TAXONOMY OF THE TRIBE JUSTICIEAE (ACANTHACEAE) BASED ON WOOD MICROSCOPY

P. C. DATTA AND R. K. MAITI (*)

INTRODUCTION

Most of the betanists have derived Acanthaceae from the **Scrophulariaceae** or stocks ancestral to them (Lawrence, 195). The family has been divided into four sub-families by Engler and Diels (1936), of which the sub-family Acanthoideae has been divided into **Contortae** and **Imbricatae. Barlerieae** is a tribe under **Contortae.** The tribes of **Imbricatae** include **Andrographideae** and **Justicieae** along with others. The classification of Bentham and Hooker (1873) is markedly different. According to them the family is divided into five tribes. The tribe **Justicieae** is composed of six subtribes of which three are common in India; **Barlerieae, Andrographideae** and **Eujusticieae** (Hooker, 1885). So, according to Engler and Diels (1936), Barlerieae are far apart from **Andrographideae** and their **Justicieae,** while according to Bentham and Hooker (1873), they are very close to **Andrographideae** and their **Eujusticieae.**

Cytological investigation on some of the species of **Justicieae** has thrown some light in solving taxonomical controversies (Datta and Maiti, 1968). Study of wood anatomy has been regarded as one of the most definite ways of tracing interrelationships (Frost 1930, 1931; Kribs, 1935, 1937; Tippo, 1946; Money **et al,** 1950; Sinnott, 1952; Bailey, 1953; Cheadle, 1944, 1953, 1963; Bierhorst, 1960). For this reason, study of secondary xylem of the species under review has been taken up to see how far this investigation can be of use. Moreover, the consultation of literatures reveals that a very few species of the tribe **Justicieae** of Bentham and Hooker have been investigated by anatomists (Metcalfe and Chalk), 1950). The selection of this group of plants for such critical investigation also lies in the therapeutic importance of all the species belonging to this taxon (Chopra **et al,** 1956).

MATERIALS AND METHODS

Samples of wood belonging to different species of the tribe were collected from plants growing in Calcutta and its suburbs. The plant species included are:

(*) Departament of Botany, Calcuta University, Calcuta-19 - India.

Subtribe **Barlerieae : Barleria prionites** Linn. **Barleria cristata** Linn. var **dichotoma** Roxb.

Subtribe **Andrographideae : Andrographis paniculata** Nees.

Subtribe **Eujusticieae : Adhatoda vasica** Nees., **Ecbolium linneanum** Kurz., **Justicia gendarussa** Burm.

Woods from different parts of a plant were collected to compensate dimensional variations of wood elements if any and to minimise statistical error. Inner regions and outer barks were totally scraped off to exclude primary xylem and the phloem. A large number of samples were collected from each species to facilitate statistical sampling. The wood specimens were boiled in 5% glycerine for two hours for softening. Thin free hand sections and macerations (following Jeffrey's methods; Johansen, 1940) stained with safranin os safraninlight green were examined. Differents were measured from different replicated slides at random to facilitate statistical sampling. Fifty random measurements (both length and breadth) were taken for each element and analyse statistically. The standard error have been given in parentheses. Degree of inclination of end walls of vessel members has been calculated by measuring the larger angle formed by the terminal and lateral walls of drawings of side views, and deducting 90°. All measurements were calculated from camera lucida drawings.

OBSERVATIONS

Barlerieae prionitis Linn.

Diffuse porous, irregular growth zones distinguishable for thick walled fibres of late wood; compact, hard (Fig. 1); pores circular, rarely angular, irregulary scattered, moderately numerous; no radial compression of pores noticed, tendency to lateral compression in clustered pore; mostly solitary, some in radial chains of 2-4, (commonly 2-3), few in clusters, proportion of solitary pores : pore clusters :: 22 : 6 : 1 approximately; fibres predominent, thick-walled; rays closely spaced; axial parenchyma sometimes partly paratracheal, vasicentric, unilayered; apotracheal diffuse parenchyma and clusters of apotracheal parenchyma common, containing a dark content.

