

A CONTRIBUTION TO THE NODAL ANATOMY OF *AZIMA TETRACANTHA* LAMK.

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Salvadoraceae is a small family comprising only three genera, viz., *Azima*, *Dobera* and *Salvadora* and about eight species, occurring in Tropical Africa, Mascarene Islands, Arabia, India and certain Islands of the East Indies. There has been a wide divergence of opinion among phylogenists about the systematic position of the family among the systems of angiosperms. Wettstein (1935) included this family in the order Celastrales under the class Choripetalae. A similar treatment has been followed by Hutchinson (1959) recently. Engler and Diels (1936) also included the family Salvadoraceae in their class Archichlamydeae but under the order Sapindales. More or less similar treatment of the family is found in the works of Gundersen (1950) and Takhtajan (1959). On the other hand, Bentham and Hooker (1873-76) and Bessey (1915) considered the family Salvadoraceae as belonging to the Sympetalae under the order Gentianales and most closely related to the families Oleaceae and the Loganiaceae. During recent years, there has been a general realization of the fact that in determining the correct phylogenetic position of a taxon, it is the totality of evidence from all aspects of the study of the plant which must be considered. In view of this, a monographic study of this family has been undertaken before summarizing its phylogeny and natural relationships. Earlier, one of us (Tiagi, 1954) studied the embryology of *Salvadora persica* and concluded that the Salvadoraceae belongs to the Choripetalae. In the present work, the authors have described the occurrence of a unilacunar, double-trace node and its modifications in *Azima tetracantha*.

Material and Methods

Vegetative and flowering material of *Azima tetracantha*, fixed in 70% alcohol was obtained through the kind courtesy of Dr. K. S. Manilal of Kerala University, Calicut (India). Young branches comprising two successive nodes and three internodes were cleared by heating in lactic acid or alternatively in 10% solution

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of potassium hydroxide. In addition, young stems were embedded in paraffin and serially sectioned through two to four nodes. Sections were stained with crystal violet and erythrosin with satisfactory results.

Observations

External Morphology.—*Azima tetracantha* is a low, spinous, highly branched bush, woody below but with pale green, herbaceous, almost quadrangular young branches. The leaves are in opposite to subopposite, decussate pairs. They are shortly petiolate, about 2×4 cm long, entire, elliptic, acute, sharply mucronate, rigid, pale green with an acute base. Usually, there are two laterally placed spines in the axil of a leaf (Figs. 1-2). The spines which morphologically represent the first pair of leaves of the axillary shoot are about three cm long, more or less, triangular in cross section, very sharp and with an indurated apex. The plant is dioecious. The flowers are borne in the axils of leaves. Generally, there is a cyme of three flowers in the axil of a leaf which in the upper branches, especially of the male plants become greatly reduced or even completely suppressed. In female inflorescences, the subtending leaf and the two laterally placed spines are mostly well developed (Fig. 2). The female flower is tetramerous and possesses four sepals of which two are medianly placed and the other two laterally placed; the four petals are diagonally placed and the four staminodes inner to them are antesealous. The gynoecium is bicarpellary, the ovary unilocular below but bilocular above. In each locus, there is an anatropous, crassinucellate, bitegmic ovule on an axile-basal (really parietal-basal) placenta; the micropyle is constituted by both the integuments. The two carpels which are medianly placed very often show an anticlockwise twisting of about 45 degrees when viewed from above. The stigma is almost sessile, globose and bilobed.

The male flower is built upon the same plan as the female flower and possesses a pistillode in the centre.

