A STUDY OF SCLEREIDS IN THE FERTILE PARTS OF TWO MEMBERS OF CUPRESSACEAE

BY A. R. RAO AND MANJU MALAVIYA (MISS)

(Botany Department, Lucknow University, Lucknow)

Received July 23, 1966

(Communicated by Prof. L. Narayana Rao, M.A., Ph.D., F.A.SC.)

ABSTRACT

The structure, distribution and ontogeny of sclereids in the male and female cones have been found to be very similar in eight species of Cupressus, namely C. macrocarpa, C. goveniana, C. funebris, C. lusitanica, C. knightiana, C. sempervirens, C. torulosa, and C. lindleyi and in some Juniperus species, i.e., J. chinensis, J. formosana, J. procera, and one unidentified species. There are no sclereids in the vegetative parts. The sclereids exhibit polymorphism. The ontogeny of different types of sclereids is the same in all the species. A parenchyma cell in the ground tissue of the female cone scale or in the microsporophylls differentiates as a sclereid initial, develops branching, undergoes "secondary sclerosis" and results in the formation of a thick-walled sclereid. The taxonomic importance of these sclereids is also discussed.

INTRODUCTION AND TECHNIQUES

The present is the ninth paper in the series on a study of sclereids in Indian conifers. Earlier papers have recorded the occurrence of sclereids in the fertile as well as the vegetative parts (Rao and Malaviya, 1963, 1964 a and b, 1965 a and b). In the present paper are included some observations on sclereids, which occur only in the sporophylls and not in the vegetative parts of the below-mentioned eleven species of Conifers, belonging to the sections Cupressineae and Juniperneae of the Cupressaceae. All the specific names used in this paper have been taken from the labelled museum specimens and checked from A Handbook of Coniferae by Dallimore and Jackson (1948). The following are the species investigated:

Cupressus macrocarpa Hartweg
   C. goveniana Gordon.
   C. funebris Endlichehr.
   C. lusitanica Miller.
Sclereids in Fertile Parts of Cupressaceae

C. *knightiana* Rehder.
C. *sempervirens* Linnaeus.
C. *torulosa* Don.
C. *lindleyi* Klotzsch.

*Juniperus chinensis* Linnaeus.
*J. formosana* Hayata (= *J. fermidane*).
*J. procera* Hoehstetter.

*Juniperus* species (unidentified) from Apharwart.

The material of young male and female cones of *Cupressus funebris* was kindly sent by Dr. B. M. Johri. All the other species were taken out from labelled specimens in the departmental museum. The mature cones were found very hard for sectioning, hence thick hand sections were cut and cleared by Foster's technique (1946, 1964 lecture series). One percent safranin in a mixture of \( \frac{1}{2} \) absolute alcohol and \( \frac{1}{2} \) xylol was used for staining as described by Foster. Alcoholic safranin and Haematoxylin-orange-G were used for microtome preparations.

**Distribution and Structure**

Sclereids were found only in the micro and megasporophylls of *Cupressus* as well as *Juniperus* and exhibit polymorphism. All the three well-known types of sclereids of Tschirch (1889), *i.e.*, the brachy, osteo and astrosclereids are met with. The ontogenetic development of all these types is more or less identical in all the species studied. In fact the nature of the sclereids\(^1\) and their distribution pattern is exceedingly similar in all of them. But attention is drawn to the differences in the sclereid structure when noted at the generic or specific levels. The two genera are being described separately to bring out the differences and for convenience of illustrations.

*Cupressus*

Sclereids are absent in the leaves and stems of all the species investigated. In *C. macrocarpa*, *C. funebris* and *C. goveniana*, sclereids are present in the male and female cones. In *C. knightiana* and *C. lusitanica* they are found in the male cones, while the female cones could not be investigated for want of material. In *C. torulosa* and *C. lindleyi*, the fertile parts were not available at all, hence they could not be examined. All the figures have been drawn from *C. funebris* and the distribution and structure of sclereids appear to be the same in all the species. In the female cones, the sclereids occur in a
diffuse manner, in large numbers in the sterile tissues of the scales (Figs. 1 and 2, Photos 1 and 2). In the male cone, the sclereids however, are confined mostly to the lower sterile sporophylls (Figs. 3 and 4). In these they occur in a diffuse manner in the ground tissue as well as near the sporangial wall. In the upper fertile sporophylls, sclereids may or may not occur next to the sporangial wall but the ground tissue does not show any sclereids.

All the three well-known types of sclereids found in the female cone show a great deal of variation in form. The rounded or oval, roughly isodiametric forms (Fig. 5 and Photo 1) showing slight variation in outlines can be referred to the category of brachysclereids. The elongated forms with either truncated, blunt or bifurcated ends (Figs. 6–11) fall into the group of osteosclereids. The majority of sclereids exhibit various modes of unequal branching, dichotomy and curving of the branches (Figs. 12–14). These may be accommodated under astrosclereids.

In the male cones, the sclereids are generally smaller in size and less branched than the ones in the female cone. Most of them fall under the category of brachysclereids (Figs. 15–17). A few of the sclereids which show some branching and bifurcation of branches (Figs. 18–20) are the astrosclereids. The elongated types found mostly next to the wall layers with either pointed or sometimes branched ends (Figs. 21–25) belong to the category of osteosclereids.

