

## Ontogeny of the Interfascicular Cambium in the Petiole of *Tabebuia rosea* DC. (Bignoniaceae)

Susy Albert\* and Jaysukhlal Jagmohandas Shah

Department of Botany, Faculty of Science, Maharaja Sayajirao University of Baroda, Vadodara 390002, India

**In order to elucidate the origin of interfascicular cambium in the petiole of *Tabebuia rosea*, transverse and tangential views of the cells in the interfascicular region during the different developmental stages of the petiole have been traced. Interfascicular cambium originates from the interfascicular parenchyma, which has been differentiated from interfascicular vascular meristem cells. Interfascicular parenchyma cells divide periclinally differentiating into the interfascicular metacambial cells and then into the cambium. Tangentially the homogenous structure of interfascicular parenchyma cells in the early stage gradually changes into a heterogenous structure with long and short cells from which fusiform and ray initials are derived, respectively. Ontogenetic pattern of the interfascicular cambium is similar to that of the fascicular one with interfascicular metacambium as an intermediate stage.**

*Keywords:* interfascicular cambium, ontogeny, petiole, *Tabebuia rosea*

There are two main views regarding the origin of interfascicular cambium. According to Esau (1943) and Soh and Choi (1995), interfascicular cambium in stem is a product of residual meristem or procambium, while some others suggest its origin from the interfascicular parenchyma (Catesson, 1964, 1974; Devdas and Beck, 1971; Butterfield, 1976; Phillips, 1976; Swamy and Krishnamurthy, 1980). According to Soh et al. (1989) in the hypocotyls of *Ricinus communis* the tissue from which the interfascicular cambium originates is structurally different from the parenchyma.

Whether the interfascicular cambium originates before or after the fascicular cambium is the second aspect of its origin. The commonly accepted view is that the interfascicular cambium originates after the fascicular cambium. In *Ricinus* the fascicular cambium originates before the interfascicular one (Fahn et al., 1972). In the stem of *Hoheria*, with the onset of secondary growth, interfascicular cambium originates from the parenchyma before the development of the fascicular cambium (Butterfield, 1976). However, these studies have dealt with the development of interfascicular cambium in the shoot or hypocotyls. The presence of interfascicular cambium and its development in the petiole have not been analyzed.

The present study aims to contribute to the understanding of the structural changes of the interfascicular tissue, in the vascular system, during the different developmental stages of the petiole in *Tabebuia rosea*, an evergreen tree of the family Bignoniaceae.

### MATERIALS AND METHODS

The petioles of *T. rosea* were collected from the

Botanical garden, M.S. University of Baroda. They were cut into small pieces and fixed in FAA solution (Johansen, 1940) immediately. The fixed materials were dehydrated in tertiary butyl alcohol series and embedded in wax (Wi et al., 2005). The embedded specimens were sectioned transversely and longitudinally at a thickness of 10  $\mu\text{m}$  with a rotary microtome (RM 2035; Leica Microsystems, Germany), and the sections were stained with Toliudine blue O.

