SECONDARY GROWTH IN THE STEM OF SOME SPECIES OF ALTERNANTHERA AND ACYRHANTHES ASPERA (AMARANTHACEAE)

by

Kishore S. Rajput & K.S. Rao

Department of Biosciences, Sardar Patel University, Vallabh Vidyanagar 388 120, Gujarat, India

SUMMARY

Secondary growth in Achyranthes aspera, Alternanthera polygamous, A. pungens, A. sessilis, and A. triandra was achieved by the development of a cambial variant resulting in successive rings of xylem and phloem. Each new cambium was developed at a distance about two to three cells external to the phloem produced by the previous cambium. The development of phloem was not synchronous in the species studied. Phloem development started either simultaneously with xylem or after the formation of a few xylem derivatives. In Achyranthes, xylem production started first followed by the development of phloem. Phloem mother cells differentiated into sieve tube elements, companion cells and axial parenchyma. Xylem was storied and exclusively composed of axial elements. Radial elements were absent in all the xylem rings of the stem. Vessels were angular and mostly solitary, but radial and tangential multiples were also observed occasionally. Xylem fibres were nonseptate and nucleated. Development of phloem and the rayless nature of the xylem is discussed.

Key words: Amaranthaceae, cambial variant, phloem, rayless xylem.

INTRODUCTION

There has been growing interest in the cambial variant of different groups of plants (Kirchoff & Fahn 1984; Lotova & Timonin 1985; Timonin 1988; Philipson 1990; Larson 1994; Rao & Rajput 1998; Rajput & Rao 1998; 1999a). Cambial variants, previously called anomalous secondary thickening, are known to occur in the stem of dicotyledons but in most instances the peculiarity is confined to some members of an otherwise normal family (Balfour 1965). However, the family Amaranthaceae is well known in the literature for its secondary thickening which is abnormal in all the investigated cases. Opinions have differed on the mode of formation of this secondary growth (De Bary 1884; Pfeiffer 1926; Joshi 1937; Metcalfe & Chalk 1950; Balfour 1965; Timonin 1988; Rajput & Rao 1999a). The cambium of the Amaranthaceae is considered to be functionally unidirectional (Balfour 1965; Philipson & Ward 1965).

Although the pattern of secondary growth in the Amaranthaceae has been studied earlier (De Bary 1884; Dastur 1925; Joshi 1931a, b, 1934, 1937) the occurrence of rayless xylem has not been reported in the species studied. Therefore, the present investigation reports the functionally bi-directional nature of the cambium and occurrence of rayless xylem in four species of Alternanthera and Achyranthes aspera.
Fig. 1. Tangential longitudinal (A) and transverse (B–E) views of cambium, xylem and phloem of *Alternanthera*. – A: Cambium showing storied arrangement of fusiform cambial cells and absence of rays in *Alternanthera pungens* (× 124). – B: Stem of *Alternanthera polygamous* showing three successive rings of xylem (× 50). – C: Newly developing cambium in *Alternanthera triandra* showing bidirectional differentiation of xylem and phloem; an arrowhead
MATERIALS AND METHODS

Four to eight internodal segments of the main stem measuring 3–20 mm in thickness were collected twice (i.e. in August and March) from ten plants of *Alternanthera polygamous*, *A. pungens*, *A. sessilis*, *A. triandra* and *Achyranthes aspera* (Amaranthaceae) growing at Bhorkheda in North Maharashtra, and on the M.S. University campus at Baroda. Samples were immediately fixed in FAA (Berlyn & Miksche 1976) and processed by routine methods to obtain transverse, tangential and radial longitudinal sections of 15–20 μm thickness. Sections were stained either with tannic acid - ferric chloride-lacmoid combination (Cheadle et al. 1953) or with safranin-fast green combination (Johansen 1940) and 4% acetocarmine for localization of nuclei in fibres.

To measure the length and width of xylem fibres and vessel elements, small pieces of stem were macerated with Jeffrey’s fluid (Berlyn & Miksche 1976) at 55–60 °C for 24–36 hrs. The length and width of sieve elements and fusiform cambial cells were measured directly from the tangential longitudinal sections. Mean values of the elements were calculated from one hundred random measurements taken for each element using an ocular micrometer scale.

