

VASCULAR CAMBIAL ACTIVITY OF *ROBINIA PSEUDOACACIA* L. FROM TEMPERATE CLIMATE OF KASHMIR HIMALAYA

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ABSTRACT

Vascular Cambium, a meristem responsible for the formation of wood by way of growth and divisions of its components i.e. ray and fusiform initials were analyzed during different months over two consecutive years in order to assess their impact on growth in *Salix alba* from temperate climate of Kashmir Himalaya. The maximum and minimum dimensions of fusiform initials and ray initials were in winter and summer seasons respectively while the horizontal and vertical diameters of ray initials were maximum and minimum in summer and winter seasons respectively.

Key words: Vascular cambium, fusiform initial, ray initial, *Robinia pseudoacacia*

INTRODUCTION

Woody trees possess a power of secondary growth by which they increase in girth. This activity of secondary growth results in the formation of wood which by nature is secondary xylem constituting vessels, fibers, rays, parenchyma etc. All these structures are formed by a meristem called vascular cambium which is made up of two types of cells; fusiform and ray initials. The fusiform initials give rise to vertically oriented structures, while ray initials give rise to horizontally oriented structures. Diametric growth of a tree is accompanied by circumferential expansion of the cambium. The increase in girth of the cambium is due primarily to the increase in number of fusiform initials. In gymnosperms and most dicotyledoneae this increase is achieved by pseudo-transverse divisions. The production of new initials in the cambium by pseudo-transverse division far exceeds the number of initials required to accommodate the cambium's needed increase in girth. Keeping in view the above fact, an attempt

was made to assess the activity of vascular cambium of *Robinia pseudoacacia* from the temperate climate of Kashmir Himalaya as its wood is suitable for veneer, pulp, plywood, laminated wood, reconstituted wood products, artificial limbs, fruit boxes, agriculture implements, furniture; tool handles and sports goods.

MATERIAL AND METHODS

Study area

The territories forming Kashmir cover a wide area, mostly; snow-covered Himalayan Mountains girdling the valley, its altitude ranges from 1,600m (Srinagar) to 5,420m (Kolahoi) On an average, the climate of the Valley is sub-Mediterranean, with bixeric regimes, having two dry spells in June and September, and high precipitation during the cool season. The Kashmir valley enjoys an enchanting climate for the major part of the year. It has continental climate, marked by four well-defined seasons a year. (1) winter (December February), (2) spring (MarchMay), (3)

summer (June-August) and (4) September November). the data on the climatic factors were collected from the meterological department Rambagh Srinagar government of India.

The geographical location along with other characteristics of the selected site and material are summarised in Table 1:

Table 1. Characteristics of study site

Factor	Site (Khrew)
North Latitude	30° 12'
East Longitude	75° 35'
Elevation (m)	2000
Soil	Sandy clay
Age in years	10- 13