### RESULTS

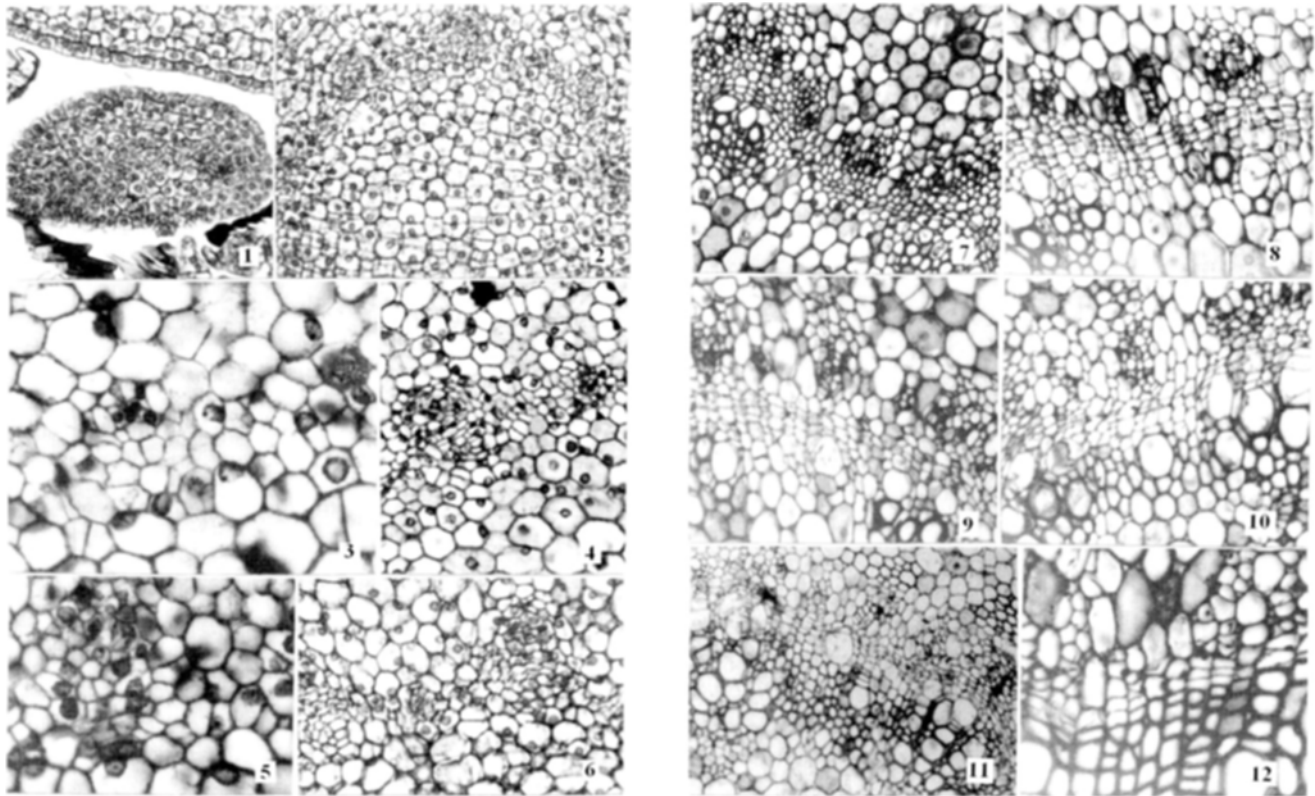
The development of interfascicular cambium has been traced in the different developmental stages of the petiole categorized on the basis of its length.

#### Transverse View

Primary vascular system in the petiole is initiated in the form of its meristem, the vascular meristem, which is noticed in the first leaf primordium. The vascular meristem is rather a template, i.e., a precursory stage of the petiole vasculature where leaf trace procambial strands will differentiate with reference to the leaf traces. The vascular meristem cells are homogeneously and densely stained and distinctly marked out from the surrounding vacuolated cells of the ground meristem (Fig. 1-1). In the youngest second leaf primordium, the vascular meristem shows 4-5 loci of cells actively dividing in varying planes (Fig. 1-2). These are the loci, which indicate development of leaf trace procambial strands. Cells of the vascular meristem, which are not committed to the procambial strand development, differentiate as the interfascicular meristem (Fig. 1-2).