The secondary wall of the adult sclereid is highly lignified, thick and lamellated (Fig. 26) with pit-canals running through the entire thickness of the secondary wall. A characteristic feature of the sclereids of *Cupressus* is that most of them contain a persistent nucleus in the lumen of the adult sclereid and also some disorganised remains of the cell protoplast. Many of the elongated types show two or even three distinct nuclei in the lumen (Photo 2). But the nucleus is generally absent in the adult sclereids of *Juniperus* which serves as a distinguishing feature between the two genera.

**The Ontogeny of Sclereids in Cupressus**

All the three types of sclereids in the species studied are transformed ground parenchyma cells of the sporophylls of the male as well as the female cones undergoing subsequent “secondary sclerosis” of the wall. In the female cones of *Cupressus*, a group of sclereid initials may be recognised from the surrounding tissue with their dense, coarsely granular protoplasm, devoid of any vacuoles and one or two distinct nuclei each with a nucleolus (Fig. 27). Initials may also be seen at the same time or a little earlier in the cone-axis (Fig. 28). Out of these, some initials develop into the brachysclereids of
one type (Figs. 29-31). There is a general increase in size (Figs. 29-31) and this is accompanied by the secondary wall formation. Other initials develop into a rounded type of brachysclereid (Figs. 32 and 33).

The osteosclereids develop by an excessive elongation of the initial cell (Figs. 34-38) whose contents generally migrate towards their elongated, curved or bifurcated ends. The elongation of the sclereid generally ceases, as soon as the secondary wall is laid down. The lignin deposition takes place in layers, resulting in a highly lamellated secondary wall. The osteosclereids may grow at both ends so as to result in a very narrow sclereid with pointed and bifurcated ends (Fig. 39). Formation of highly lignified secondary wall, renders the sclereid very conspicuous against the surrounding background of thin-walled cells, specially when stained with a phloroglucin-concentrated hydrochloric acid stain (Foster, 1949).

The astrosclereid initial very soon starts sending out small tubular arms in the surrounding intercellular spaces (Fig. 40). These branches grow by intrusive growth (Fig. 41), they increase in size and the contents generally migrate into them. Figure 42 shows another type of young astrosclereid. Further development is similar to that of the other two types. Sometimes an assemblage of different types of sclereids developing simultaneously may be seen (Figs. 43-45). Whenever a sclereid does not find sufficient space in one direction, it takes a curve and changes its direction of growth (Fig. 45). The tendency of these sclereids is to grow in any direction where they meet no resistance. The ultimate form of the sclereid thus seems to depend to some extent, on the nature of the surrounding tissue and the ability of the sclereid initial to grow through these. This has already been discussed earlier while dealing with the sclereids of Araucaria (Rao and Malaviya, 1964 b).

The initials in the male cone also develop like those of the female cones. The size of the initial is smaller and it is more elongated near the wall region (Fig. 46) than in the ground tissue of the sterile sporophyll (Fig. 47). The initial enlarges and develops very soon (Fig. 48) into a brachysclereid, or it may show lobed form (Fig. 49). Other initials may send out small branches (Figs. 50 and 51) or may simply elongate (Figs. 52 and 53). In the male cone also brachy and osteo or astrosclereids develop together in the same sporophyll, at different places (Fig. 54).

Summing up the ontogeny, it is noted that basically the trend of sclereid development is the same in all the three types of sclereids. There is an increase in the size of the initial which is either immediately followed by deposition of secondary wall layers, or there is the formation and elongation of small
tubular branches, which retain the capacity to grow and push their way in the surrounding tissue for sometime. Secondary wall formation follows after the branches have stopped growing and an osteo or an astrosclereid is formed.

**Distribution, Structure and Ontogeny of Sclereids in Juniperus**

The distribution pattern of sclereids in *Juniperus* more or less follows a similar course as in *Cupressus*. The sclereids occur in a diffuse manner in the outer sporophylls of the female cone (Fig. 55), in the ground tissue of the basal sterile sporophylls and the cone axis (Figs. 56 and 57). In the male cone the sclereids occur in small numbers in the few sterile basal sporophylls and in larger numbers in the upper sporophylls (Fig. 58). In the microsporophyll the sclereids are mostly confined to the basal broad portion by which the scale is attached to the axis (Fig. 59). In the male cone of *Juniperus* however, the sclereids are not confined near the sporangium wall as in *Cupressus*. The brachysclereids show various forms, *i.e.*, rounded, lobed, and oval (Figs. 60-66). The elongated bone-shaped osteosclereids have either blunt, or pointed ends (Figs. 67-76). The astrosclereids include forms with branched and curved ends, also with very irregular shapes (Figs. 77-85). From the figures 60-85 it becomes quite evident that the sclereids of *Juniperus* exhibit more variety in form than those of *Cupressus*.