Vascularization of the Leaf.—A transverse section of an internode is, more or less, quadrangular in outline. Its eustele consists of a ring of about twenty-four, conjoint-collateral, open, endarch vascular bundles. A study of serial transverse sections through two or more nodes (Figs. 3-23) of the stem reveals that a transverse section through the middle region of an internode shows the differentiation in its eustele of the vascular supply of two successive pairs of leaves above it. The two opposite groups of two vascular strands each which are subjacent to the pair of leaves just above are slightly larger than those subjacent to the next higher pair of leaves (Fig. 3). Thus, each leaf is vascularised by two separate vascular strands that are related to a single gap two internodes down in the eustele. In other words, *Azima tetracantha* possesses a unilacunar, double-trace node. It may be mentioned here in

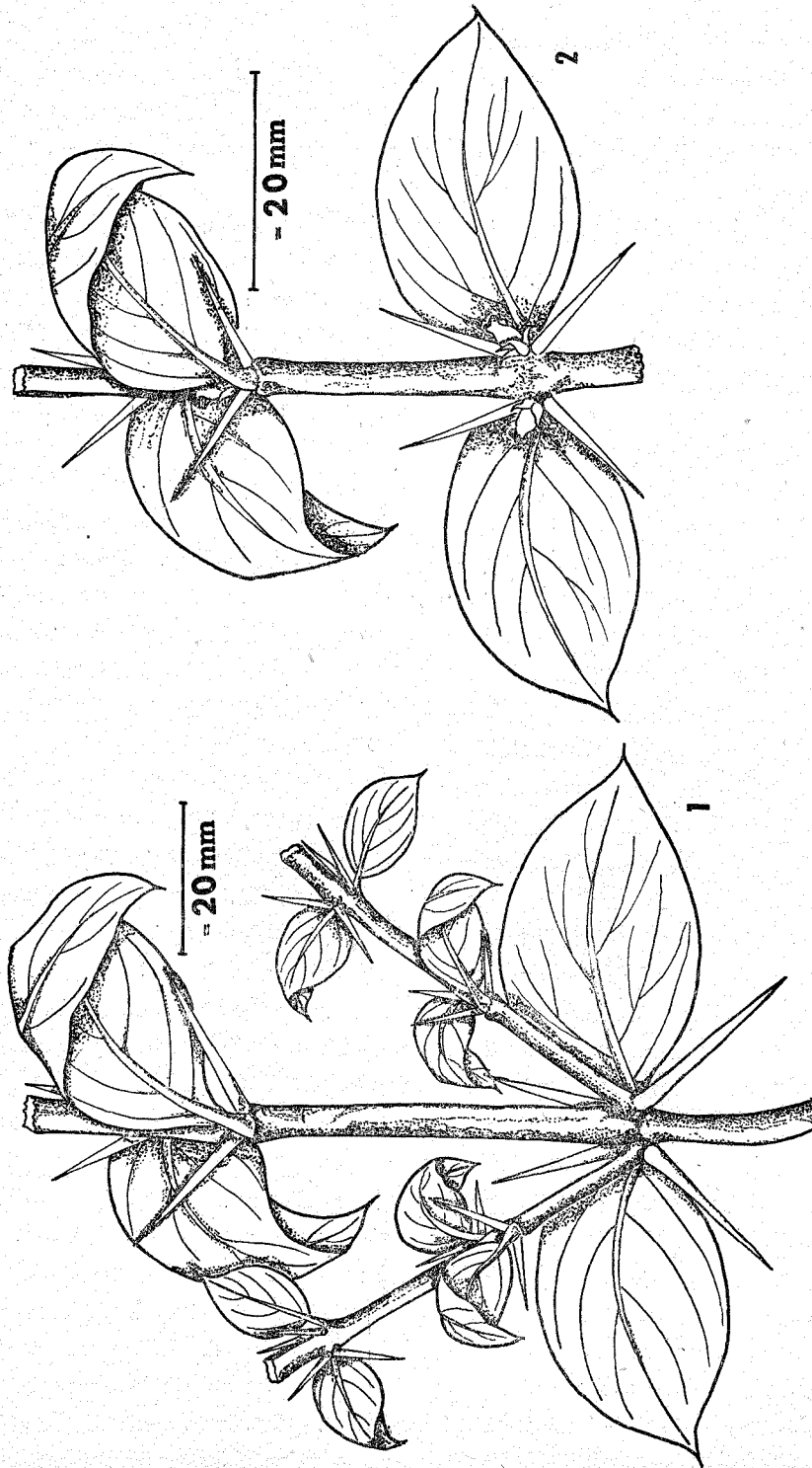


Fig. 1. A vegetative twig showing the axillary branches and the laterally placed spines at their bases.
Fig. 2. A reproductive branch from a female plant; note the axillary flower, flanked on either side by a spine.