RESULTS

Structure and development of cambium

The cambium was storied and exclusively composed of relatively short fusiform cambial cells varying 215–287 μm in length in all the species studied (Fig. 1A). In transverse view, the cambium appeared two- to three-layered when non-dividing and three- to six-layered during the development of xylem and phloem. In the species of *Alternanthera* the cambial rings were not always continuous and often appeared as overlapping segments, leading to the union and anastomosing of the adjacent rings. However, no such anastomosing and overlapping of cambial rings were observed in *Achyranthes aspera*.

Developmentally each cambial ring was divided into two distinct alternative segments, the segments of the cambium giving rise to conducting elements of xylem and phloem, and other segments producing only xylem fibres centripetally and phloem parenchyma centrifugally. Like other dicotyledons, the first cambium formed a complete cylinder and differentiated bi-directionally producing xylem inside and phloem outside. Later it ceased to divide and a new ring of cambium developed at a distance about two to three cell layers external to the phloem produced by preceding vascular cambium (Fig. 1C). Those two to three layers of parenchyma mature into conjunctive tissues.

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indicates already differentiated sieve elements (× 350). – D: Outermost cambium surrounded by xylem and phloem (× 350). – E: Development of one sieve tube element and two axial parenchyma strands from a single phloem mother cell (arrow); an arrowhead indicates one sieve tube element and one axial parenchyma cell formation from a single phloem mother cell (× 580).
Structure and development of secondary phloem

The secondary phloem of all the species was composed of sieve tube members, companion cells and axial parenchyma whereas rays were absent. In transverse view, the phloem parenchyma contained the largest cells and single companion cells occur-
ring at the margin of each sieve tube member were the smallest cells. Sieve tube members possessed a simple sieve plate with minute sieve pores on their slightly oblique to transverse end walls. Lateral sieve areas were observed on both radial and tangential walls of the sieve tube members. Variable amounts of callose deposition were observed on the sieve areas of functional sieve elements, whereas heavy accumulation was seen on non-functional ones. Compared to fusiform cambial cells, the average length and width of sieve tube members were slightly less. The length was 248–270 μm in *Achyranthes aspera* and 198–218 μm in *Alternanthera sessilis*. The width of sieve tube members varied from 13–20 μm in different species; however, it was highest (16–20 μm) in *Alternanthera polygamous* and lowest (13–18 μm) in *Achyranthes aspera*.

Development of secondary phloem was not synchronized in all the species of *Alternanthera* whereas in *Achyranthes* xylem production always preceded that of phloem. In *Alternanthera* phloem development occurred either simultaneously or after the formation of a few xylem derivatives (Fig. 1C, D). This variation was seen not only in the different species but even in the same species. In some of the cambial segments phloem and xylem development occurred simultaneously while in other segments phloem developed after the formation of 4–6 xylem derivatives (Fig. 1D). The rate of cell division towards the phloem side was very slow. The ratio of xylem to phloem production ranged from 6:1 to 9:1.

The phloem mother cells underwent further divisions and differentiated into one sieve tube mother cell and an axial parenchyma cell each or directly differentiated into axial parenchyma cells (Fig. 1E). Development of conducting elements of xylem and phloem was restricted to certain segments of the cambium whereas other areas of the cambium formed only thick-walled xylem fibres (conjunctive tissue) centripetally and axial parenchyma centrifugally. But in *Alternanthera sessilis* and *A. polygamous* some of the segments of the outermost ring also showed formation of sieve tube elements centrifugally where it produced only thick-walled xylem fibres centripetally.