The secondary wall in all sclereids of *Juniperus* is very thick, lamellated, lignified with numerous pit-canals (Fig. 86). Unlike the sclereids of *Cupressus* the adult sclereids of *Juniperus* are devoid of nuclei and any residual protoplast or secondary wall substance (Fig. 86).

The development of all these sclereid forms basically follows the same course as in *Cupressus*. The initials however, do not have very dense cell contents and the protoplast is very much scattered giving the appearance of small vacuoles in the lumen (Figs. 87 and 88). The latter development of these sclereids is just like those of *Cupressus*.

**Discussion**

Sclereids are altogether absent in the vegetative organs of all the species investigated but occur in the fertile parts. Except for some minor differences sclereids of *Cupressus* and *Juniperus* are similar in their features. This sclereid similarity constitutes another close affinity between these two genera in addition to other morphological and anatomical similarities. At the same time minor differences like the presence of nuclei and some disorganised contents in the adult sclereids of *Cupressus* separates this from *Juniperus*. It will
thus be seen that details of sclereid character, if carefully noted, may furnish distinguishing characters to separate allied genera.

The above studies on the sclereids of *Cupressus* and *Juniperus* and also those of *Podocarpus* (Rao and Malaviya, 1965 b), *Taxus* (Rao and Malaviya, 1965 a) and *Picea* (Rao and Malaviya, 1967) have brought out some points of general interest. The distribution pattern of sclereids though varied in stem and leaf, is uniform in fertile parts. The ontogeny too does not deviate much from the basic pattern. In the microstrobilus, sclereids are present mostly in the sterile sporophylls not in the fertile ones and there are no sclereids in the sporangial tissue. In the megastrobilus sclereids are present in the cone axis, sterile sporophylls, body of seed scale and in the seed integument. These are mostly of the brachysclereid type, with very few osteosclereids. In spite of this uniformity, the structural differences of sclereids in the fertile parts and the form and distribution pattern of sclereids in the vegetative parts varies in the different genera of conifers. This serves as an aid to distinguish them from each other.

**Acknowledgement**

We are very grateful to Prof. B. M. Johri for supplying us the material of young male and female cones of *Cupressus funebris*. We are also thankful to the University Grants Commission and the Council of Scientific and Industrial Research respectively for financial aid.

**References**

Dallimore, W. and Jackson, A. N.  

Foster, A. S.  


Rao, A. R. and Malaviya, M.  


Rao, A. R. and Malaviya, M.  


**EXPLANATION OF PLATE VIII**

**PHOTO 1.** A part of the longitudinal section of the female cone of *C. funebris* showing the sclereids, ×8.

**PHOTO 2.** A few sclereids magnified in the female cone scale of *C. funebris* showing the persistent nuclei, ×153.7.

(gt, ground-tissue; n, nucleus; ov, ovule; scl, sclereid.)

**EXPLANATION OF TEXT-FIGURES**

**Figs. 1–39.** Fig. 1. Diagrammatic sketch of a longitudinal section of the female cone-scale showing the sclereids, ×6. Fig. 2. A portion of the cone scale magnified, ×50. Fig. 3. Diagrammatic sketch of a longitudinal section of the male cone, ×6. Fig. 4. Magnified view of the sterile sporophyll of the male cone, ×40. Fig. 5. A brachysclereid, ×64. Figs. 6–11. The osteosclereids, ×64. Figs. 12–14. The astrosclereids, ×64. Figs. 15–17. The brachysclereids of male cone, ×64. Figs. 18–20. The astrosclereids from the male cone, ×64. Figs. 21–25. The osteosclereids from the male cone, ×64. Fig. 26. A macerated sclereid magnified, ×200. Figs. 27–39. Various stages in the ontogeny of sclereids in the female cone showing the development of brachy and osteosclereids, ×148.

(ca, cone-axis; gt, ground tissue; i, initial; l, lumen; n, nucleus; pc, pit-canal; pt, protoplast; rc, resin-canal; scl, sclereid; sp, sporangium; sw, secondary wall; wa, wall.)

**Figs. 40–88.** Figs. 40–45. Different stages in the ontogeny of astrosclereids in the female cone, ×148. Figs. 46–54. The developmental stages of sclereids in the male cone, ×148. Figs. 46–88. *Juniperus* Fig. 55. Diagrammatic representation of the tangential longitudinal section of the female cone showing the distribution of sclereids, ×8. Figs. 56–57. Magnified portions of the cone scale and cone respectively showing the diffusely scattered sclereids, ×32. Fig. 58. Diagrammatic representation of the longitudinal section of the male cone showing the sclereids, ×8. Fig. 59. Lower part of longitudinal section of the male cone showing the detailed structure, ×32. Figs. 60–66. Various forms of brachysclereids, ×60. Figs. 67–76. The osteosclereids, ×60–Figs. 77–85. The astrosclereids, ×60. Fig. 86. A mature sclereid showing the detailed structure, ×140. Figs. 87–88. Female cone scales enlarged to show the detailed structure and the initial stages in the development of sclereids, ×32.

(gt, ground tissue; i, initial; n, nucleus; nc, nucellus; ov, ovule; pc, pit-canal; pt, protoplast; scl, sclereid; sw, secondary wall; wa, wall.)