parenthesis that excepting these four pairs of vascular strands which constitute the vascular supply of the two successive pairs of leaves above, the other vascular bundles may increase or decrease in number by radial divisions and fusions all along the length of the internode as a result of which the number of vascular bundles in a cross section of an internode may be more or less than twenty-four.

At the level of a leaf base, its two vascular traces diverge out from the eustele of the stem, become laterally united to form a single arc-shaped vascular strand which enters the leaf base and then ascends upward in the petiole. However, at places along the length of the petiole, the lateral fusion of the two bundles may not be very complete (Fig. 9) giving clear indication of the double nature of the petiolar strand. The same process is repeated below each pair of leaves (Figs. 11-14, 15-20, 21-23).

Vascularization of the Axillary Shoot and its Spines.—Two vascular strands, one on either side of the paired traces of a leaf form the vascular supply of its axillary shoot (Figs. 8, 13, 16-18, 21-22). These vascular strands radially divide even inside the parent stem to form the eustele of the axillary shoot. From this, two separate vascular strands that are related to a single gap are set off on either side for the spine of that side (Figs. 5, 9). Each spine is, therefore, vascularized, like the leaf, by two discrete vascular strands that are related to a single gap in the eustele of the axillary shoot. Further, the two traces of a spine fuse to form a single strand which quickly splits up to form three bundles in the base of the spine (Figs. 6-7, 10-11). Inside the spine, of the three vascular bundles, the median one is placed towards the abaxial and the lateral ones more towards the adaxial side (Figs. 7, 11). Further, throughout its longitudinal course, the median bundle of the spine gives out lateral branches which run outward and downward and connect with the lateral bundles. In the apex of the spine, all its three bundles once again become united. In their attachment to a unilacunar, double-trace node of the axillary shoot, the spines, anatomically are exactly similar to the leaves, modifications of which they are.

