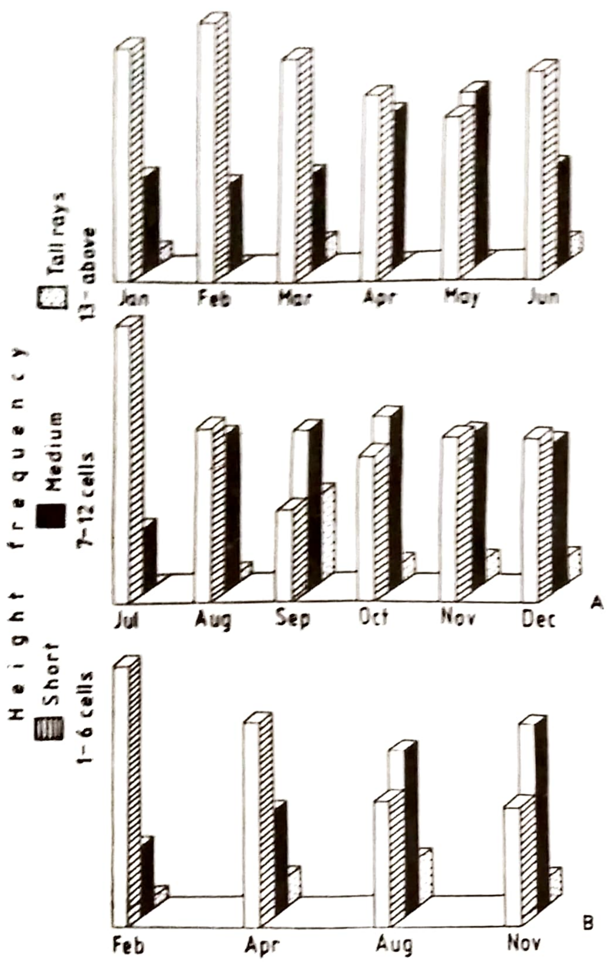

Fig. 5. Seasonal variations in the frequency of different height classes of the ray initial strands of the twigs (A) as well as the main trunks (B) for different months of the year. During the months of April to August the cell rays are fewer or absent.

strands during the active period of cambium shows a positive correlation with the variations in the dimensions of the fusiform initials (Fig. 3A, 5). Tall rays are very few or absent at the active phase of cambium in the twigs while these are present throughout the year, although their frequency is low as compared to the short and medium ray strands, in the cambium of the main trunks. The frequency of shorter rays is high in the first half of the year and then decrease to the identical value or slightly less to the frequency of the medium ray strands for the later half of the year, both in the twigs as well as the main trunks (Fig. 5A, B). The short rays (composed of 1-6 cells) contribute to 55.75%, medium rays (7-12 cells) 38.83% and tall ones (13 and above cells high), the remaining 8.13% of the total population of rays. The area occupied by ray initial strands and their number per mm^2 in the cambium vary throughout the year (Fig. 3A). On an average, the ray initials contribute to 14.42% of the area in the cambial zone and number of ray strands/mm^2 figured out to 110.14.

In the main trunk the maximum length and width of the fusiform initials has been recorded in August (518.70±76.32mm and 19.92±1.88mm), respectively. The minimum value for length and width of these was recorded during January (460.75±16.70mm x 14.28±2.36mm), the inactive phase.