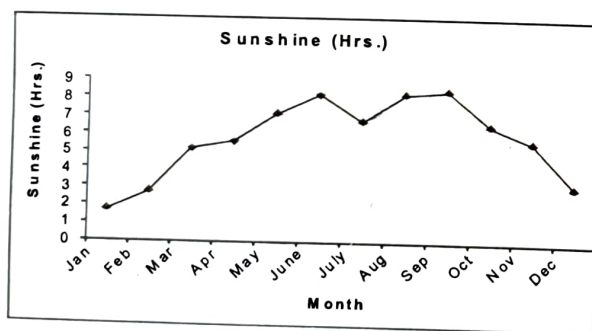


Fig. 1. Sunshine hours during different months of Year

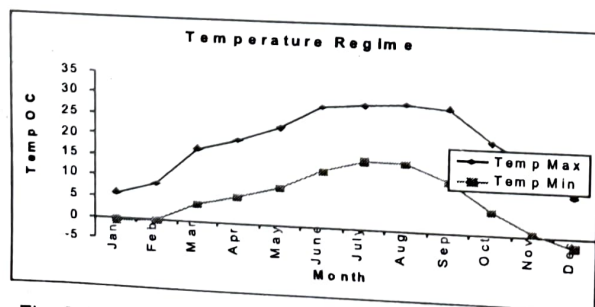


Fig. 2. Minimum and maximum temperature during the different months of year

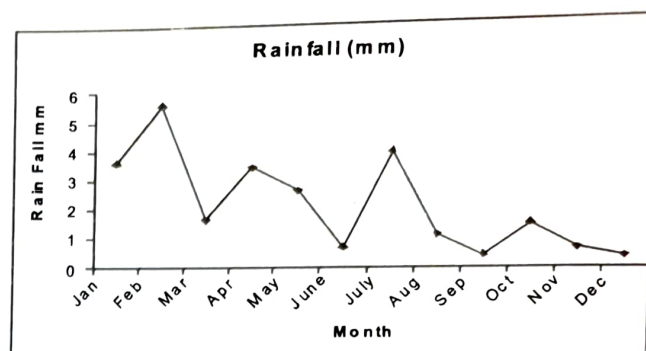


Fig. 3. Rainfall during the different months of year in mm

Cambial Characteristics

Field Study

To study the cambial characteristics (i.e. cambial activity in relation to season, twelve trees of *Robinia pseudoacacia* L., were selected from the natural habitats. Periodical monthly observations of cambial activity were made during two consecutive years (2005 and 2006). Cubical samples 2x2x2 cm comprising tissues along with outer bark and inner sapwood were chiselled out at breast height (1.3 m) from selected trees. Two trees were used on each occasion and no samples were collected from the same tree within three months. The samples were fixed in formalin Aceto-alcohol (F.A.A.) (Johansen 1940). After three days the material was transferred to 70% ethyl alcohol for preservation.

Laboratory Study

To assess the cambial activity 10-15 m thick tangential longitudinal sections were cut with Reichert sliding microtome and also manually with the help of sharp razor.

Staining procedure

For staining the section Foster (1934) and chedale (1953) method was followed. The section were first

put in 1% Tannic acid for 5-10 minutes and then washed 3-5 times with distilled water. After washing with distilled water, sections were transferred to ferric chloride for 5-10 minutes. Again sections were washed with distilled water 3 to 5 times. After that the section were transferred to sodium bicarbonate solution for 30 minutes, then sections were put in Lacomoid blue for 12 to 18 hours and after then washed with sodium hydrogen carbonate for few seconds. Now sections were transferred to clove oil for 10-15 minutes. After that they were transferred to different grades of alcohol and finally to Xylene in order to achieve complete dehydration and finally mounted on glass slides with Diphenyl Pthalte Xylene (DPX) as mountant.

Microscopy

To determine cell size, micrometry was done. The stages and Ocular micrometers were calibrated. A minimum of 100 randomly selected elements per block of each species were measured. The cell size was determined for each month after pooling all readings taken from relevant samples obtained during the two collection years as per Siddiqui (1991). The relative proportion of fusiform and ray initials was determined from Camera lucida drawings as per Ghouse and Yunus (1973). The ray height was classified into different categories viz. short (up to 300 m) medium (301-600 m) and tall (601 m onwards), while these of varying width as uni-bi-tri and multiseriate.

Statistical analysis

The data collected for present study was

statistically analyzed by using Sigma Plot 12.0 statistical software (SPSS. Chicago. IL.USA) and Minitab 11.0 for windows.

RESULTS

The Vascular cambium of *Robinia pseudoacacia* L. comprises of both fusiform and ray initials. The tapering ends of vertically aligned fusiform initials overlap one another at a same position rendering the cambium storied or stratified type. The cambium reactivates once a year after completing a definite period of rest. The cambial initials show certain changes in their size and content under diverse climatic conditions. The average length and width of fusiform initials vary from 144 μm (January) to 182 μm (August) and 16 μm (June) to 21 μm in (February) respectively. On whole the initials are shorter during winter than during the rest period of the year (Table 2). The average vertical and horizontal diameters of ray initials vary from 31-37 μm and 25-32 μm respectively. The fusiform initials divide pseudo transversely resulting into two initials. The ray initials also divide to increase their number. All this happens in order to cope with the expansion of the stem axis. New ray initials are also added by conversion of fusiform initials into ray initials through transverse, terminal or lateral segmentation.