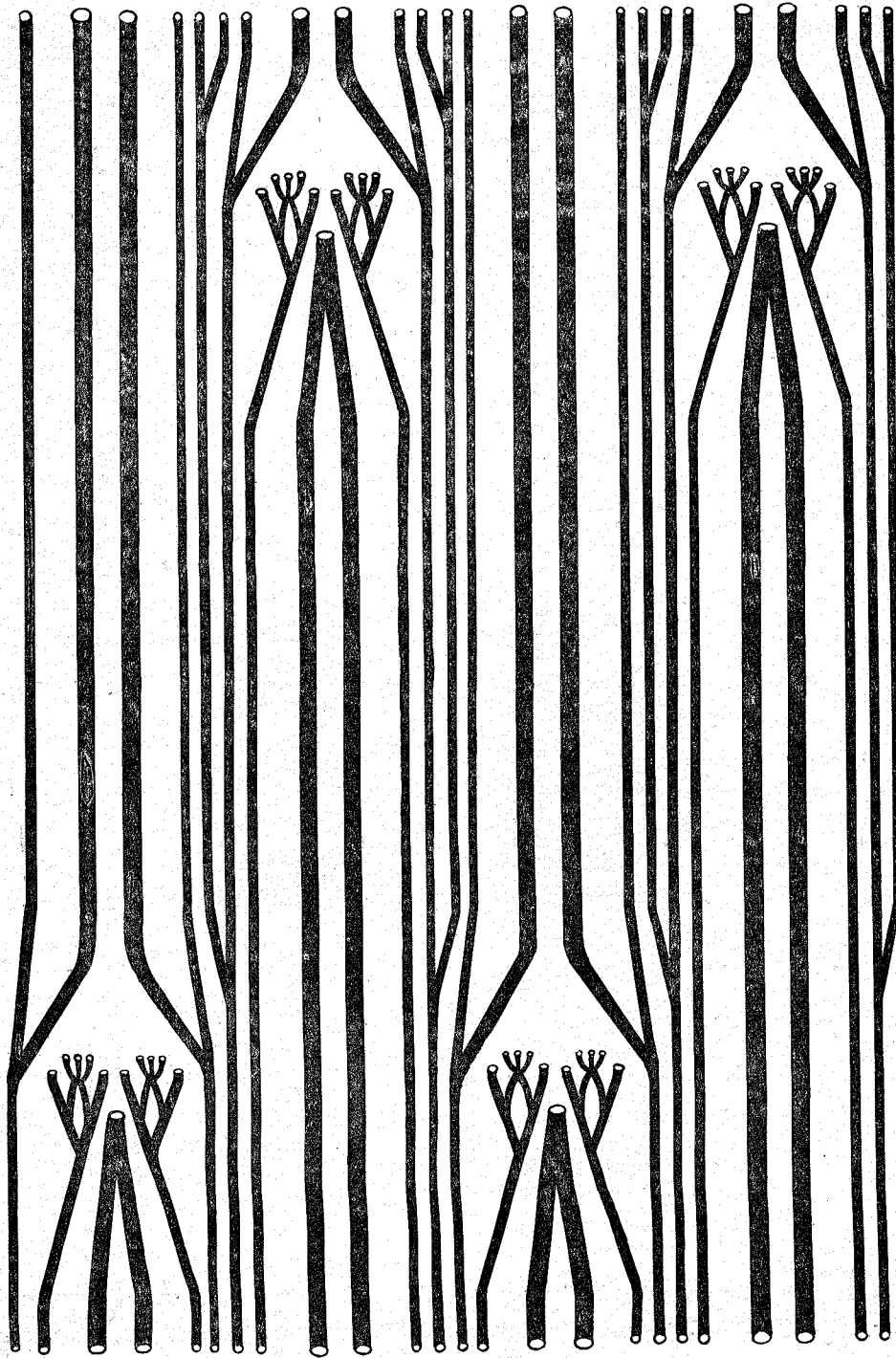
The longitudinal course of traces of the leaves, their axillary branches and of the spines through two successive nodes may be reconstructed from the study of serial transverse sections and is depicted diagrammatically by a stereodiagram which is cut longitudinally on one side and spread open (Fig. 24).