The cells of the interfascicular meristem transectionally appear more or less polygonal, compactly arranged

\*Corresponding author  
e-mail drsusyalbert@rediffmail.com



**Figure 1.** 1, Leaf primordium at the first node showing densely stained vascular meristem. X400. 2, Leaf primordium at the second node showing densely stained procambial strands, intervening interfascicular meristem. X380. 3, Enlarged view of a leaf procambial strand and intervening interfascicular meristem. Note the interfascicular meristem cells compactly arranged without intercellular spaces. X769. 4, Petiole at the third node showing young vascular strand and intervening parenchyma cells. X208. 5, Enlarged view of a developing vascular strand with rectangular procambial cells and the interfascicular parenchyma. X700. 6, Young vascular strands with intervening interfascicular parenchyma cells. X210. 7, 2 cm long petiole. Interfascicular parenchyma cells showing periclinal division. X200. 8, 5 cm long petiole. A complete metacambial cylinder formed of 3-4 radially flattened metacambial cells. X380. 9, 5 cm long petiole. New metaxylem elements developing from metacambial cells. X187. 10, Xylem parenchyma cells associated with new xylem elements lignified. X187. 11, 8.5 cm long petiole. Late metacambial cylinder. X187. 12, Vascular cambium in a mature petiole showing 2-3 layers of fusiform cells intervened by ray cells. X344.

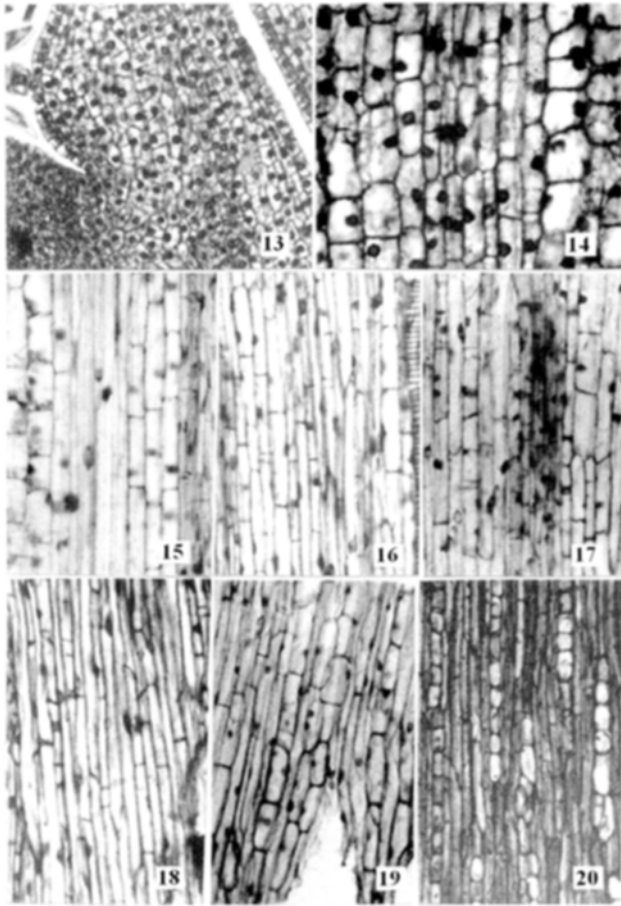
and each of them with a large prominent nucleus (Fig. 1-2). They are easily distinguishable from large and more or less rounded vacuolated cells with intercellular spaces belonging to the pith and cortex. The procambial strands appear as groups of small, actively and irregularly dividing polygonal or rectangular densely stained cells (Fig. 1-2, 1-3).

At the third node of the shoot apex petiole and lamina are distinguished. Vascular strands are discrete with protoxylem and protophloem and the interfascicular meristem cells differentiate into parenchyma cells (Fig. 1-4, 1-5, 1-6). The interfascicular parenchyma differentiated from interfascicular vascular meristem are round or oblong, smaller in size and are compactly arranged when compared to the large pith and cortex cells having intercellular spaces. At this stage procambial strands are still present in the vascular meristem (Fig. 1-4). The procambial cells within the strands are rectangular/isodiametric in shape.