Occasionally, fusion of two or more vertically aligned rays gives rise to exceptionally tall or wide rays. This is brought about by the conversion of the intervening fusiform initials into a group of ray initials, as this connects the already existing groups of the ray initials. The newly produced rays, with limited height in the beginning, grow into tall

structures by further divisions of the existing initials. At times, the fusiform initials intrude into a panel of ray initials, causing splitting of a broad or tall ray into several smaller rays (Fig.4 A and B). Depending upon the cambial makeup, the fusiform and ray initials in a tangential plane constituted 62 percent and 38 percent respectively. On the basis of

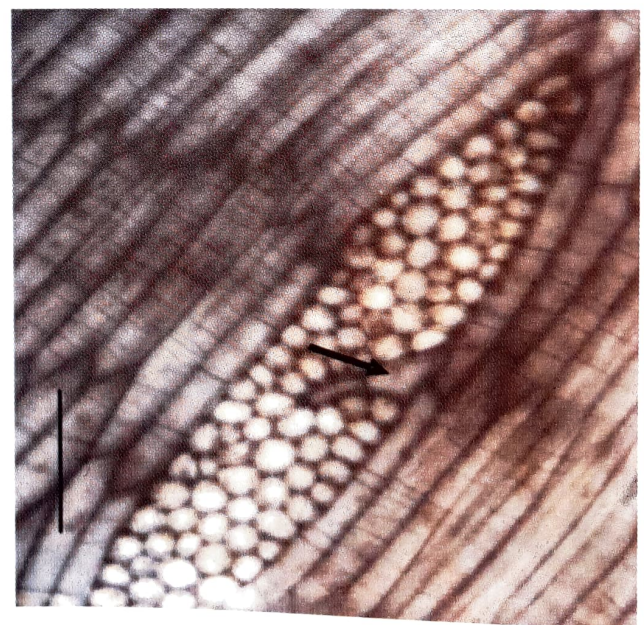
height the cambial rays classified into three groups viz., short (300 μm or below), medium (301-600 μm) and tall (above 601 μm). The rays of different height and width occur but tall cambial rays were predominantly present with average height and width of 1902 and 203 μm respectively.

Table 2. Dimensions of fusiform and ray initials (μm) in *Robinia pseudoacacia* during different months of year

Months	length of fusiform initials	Width of fusiform initials	Vertical diameter of ray initials	Horizontal diameter of ray initials
January	144	20	30	26
February	148	21	32	27
March	145	20	31	25
April	155	20	34	29
May	170	18	33	31
June	175	16	36	30
July	180	18	37	32
August	182	19	35	32
September	165	18	36	30
October	162	17	34	27
November	158	19	32	29
December	155	18	30	28
LSD ($P \leq 0.05$)	4.11	2.63	3.15	2.61
($P \leq 0.01$)	5.59	3.58	4.29	3.55



A



B

Fig. 4. (A) T.L.S. of cambium showing storied structure with septations and initiation of formation of ray initial. (B) T.L.S. of cambium of *Robinia pseudoacacia* showing splitting of cambial ray by intrusion of fusiform initial. Scale bar = 40 μm for A&B

DISCUSSION

The basic composition of vascular cambium is elongated fusiform initials and roughly isodiametric ray initials. Bailey (1923) has recognized two basic patterns of cambial structure; storied or stratified and non storied or non stratified. In former the fusiform initials occur in horizontal tiers with end of cells appearing approximately at the same level in a given tier, and in the latter the end walls of the adjacent initials overlap to a considerable extent. In the presently investigated species of *Robinia pseudoacacia*, arrangement of cambial initials depicts a clear stratified structure. The stratified structure of the cambium has been reported earlier by Khan (1977), Iqbal (1979), Khan (1980), Siddiqui (1983), Ajmal (1985), Kafeel (1986), Khan (2001), Khan *et al.*, (2005), Khan and Siddiqui (2007a), Venugopal and Liangkuwang (2007) and Wani and Khan (2008,2009).