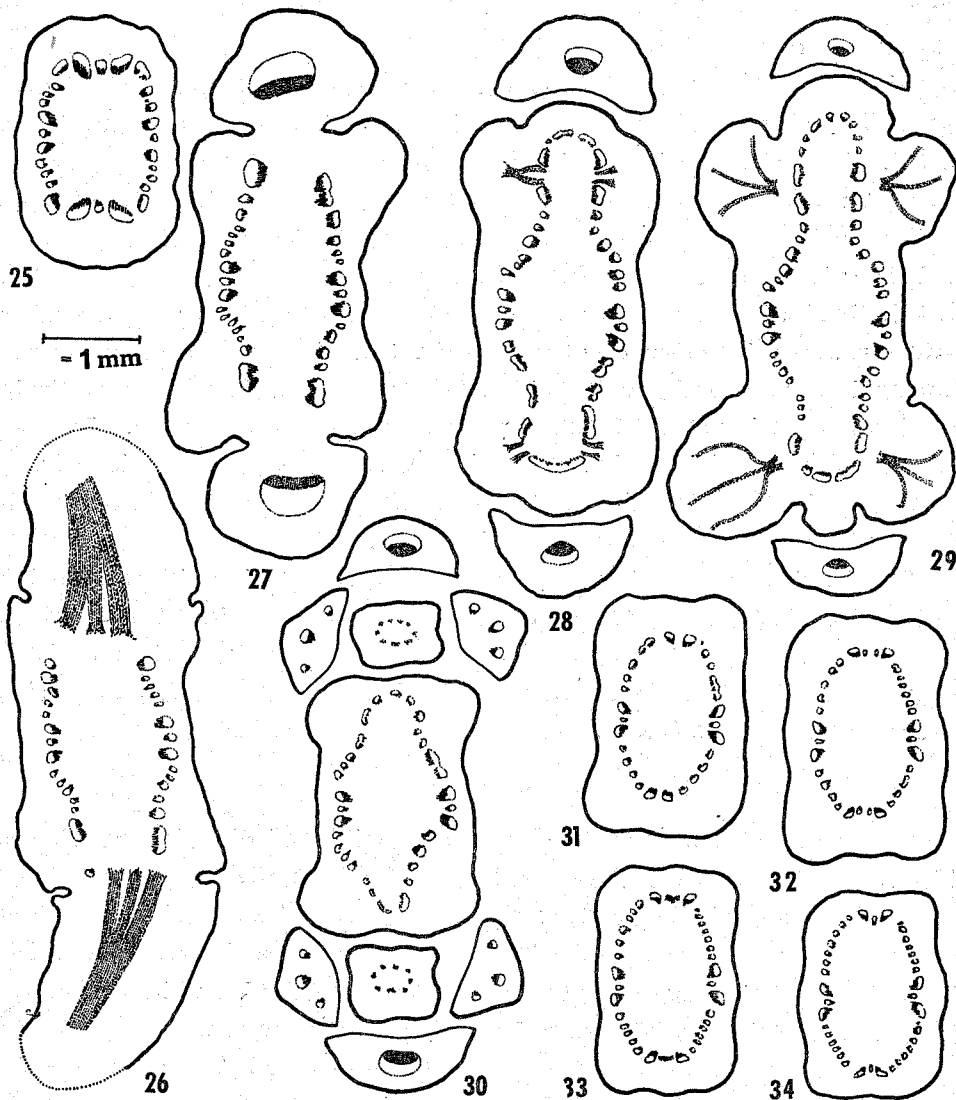
The above described type of node is found mostly in reproductive branches. Even here, in a number of cases and in the vegetative branches, a modification of this basic, unilacunar, double-trace type of node is found. In these also, the vascular supply of two successive pairs of leaves can be seen in the transverse section through the middle region of an internode. However, each of the four groups of vascular traces for the two successive pairs of leaves above consists



Figs. 3-23. Serial transverse sections of a vegetative twig, through four successive nodes, viz. Figs. 3-11, 12-14, 15-20, 21-23, showing the mode of origin of the vascular supply of the leaves, their axillary shoots and the spines: in this series, the leaf is vascularised by two traces (for explanation, see text).

of three vascular strands—two larger strands with a smaller strand inbetween (Fig. 25). The leaf, therefore, receives not the two even number but the three odd number of vascular strands. The mode of transition from an even number (two) to an odd number (three) of foliar vascular strands has been made out,





Figs. 25-34. Serial transverse sections of a vegetative twig comprising the top portion of an internode, a node and the basal portion of the internode above showing odd number of traces, viz. three for a leaf (for explanation, see text).

both by clearing techniques, as well as, by the study of serial transverse sections through two successive nodes and three internodes of the stem. In

Fig. 24. Stereo-diagram of the primary vascular skeleton of a vegetative twig comprising one complete internode with a node and portion of an internode both at its top and the base, cut longitudinally on one side and spread open, showing the mode of origin of the vascular supply of the leaves (two-traced), axillary branches and the spines (for explanation, see text).