At any given level of the leaf development in *T. rosea*, the developmental status of the vascular strands varies from one to the other. Nonetheless, the general devel-

opment of the strands has been studied and each strand passes through almost similar stages. In a 2 cm long petiole vascular strands show a zone of cambiform cells (cambium like) termed as metacambium, a term coined by Larson (1976), a transition stage between procambium and cambium (Susy and Shah, 1998). At some loci, periclinal divisions are evident in the interfascicular region and they are tangentially continuous with the 2-3 layered metacambium of the vascular strand (Fig. 1-7). Thus a tangential advancement of periclinal divisions occurs from the fascicular region to the interfascicular region. As these cells appear similar to the metacambium of the strand and as they are present between the vascular strands, the term "interfascicular metacambium" has been used to designate the periclinally dividing cells in the interfascicular region.

Interfascicular metacambium becomes more distinct in a 5 cm long petiole with 3-4 layered radially flattened cells in radial seriation and in continuity with the band of metacambial cells in the vascular strand and phloic strand to form a complete metacambial cylinder (Fig. 1-8, 1-9). New xylem elements develop opposite



**Figure 1.** Continued. **13**, Densely stained vascular meristem cells in the first leaf primordium. X300. **14**, Elongated procambial cells with large conspicuous nucleus. X400. **15**, Interfascicular parenchyma in the petiole at the third node. Note the cortical cells with transverse end walls. X240. **16**, Periclinal divisions in the interfascicular parenchyma cells of 2 cm long petiole. X240. **17**, Elongated interfascicular metacambial cells with long and short cells. X240. **18**, 5 cm long petiole. Interfascicular metacambial cells showing long and short cells. X240. **19**, 8.5 cm long petiole. Late interfascicular metacambium. X252. **20**, Fusiform and ray cells in a mature petiole. X161.

to the phloic strands in the interfascicular region (Fig. 1-9). These elements are considered to be metaxylem elements because they appear simultaneously with the metaxylem elements formed from the metacambium cells in the vascular strand. These elements can also be termed as late metaxylem elements because they appear after the differentiation of protoxylem and early metaxylem in the vascular strands. Thus the differentiation of primary xylem occurs in two stages. Initially a number of protoxylem and metaxylem elements develop within the strand. These are gradually linked into a complete cylinder of primary xylem by the differentiation of the interfascicular metacambial derivatives. This was another additional criterion to coin the term interfascicular metacambium.

Gradually, the parenchyma cells associated with xylem elements undergo lignification (Fig. 1-10) gradu-

ally forming a complete cylinder of xylem as seen in 8.5 cm long petiole (Fig. 1-11). In a mature petiole (one which has ceased elongation), 2-3 layers of radially flattened fusiform cells of the vascular cambium are interrupted by the elongated ray cells (Fig. 1-12). The identity of individual vascular strands is not observed, but a complete vascular cylinder is observed.

### Longitudinal View

Longitudinally the vascular meristem cells in the first leaf primordium appear densely stained, polygonal and are well demarcated from the large cortical and pith cells (Fig. 1-13).

In the second leaf primordium where procambium strands are distinct, the procambial cells are elongated with a large conspicuous nucleus (Fig. 1-14). Cells of the interfascicular meristem are also elongated with a conspicuous nucleus but are more lightly stained than the procambial cells.

Parenchyma cells differentiated from interfascicular vascular meristem cells show transverse end wall with a prominent nucleus (Fig. 1-15). Cortical cells are short and broad with transverse end walls (Fig. 1-15).

These interfascicular parenchyma cells elongate with the elongation of the petiole (Fig. 1-16) and in a 2 cm long petiole, where metacambium is well developed within vascular strands periclinal divisions are noticed in the interfascicular parenchyma cells (Fig. 1-17). Some of the cells elongate and have their one end wall oblique.

In a 5 cm long petiole, the metacambium of the vascular strands and the interfascicular metacambium show the long and short cells (Fig. 1-18). Long cells show tapering or transverse end walls and short cells show transverse end walls.

In an 8.5 cm long petiole, the metacambium in micromorphology, assume characteristics more resembling those of the vascular cambium (Fig. 1-19). The long cells later differentiate as fusiform cells and the short ones, as ray cells.