DISCUSSION

The seasonal behaviour of cambium in *Q. leucotrichophora* suggested a highly positive correlation with the moisture status of the atmosphere since intense combial activity was seen when the latter was high alongwith the long photoperiod. On the other hand, Faraggittaki *et al.* (1984) have observed that the cambial activity in *Q. coccifera* growing in Greece, does not show such a correlation and in the evergreen Chilean and Californian shrub species, the cambium continues to cut off cells, throughout the year up to a certain extent even during the winter and severe drought periods and its maximum activity occurs during spring or in early summer (Avila *et al.*, 1975). That the rainfall has a direct bearing on the cambial activity by increasing water in the soil as well as the metabolic processes of the tissue was proposed by Glock & Agerter (1962) perhaps for the first time. Later Little (1975) stated that the water stress directly inhibits it by lowering the turgor pressure of the cells and indirectly by reducing the growth of leaves and supply of the hormones. In *Acacia raddiana* and *Zygophyllum dumosum* the cambial activity is also affected by water shortage (Fahn *et al.*, 1968; Waisel *et al.*, 1970). This condition holds true for the presently investigated taxon *Q. leucotrichophora* since the activity of cambium was seen to be augmented with the onset of rain and sprouting of new leaves and reaches the maximum level in June when the precipitation is highest.

Probing a little deeper, one finds that besides other parameters, the temperature may affect the cambial activity through its influence on the physiological processes also in the plants. Synthesis also contributes towards yet another indirect effect. In *Robinia pseudoacacia,* under controlled environmental conditions, the cambial activity is triggered by temperature and photoperiod (Waisel & Fahn 1965). Other workers like Priestley (1930), Kozlowski *et al.* (1962), and Reinders-Gouwentak (1965) have brought to light that the temperature determines reactivation of cambium, during the early part of the season in the temperate regions. In the hilly regions the day-length (sunshine) is yet another environmental factor which undergoes alteration with seasons. Similar observations have been recorded by Eggler (1955), Wareing (1958), Alvim (1964), Paliwal & Prasad (1970), Ghouse & Hashmi (1983) and Iqbal & Ghouse (1985). The presently investigated species brings to light identical conclusions since here the cambial activity increases with the increase in sunshine and temperature. Besides the above, Wareing (1958) has provided evidence to suggest that the increase in the ring width is attributable to the fall in temperature at high altitude alongwith the change of topography. The concept that a correlation between an external factor and the activity of cambium does not necessarily mean a simple relation between the two is now gaining ground (*see* Munting & Willemse, 1987).

Previous workers have reported that cambial reactivation progresses very rapidly or is simultaneous throughout the circumference in the ring porous trees (Priestley, 1935: Wareing, 1951; Ladefoged, 1952). Also the vessel expansion occurs in them more or less 3 weeks before bud-break, prior to the increase in the loss of water vapour that accompanies the expansion of the foliage (Dougherty *et al.* 1979), at a time when the water potential would be high. The auxin levels have been shown to be rather low in the cambium of ring porous species at this time (Aloni 1987). Also worthy of note is the suggestion of Wareing *et al.* (1964) that the rapid spread of cambial activity in ring porous species is due to the presence of tryptophan (an auxin precursor) in the bark, before bud-break. In *Q. leucotrichophora* an identical situation has been encountered and this fully supports the earlier propositions for similar (ring porous) taxa. On the other hand, in sycamore, a diffuse porous species, vessel expansion did not occur in the main stem until after bud-break (Denne & Atkinson, 1987), by which time rate of transpiration would be increasing and water potential declining.

Efforts have been made in the past to deduce the correlation between the size variations among the fusiform initials and cambial activity by alterations in the environmental factors in both the temperate (Bailey , 1920; Bannan, 1951; Catesson, 1974); and tropical trees (Paliwal & Prasad, 1970; Paliwal *et al.*, 1975; Iqbal & Ghouse, 1985). Usually the initials elongate with the onset of cambial activity and attain the maximum length, after the activity had reached its peak value, with some minor alterations. In the present study, the fusiform initials gain in length in the early growth period and then experience a decline in the month of June, due to frequent anticlinal divisions. Their average length later gains gradually and reaches a maximum value in September. Further fluctuations in the cell length during the subsequent months have also been observed. The maximum area occupied by the fusiform initials is 90% (in July) which is nearly equal to that suggested by Wilson (1963) and Kozlowski (1971) for temperate species.

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