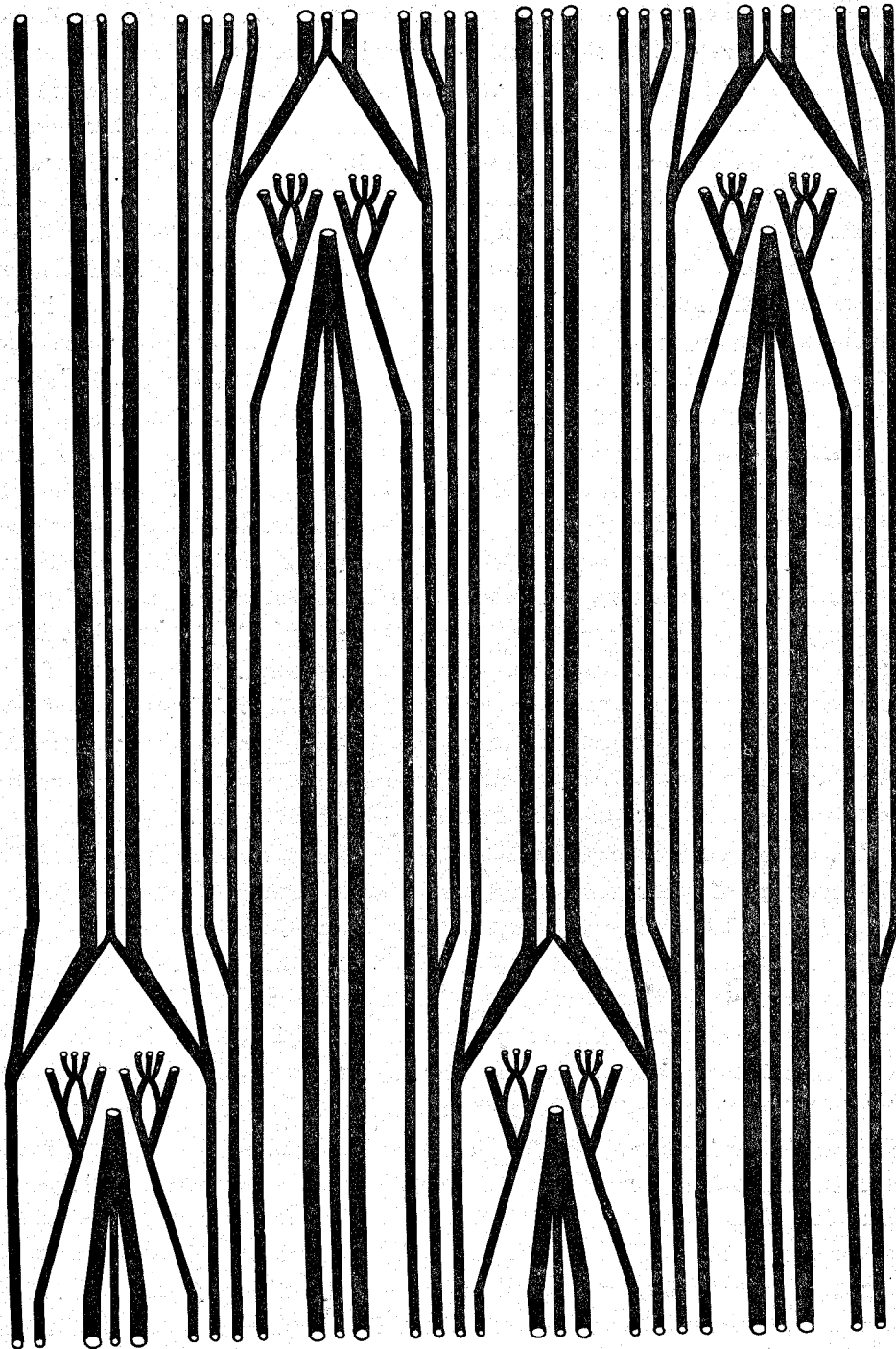
the subnodal region, just below the leaf base, the three vascular traces of a leaf diverge outward (Fig. 26) and become laterally united to form a single vascular strand of the petiole (Fig. 27). Here, the mode of vascularization of the axillary shoot and its two spines is similar to that described earlier (Figs. 27-30). After the vascular supply to the leaves and their axillary shoot has been set off, two vascular strands, one from either side of the gap (which earlier furnished the vascular supply to the subtending leaf, its axillary branches and the spines) are given off in the base of the internode just above (Figs. 30-31). Each of these vascular strands bifurcates into a larger and a smaller strand towards its other counterpart (Fig. 32). The two smaller strands quickly become laterally united (Fig. 33) to form a single, comparatively smaller, intervening strand between the two larger ones (Fig. 34). In some cases, the median intervening strand is produced, not by the fusion of two strands but by the bifurcation of only one of the two large strands, the branch shifting inward and assuming a central position.