Rays juvenile type, predominently uniseriate consisting of elongated upright cells; multiseriate rays also composed of upright cells; proportion of uniseriate : multiseriate :: 9 : 1 approximately. Ray type approaches heterogeneous I of Kribs (1935).

Vessel members mostly with inclined end walls, generally one end inclined, other truncated, long tails common; mean inclination of end walls 25.56; perforation plate simple having rim, pits round to ellipsoidal, alternate; L = 345.90 (± 17.36) /u, D = 29.55 (± 1.90) /u.

Fibres - fibre tracheids, thick-walled, smooth; apex gradually pointed; L = 1030.30 (± 30.25) /u; D = 11.12 (± 1.14) /u.

Barlaria cristata Linn. var. **dichotoma** Roxb.

More or less similar to that of **Barlaria prionitis**; wood diffuse porous; growth zones seen; wood compact, soft; pores circular to angular, scanty, scattered, not uniformaly spaced, smaller in siza, mostly solitary, rarely in short radial pore chains; proportion of solitary pores : pore chains : pore clusters :: 8 : 3 : 1 approximately; rays fine, wary, very closely spaced unlike that of **B. prionitis**, fibres predominate, not much thick-walled (Fig. 2); axial parenchyma diffuse and in apotracheal clusters, contain a gummy substance.

Ray cells elongated, slightly juvenile, rectangular, simple pitted, ray type approaches heterogeneous I of Kribs (Fig. 3), having comparatively low uniseriate rays.

Vessel members with inclined to truncated ends; mean inclination 22.36°; tail present, perforation simple; intervessel pits smaller, roundish, minute, numerous closely and uniformly spaced, alternate; L = 975.02 (± 38.90) /u, D = 79.98 (± 1.72) /u.

Fibres - fibre tracheids, thin walled, much larger in comparison to those of B. prionitis, L = 703.80 (± 38.86) /u, D = 15.74 (± 1.22) /u.

Andrographideae

Andrographis paniculata Nees. Wood semi-ring porous, compact; pores ovoidal, scanty, smaller in size, in radial pore chains of 2-16, a few arranged in pore clusters of 3-6; solitary pores few; proportion of solitary pores : pore chaine - pore clusters :: 2 : 3 : 1 approximately; wavy, not closely and uniformly spaced (Fig. 4); axial parenchyma rarely paratracheal, diffuse apotracheal or grouped apotracheal common.

Rays uniseriate mainly, consisting of upright cells; multiseriate rays having both upright and procumbent cells; juvenile, heterogeneous type I of Kribs.

Vessel members' ends inclined to truncated; mean inclination 58.90°; perforation plate simple; vessel mostly narrow, intervessel pits numerous, closely spaced, medium sized, oval, 5-8 seriate, opposite to alternate; L = 402.05 (± 21.50) /u, D = 57.83 (±1.29)/u.

Fibre-tracheids thin-walled, gradually narrowed and tapering at both ends; septate fibres present; L = 655.32 (± 29.02) /u, D = 26.66 (± 1.29) /u.

Enjusticieae

Adhatoda vasica Nees.

Semi-ring porous (Fig. 5), compact; pores oval to angular, medium sized, showing tendency of radial compression, arranged in radial pore chains of 2-11, also sparsely in pore clusters; solitary

pores sparse; proportions of solitary pores : pore chains : pore clusters :: 1 : 7 : 1 approximately; axial parenchyma diffuse apotracheal; walls with simple pits, larger than the vessel pits, closely spaced.

Wood rays heterogeneous type I of Kribs, consisting mostly of upright cells and fewer procumbent cells; multiseriate rays 2-3 (mostly 3) seriate, sometimes a short multiseriate portion of about 30 cells is alternated by a long uniseriate portion of about 60 cells high; ray langth 6-7 cells to very long rays many cells high; uniseriate rays 3-25 cells high, homocellular consisting of upright long linear cells, few heterocellular consisting mostly of upright cells, cells containing crystals (Fig. 6).