**Structure of xylem**

The stems of all the species were composed of successive rings of xylem alternating with phloem (Fig. 1B, 2A). The rings were not always complete but often appeared as overlapping segments. The secondary xylem was composed of vessels, tracheids, nucleated xylem fibres and axial parenchyma while rays were absent (Fig. 2B, C). Vessels were either solitary or in radial multiples of 2 or 3. They possessed alternate bordered pits with a simple perforation plate in transverse to slightly oblique end walls. The length and width of vessel members differed from species to species and was found maximal in *Alternanthera polygamous* (189–205 μm and 45–69 μm) and minimal in *Achyranthes aspera* (147–163 μm and 39–58 μm), respectively. Xylem fibres retained their living protoplast and nucleus even after the development of secondary walls (Fig. 2D). The nuclei were oval to oblong and fusiform measuring from 5–9 μm in length and 3–4 μm in width among the species studied. Fibre length did not show much variation but was found longest in *Achyranthes aspera* (507 μm) and shortest in *Alternanthera pungens* (472 μm).
DISCUSSION

The pattern of secondary thickening in Chenopodiaceae, Nyctaginaceae and Amaranthaceae deviates from that of many dicotyledons investigated (Balfour 1965; Fahn & Zimmermann 1982). Much attention has been paid to the question whether each growth increment of secondary vascular tissue arises from a separate cambium layer or from a residue of previous cambium (Balfour 1965; Philipson & Ward 1965; Baird & Blackwell 1980; Fahn & Zimmermann 1982). However, the composition and development of their derivative tissues remained neglected (Rao & Rajput 1998; Rajput & Rao 1998; 1999a). Development of the secondary thickening meristem in Alternanthera sessilis and Achyranthes aspera has been studied thoroughly by De Bary (1884), Dastur (1925), and Joshi (1931a, b, 1934, 1937), but the occurrence of nucleated xylem fibres and absence of a radial system in xylem and phloem remained unreported.

In Alternanthera, with the cessation of activity of a cambial ring, secondary growth is either succeeded by another complete ring of cambium or by a large cambial arc. However, at some places cambium arises only to the outer side of phloem strands of the preceding cambium and, to complete the ring, it becomes joined laterally with the pre-existing cambial segment that produces only xylem fibres.

In all members of the Amaranthaceae, cambia are reported to be functionally uni-directional (Balfour 1965; Philipson & Ward 1965). In the present investigation, all the species showed bi-directional differentiation of xylem and phloem. A similar observation has also been made in Pupalia lappacea (Rajput & Rao 1999a), a member of the same family. Among all the species investigated the time of xylem and phloem differentiation is not necessarily synchronized, phloem development may start simultaneously or after the development of few xylem derivatives. However, the lower production of phloem elements as compared to xylem gives a false impression of a functionally uni-directional cambium (Rajput & Rao 1999a).

Rayless xylem has also been reported in some genera and is restricted to a small portion of dicotyledons (Gibson 1978; Carlquist 1988; Lev-Yadun & Aloni 1995; Rao & Rajput 1998; Rajput & Rao 1999a). Raylessness may be total, i.e., rays are not formed at all during the entire life span of the plants, or it may be temporary, i.e., raylessness is restricted to the juvenile wood (Lev-Yadun & Aloni 1995). All the species in the present investigation showed the absence of rays even in the samples collected at the end of the growing season (i.e., in March). Rayless xylem has also been reported in some members of the Amaranthaceae (Metcalfe & Chalk 1950; Rajput & Rao 1999a). According to Gibson (1978), raylessness in dicotyledonous wood is most frequently observed in species with reduced cambial activity, short fusiform initials and highly specialized cell types in secondary xylem. These characteristic features occurred in all the species studied.

It has been considered that rayless xylem tends to occur in groups of plants in which normal cambial activity is lost during the course of evolution towards a herbaceous mode of structure (Carlquist 1988). Furthermore, raylessness may represent a way of increasing mechanical strength in short-lived stems, in which the selective
value for radially oriented parenchyma is minimal. All the species of *Alternanthera* have prostrate stems which later bend upwards while *Achyranthes* is an erect herb with spreading branches.

Fibres with living protoplast are considered to be an adaptive feature, exhibiting transition forms towards parenchyma cells prevailing in the herbaceous plants (Fahn & Leshem 1963, Rajput & Rao 1999b). Occurrence of nucleated fibres may be associated with the rayless nature of the stem and the fibres may be functioning both as mechanical and storage tissues (Rajput & Rao 1999a, b).

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### REFERENCES