The longitudinal coursing of the traces of leaves, their axillary branches and of the spines through two successive nodes and three internodes may be reconstructed from the study of serial transverse sections and is depicted diagrammatically by a stereo-diagram which is cut longitudinally on one side and spread open (Fig. 35).

Discussion

As a result of a wide reconnaissance of families of dicotyledons, Sinnott (1914), Sinnott and Bailey (1914, 1915) concluded that the trilacunar type of node is the most primitive among the angiosperms. The significance of the occurrence of a unilacunar, double-trace node among the angiosperms and which is so characteristic of gymnosperms was not realised for long. Its occurrence in the stem, cotyledons, stamens and carpels of certain taxa was considered to be either as abnormalities or as examples of fused organs. The odd-numbered types of node were thought to be characteristic of angiosperms and even-numbered of the gymnosperms. However, a recent revaluation of the whole problem in the light of the occurrence of a double-trace unilacunar node in certain taxa of the order Ranales (*sensu lato*) together with its widespread occurrence among the gymnosperms and the seedlings of angiosperms has now led Bailey (1956) to conclude that the unilacunar, double-trace node is in all probability the most primitive type among the angiosperms. Takhtajan (1964)

Fig. 35. Stereo-diagram of the primary vascular skeleton of a vegetative twig comprising one complete internode, with a node and portion of an internode both at its top and the base, cut longitudinally on one side and spread open, showing the mode of origin of the vascular supply of the leaves (three-traced), axillary branches and the spines (for explanation, see text).



holds a similar opinion. Probably, the most primitive condition of the foliar vasculature and of the node is present in *Austrobaileya*. Bailey and Swamy (1949) demonstrated that the decussate, pinnately veined leaves in this genus are vascularized by two separate vascular strands from a single gap in the eustele (the two strands are not produced by the bifurcation of a single strand but are related independently to a single gap in the eustele of the stem when traced downward in the internode). Further, the two strands frequently remain separate throughout the length of the petiole and of the midrib, each half of the leaf being vascularized by the ramifications of an independent system or at the most, the two strands fusing to form a single midrib in the middle and upper parts of the lamina. A similar condition of foliar vasculature has been reported in *Trimenia* of the Monimiaceae (Money, Bailey and Swamy, 1950), *Ascarina* of the Chloranthaceae (Swamy, 1953) and certain species of the Lactoridaceae, Verbenaceae, Labiatae and the Solanaceae (Marsden and Bailey, 1955). Paired, free or fused median traces are quite common in cotyledonary nodes of angiosperms (Bailey, 1956), carpels of divergent taxa—*Clethra*, *Pyrola*, *Epigaea* of the Ericaceae, *Anagallis* of the Primulaceae (see Eames, 1961, for a review), *Austrobaileya* of the Austrobaileyaceae (Bailey and Swamy, 1949) and certain species of the Cactaceae (Tiagi, 1958, 1960, 1963). Double-trace stamens although not of very common occurrence have been found in divergent taxa, both primitive and advanced—*Austrobaileya* (Bailey and Swamy, 1949), *Sarcandra* (Swamy and Bailey, 1950), *Victoria*, *Nuphar*, *Casuarina*, *Cyrtandra*, *Eranthemum*, *Doryanthes*, *Peristrophe* and certain species of the Betulaceae and the Fagaceae (see Eames, 1961, for a review).