Vessel members' ends mostly truncated having a small tail, in some having inclined end walls; some perfectly truncated without any tail; mean inclination 19.00°, perforation plate simple; intervascular pits numerous, roundish to somewhat elliptical, minute, closely spaced (Figs. 14 - 15); L = 261.44 (± 8.16)/u, D = 64.09 (± 2.86)/u.

Fibre tracheids moderately thick walled, lumen broad, apex gradually attenuated but in some abruptly attenuated; libriform fibre absent; L = 442.36 (± 15.86)/u, D = 17.58 (± 0.97)/u.

Ecbolium linneanum Kurz.

Wood ring porous compact, moderately soft (Fig. 7); pores circular to oval, small, numerous, acattered, not uniformly spaced, mostly solitary, few in radial pore chains of 2-6 cells, obliquely placed at some places, pore clusters rare; proportion of solitary pores : pore chains : pore clusters :: 15 : 6 : 1 approximately; majority of wood elements fibrous; axial parenchyma diffuse apotracheal, granular crystals in parenchyma cells present.

Wood rays heterogeneous type I of Kribs; multiseriate rays generally 2 seriate with uniserriate extensions, 8-18 cells in height; uniseriate rays 6-15 cells in height. The ratio of uniseriate : multiseriate rays :: 1 : 1 approximately; short multiseriate portion alternated by long uniseriate portions; cells upright and elongated mainly (Fig. 8).

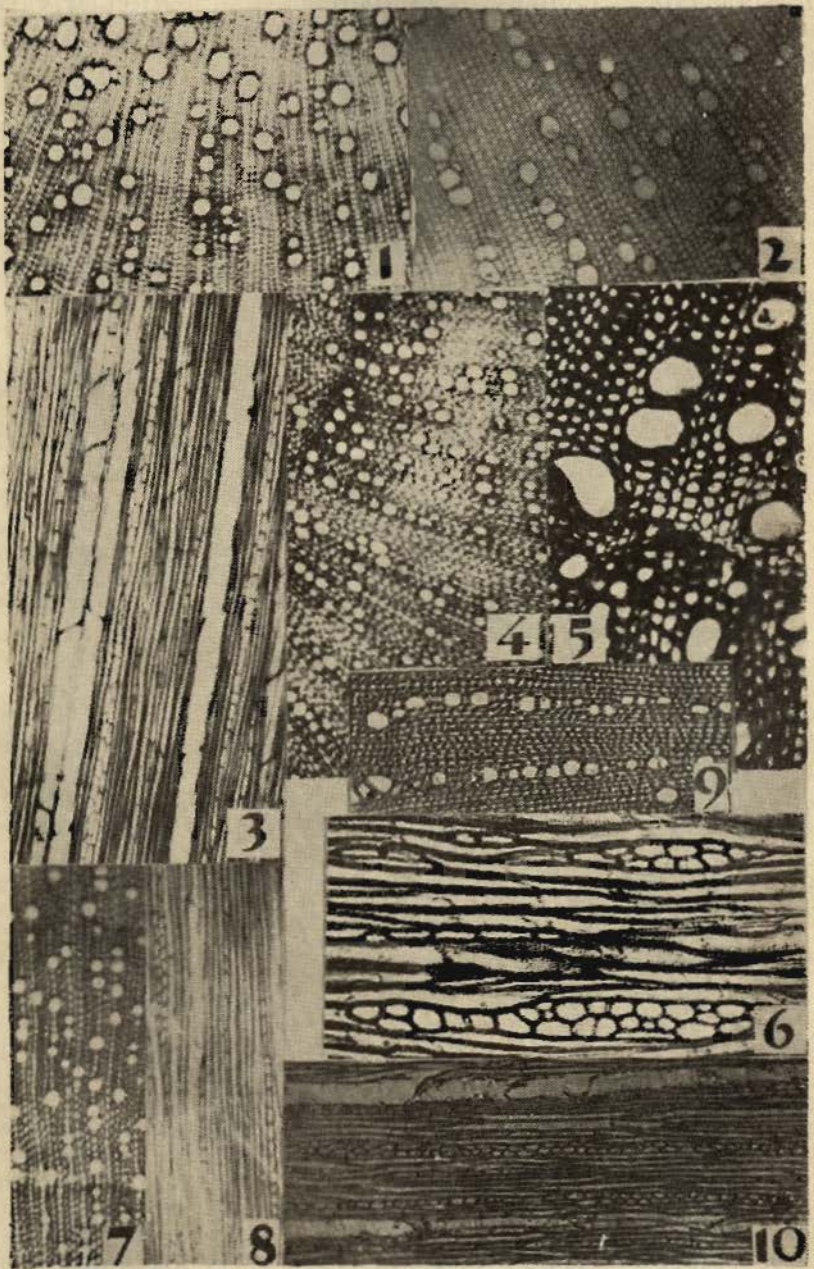
Vessel members provided with inclined ends at both ends and simple perforation plate, mean inclination of end walls 40.50°; pits minute, closely spaced, alternate; L = 349.00 (± 14.23) /u, D = 29.04 (± 0.70) /u.