In a mature petiole, the vascular cambium with two distinct systems of cell is seen, i.e., the long fusiform cells with oblique or fusiform end walls and they appear in long axial files (Fig. 1-20). Rays are uniseriate or biseriate.

### DISCUSSION

When procambial strands are first discernible in the vascular meristem of the leaf, they appear as discrete groups or islands of cells in transverse sections. The cells placed adjacently between and interconnecting the procambium strands have been referred to as interfascicular residual meristem (Larson, 1975). This region has been referred to as the interfascicular vascular meristem in the petiole of *T. rosea*.

According to some authors (Butterfield, 1976; Esau, 1977; Swamy and Krishnamurthy, 1980; Fahn, 1982) the interfascicular meristem is a secondary meristem, i.e., it is a derivative of differentiated interfascicular parenchyma through the resumption of meristematic activity. Siebers (1971) suggested that a direct ontogenetic continuity exists between procambium and interfascicular cambium. Some investigators have also suggested that the interfascicular cambium originates from the dedifferentiation and redifferentiation of the interfascicular parenchyma and the xylem and phloem elements derived are all secondary in nature (Catesson, 1964; Devdas and Beck, 1971; Phillips, 1976). Eames and Mac Daniels (1947) and Catesson (1964) have argued that formation of cambial strips takes place in the interfascicular areas only in line with those strips of fascicular sectors and that too under the influence of fascicular cambium, which originates earlier. According to Soh et al. (1989) in the hypocotyls of *Ricinus* the tissue from which interfascicular cambium originates is structurally different from the parenchyma. In the petiole of *Tabebuia* when the primary vascular system is well established, the vascular strands are discrete with interfascicular parenchyma. Many cells of the interfascicular vascular meristem are incorporated in the developing and enlarging procambial strands. The interjacent or buffer cells, lying between these strands which do not contribute to the development of fascicular tissue become highly vacuolated and differentiate as interfascicular parenchyma.

Ontogeny of the fascicular cambium reveals an intermediate stage of metacambium between procambium and cambium. Metacambium is an advanced stage of procambium in which procambial cells undergo frequent periclinal divisions forming a distinct radial seriation of cell layers extending across the entire strand to form a tangentially continuous zone. It is during this stage of the petiole that periclinal divisions are apparently noted in the interfascicular parenchyma. Successively the vascular strands are bridged by periclinal divisions in the interfascicular parenchyma that advance acropetally and laterally in line with the fascicular metacambial cells. The resultant derivatives have been termed as the interfascicular metacambial cells. The increased tempo of the cell division activity results in the formation of a continuous metacambial cylinder of cells.

Cambie (1967), Soh (1974a, b) and Butterfield (1976) reported the procambium in shoots of some species forming an almost complete cylinder from its early or late appearance. They do not, however, indicate the presence of the intermediate metacambial stage. Larson (1976) who coined the term metacambium has reported periclinally dividing parenchyma cells bridging the primary vascular strands in the seedlings of *Populus* but he has not designated them as interfascicular metacambium. It is for the first time that interfascicular metacam-

bial stage has been recognized during the development of interfascicular cambium in the petiole of *Tabebuia*. The interfascicular parenchyma therefore by dedifferentiation and redifferentiation gives rise to the interfascicular metacambium, which later differentiate as the cambium composed of fusiform and ray cell system.

The common view regarding the time of origin of the interfascicular cambium is that the fascicular cambium precedes the interfascicular cambium. Fahn et al. (1972) presume that the interfascicular cambium is first to arise, although belated in commencing activity, thereby influencing the formation of fascicular cambium. According to Butterfield (1976) and Soh et al. (1989), it originates following the onset of the secondary growth or on the completion of the organ elongation. The primary vascular strands are discrete during early metacambial development. At a later stage, the vascular strands are bridged by periclinal divisions in the interfascicular parenchyma that advance acropetally and laterally in line with the fascicular metacambial cells, in this development regular divisions usually occur earlier in the strands than in the interfascicular sectors.