Bailey (1923) while working on wide variety of temperate and tropical trees and concluded that cambial initials vary in length in non- stratified type from 460-4400 μm and are generally short in stratified type of cambium. The observations regarding this aspect indicate that in the present investigated species of *Robinia pseudoacacia*, the mean length of fusiform initials vary from 144- 182 μm , thus goes in agreement with result of Bailey (1923).

The anatomical variations which the cambium experiences during different seasons of the year have been analyzed by a number of workers like Eames and MacDaniels (1947), Esau (1965), Srivastava and O'brien (1966), Mahmood (2001),

Rensing and Samuel (2004) and Venugopal and Liangkuwang (2007). The radial walls of the fusiform initials have been reported to be usually thicker than the tangential walls, especially during dormancy and the primary pit fields appear deeply depressed in tangential longitudinal view giving a beaded appearance to the radial walls. Similar observations have been made in the present investigated species.

Cambial initials periodically undergo anticlinal and periclinal divisions Bailey (1923), Eames and MacDaniels (1947), Bannan (1956), Rao and Dave (1986), Han and Woong (1991), Mahmood (2001) and Khan (2001). The anticlinal divisions add to the cambial population while the periclinal ones increase the number of cambial derivatives emanating new phloem and xylem elements. Two fundamental types of anticlinal divisions have been recognized in the cambium of vascular plants. In one type, the anticlinal division occurs in a radial longitudinal plane and in the other pseudo-transverse wall formation takes place running a skew intersecting the two radial walls at two different levels as also reported by Philipson *et al.* (1971), Khan (1980), Khan and Siddiqui (1980), Zargoska-Marek (1984), Han and Woong (1991), Venugopal and Krishnamurthy (1989), and Mahmood (2001). In the present study the anticlinal divisions in the cambium have been noted to be radial longitudinal as it has been found in the majority of forms having stratified

Earlier works on ray initial formation indicate that the ray initials may originate in more than one way. Sometimes, they arise as a single cell which may be cut ends of fusiform initials as terminal segments

(Khan,1980; Siddiqui,1983). They may also arise either by transverse fragmentation of fusiform initials (Khan,1980; Siddiqui,1983; Mahmood, 2001) or a declining fusiform initial may reduce to a single ray initial (Barghoorn,1940, 1941a).

As for as the presently investigated species is concerned , it show the first two types of ray formation .After their development .they continue to increase in number to a considerable extent mainly through multiplication of the existing initials as has been reported by many authors (Barghoorn,1941a; Ghose and Yunus,1973; Khan, 1977; Khan,1980; Mahmood, 2001 ; Khan, 2001). In the presently investigated species, rays also showed increase in width and height by fusion of two or more vertically and radially aligned rays. Such fusions result from intervening fusiform initials or by multiplication of already existing ray initials of the adjacent panel of rays (Bartwal *et al.*, 1983; Rao, 1988). Splitting of rays also occurs as a result of intrusive growth of fusiform initials in the present investigated species, as it has also been reported by Khan (1980) in *C. citrinus*, *E. maculata* and *E. jambolana* and Khan *et al.*, (1983) in *C. sinensis*

Bailey (1923), while studying the structure of *Pinus strobus*, reported that the fusiform initials constitute about 87.5% of the total area of the cambial zone. Wilson (1963) calculated the surface area of the cambial zone of *Abies concolor* and found that the fusiform cells constitute more than 90% by volume of the cambium and its derivatives. In the present study the fusiform initials constitute 62% in *Robinia pseudoacacia* , of the

tangential area of the cambial cylinder which is much lower as compared to that of Bailey (1923) and Wilson (1963) observations, but are almost in accordance with the workers like Ghose and Yunus (1973), Khan (1977), Khan and Siddiqui (1980, 1983), Venugopal and Krishnamurthy (1989), Mahmood (2001), Khan (2001) and Wani and Khan (2008,2009).

CONCLUSIONS

The anatomical variations which the cambium experiences during different seasons of the year have been analyzed. The radial walls of the fusiform initials are usually thicker than the tangential walls, especially during dormancy and the primary pit fields appear deeply depressed in tangential longitudinal view giving a beaded appearance to the radial walls in the presently investigated species

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