There are several modifications of the nature of specialization of the primitive type of foliar vasculature and node as exhibited by *Austrobaileya*. In the family Calycanthaceae (Fahn and Bailey, 1957), the two traces of a leaf which are attached to a single gap in the eustele traverse upward through two successive internodes, a condition similar to that reported in *Azima* in this paper. However, in the subjacent region of the leaf inside the internode, the two traces become laterally united at varying depths from the leaf base, slightly below the node in *Chimonanthus praecox*, about half-way down in the internode below in *Calycanthus fertilis* and slightly more than one internode down in *Calycanthus floridus*. In *Azima tetracantha*, as reported in this paper, the fusion of the two or three leaf traces occurs almost at the level of the leaf base or in the leaf base itself. A similar condition has been reported by Tiagi (1958) in the stem of several species of *Opuntia*, *Nopalea*, and in the bracteoles present on the hypanthium of the inferior ovary of several species of *Pereskia*. In fact, in such cases, especially those where the fusion of the two leaf traces occurs at deeper levels inside the internodes, the node is likely

to be mistaken to be of the unilacunar, one-trace type, unless serial transverse sections are studied through several successive nodes and internodes. The tendency towards fusion of the basically two foliar vascular strands that are separate and independent at lower levels inside the stem but related to a single gap in the eustele is also known to occur in several species of the Monimiaceae (Money, Bailey and Swamy, 1950).

In certain other taxa, such as the family Cactaceae (Tiagi, 1958, 1960; Bailey, 1960), the two traces of a leaf which differentiate in the eustele undergo radial divisions to form two aggregations, each of several strands and at varying depths from the leaf base inside the internode. The number of vascular strands may be even or odd but there is no conspicuous larger strand occupying a median position in the arc of discrete bundles, the two aggregates of which are separated by a more or less conspicuous patch of parenchyma.

In still other taxa, certain species of the Monimiaceae (Money, Bailey and Swamy, 1950) and the Chloranthaceae (Swamy and Bailey, 1950; Swamy, 1953), each of the two leaf traces undergo radial divisions and an arc of four or more but even number of bundles is formed in which the two central bundles sooner or later become laterally fused to form an odd median strand. In the vegetative shoots and also frequently in the reproductive shoots of *Azima tetracantha*, each of the two traces of a leaf soon after their separation from a single gap in the eustele, bifurcates once to form an arc of four vascular strands, of which the two median ones quickly become united to form a single, comparatively smaller, median strand, the three vascular strands then traversing upward through two successive internodes before entering a leaf (Fig. 35).

In conclusion it may be said that although it is still too early to say anything about the phylogenetic relationships of the family Salvadoraceae, but the occurrence of a double-trace, unilacunar node and its modifications in *Azima tetracantha* is of significance in any general discussion about the possible relationships of this family to the other families of dicotyledons.

Summary

In the present investigation, morphology and nodal anatomy of *Azima tetracantha* Lamk. has been described. The two laterally disposed spines in the axil of a leaf in this species represent morphologically as well as anatomically the first pair of leaves of the axillary shoot.

The node is of the unilacunar, double-trace type. Each leaf is vascularised by two traces which descend down through two successive internodes before becoming attached independently to a single gap in the eustele. A common modification of this basic, double-trace foliar vasculature is produced by the bifurcation of the two foliar strands just after their separation from the gap

to form four strands, the two smaller middle members quickly fusing to form a single median, comparatively smaller odd member flanked on either side by a larger bundle. In any case, the two or the three foliar strands become fused at the level of the leaf base to form a single petiolar bundle.

The mode of vascularization of the spines is similar to that of leaves. Each spine is furnished with two separate strands that are related to a single gap in the eustele of the axillary shoot. These fuse to form a single strand in the base of the spine and this quickly forks into three strands which traverse up to the apex of the spine.

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