Thus in *Tabebuia* there is a lateral spread or advance of periclinal divisions from the fascicular to the interfascicular region. Consequently, the interfascicular cambium is delayed in development when compared with that of the fascicular cambium.

## ACKNOWLEDGMENT

I would like to thank the Head, Department of Botany, The MS University of Baroda, Vadodara for providing me with all the facilities required to conduct the study.

Received August 3, 2005; accepted April 10, 2006.

## LITERATURE CITED

- Butterfield BG (1976) The ontogeny of the vascular cambium in *Hoheria angustifolia* Raoul. *New Phytol* 77: 409-420
- Catesson AM (1964) Origine, fonctionnement et variations cytologiques saisonnières du cambium de l'*Acer pseudo-platanus* L. (Aceracées). *Ann Sci Nat Bot* 5: 229-498
- Catesson AM (1974) Cambial cells, In AW Robards, ed, *Dynamics Aspects of Plant Ultrastructure*. McGraw Hill, New York, pp 358-390
- Cumbie BG (1967) Development and structure of the xylem in *Canavalia* (Leguminosae). *Bull Torrey Bot Club* 111: 42-50
- Devdas C, Beck CB (1971) Development and morphology of stelar components in the stems of some members of the Leguminosae and Rosaceae. *Amer J Bot* 58: 432-446
- Eames AJ, MacDaniels LH (1947) *An Introduction to Plant Anatomy*. Ed 2, McGraw Hill, New York

- Esau K (1977) Anatomy of Seed Plants. John Wiley and Sons, New York
- Fahn A (1982) Plant Anatomy. Ed 3, Pergamon Press, Oxford
- Fahn A, Ben-Sasson R, Sachs T (1972) The relation between the procambium and the cambium, In AKM Ghouse, M Yunus, eds, Research Trends In Plant Anatomy. McGraw Hill, New Delhi, pp 161-170
- Johansen DA (1940) Plant Microtechnique. McGraw Hill, New York
- Larson PR (1975) Development and organization of the primary vascular system in *Populus deltoidees* seedlings. Amer J Bot 63: 1332-1348
- Phillips IDJ (1976) The cambium, In MM Yeoman, ed, Cell Division in Higher Plants. Academy Press, New York, pp 347-390
- Siebers AM (1971) Initiation of radial polarity in the interfascicular cambium of *Ricinus communis* L. Acta Bota Neerl 20: 211-220
- Soh WY (1974a) Early ontogeny of vascular cambium: II. *Aucuba japonica* and *Weigela coracensis*. Bot Mag 87: 17-32
- Soh WY (1974b) Early ontogeny of vascular cambium: III. *Robinia pseudoacacia* and *Syringa oblata*. Bot Mag 87: 99-112
- Soh WY, Choi PS (1995) Development and ultrastructure of interfascicular cambium in stem of *Ginkgo biloba* seedling. J Plant Biol 38: 281-288
- Soh WY, Kang KD, Yang WY (1989) Ontogeny of the interfascicular cambium in the seedling roots of *Acer saccharinum* L. Kor J Bot 34: 311-316
- Susy A, Shah JJ (1998) Early ontogeny of vascular meristem in the petiole of *Gmelina arborea* (Verbenaceae) and *Tabebuia rosea* (Bignoniaceae). Phytomorphology 48: 187-194
- Swamy BGL, Krishnamurthy KV (1980) On the origin of vascular cambium in dicotyledonous stems. Proc Indian Acad Sci 89: 1-6
- Wi SG, Chung BY, Kim JH, Baek MH, Yang DH, Lee JW, Kim JS (2005) Ultrastructural changes of cell organelles in *Arabidopsis* stem after gamma irradiation. J Plant Biol 48: 195-200