Fibres - fibre tracheids, broad lumened, not uniformly tapering, lumen somewhat wavy, wall moderately thick. L = 474.20 (± 18.86) /u, D = 13.78 (± 0.40) u.

Justicia gendarussa Burm.

Wood semi-ring porous, not compact; pores arranged in distant regular radial series (Fig. 9), ovoidal to angular, scanty, small, arranged in long radial chains of 2-25 pores; uniseriate pores

few, relegated to lateral sides, 2-3 seriate; proportion of solitary pores : pore chains : pore aggregates :: 1 : 6 : 1 approximately; rays



closely spaced; axial parenchyma sometimes partly vasicentric, unilayered containing coloured contents, few apotracheal.

Ray type - I of Kribs, commonly multiseriate, 2-3 seriate; long multiseriate portion being intervened by short uniseriate cons-

trictions in tangential section; multiseriate rays containing larger number of procumbent cells; uniseriate rays of 3-25 cells' height: prismatic crystals common in all parenchyma, particularly ray-parenchyma; proportion of uniseriate : multiseriate rays :: 5 : 2 approximately (Fig. 10).

Vessel members' ends highly inclined to slightly inclined, mean inclination angle of end walls 28.14°; perforation plate simple; intervessel pits transitional from opposite to alternate, round, minute, 6-8 seriate, uniformly spaced close to each othrer (Fig. 11); L = 431.72 (± 19.35) /u, D = 123.84 (± 5.16) /u.

Fibres mainly fibre tracheids, thick walled, large lumened, pointed; L = 1014.37 (± 35.90)/u, D = 30.10 (± 1.09)/u.

DISCUSSION

Delimitation of tribes and subtribes:

Out of three subtribes of Bentham and Hooker (1873), under investigation, the subtribe **Eujusticieae** include **Justicia**, **Adhatoda** and **Ecbolium**. But in the Englerian system, **Ecbolium** has been placed in a separate taxon **Odontonemeae** (vide Torre and Harms, 1900-1907). From the anatomical investigation of secondary xylem, it is found that all the three genera possess some common characters e. g. heterogeneous type I ray, high frequency of multiseriate rays, alternation of multiseriate and uniseriate portions in high rays, thin-walled broad lumened fibre tracheids. Short multiseriate portions alternated by long uniseriate portions are common in **Ecbolium linneanum** and **Adhatoda vasica. Justicia gendarussa** possesses long multiseriate portions interrupted by short uniseriate constrictions in tangential section. These features support the placement of these species within the same taxon. **E. linneanum** is, however, distinct for its ring-porous wood, high frequency of solitary pores. Similarly, **J. gendarussa** can be isolated for very long radial pore chains, distantly spaced. **A. vasica** stands solitary in its indefinitely high rays.

From the study of the two species of **Barleria**, it is found that there are some distinct characters in this genus. Preponderance of uniseriate rays mostly of upright cells (a tendency towards the homogeneous type III of Kribs), clearly differentiates this genus from all other genera studied. This point has also been hinted at by Metcalfe and Chalk (1950), records of whom show that the genus **Barleria** is distinct from most of the other genera of Acanthacea in this particular aspect. In addition, absence of pore chains (or presence of very short ones), generally circular pores, thin walled parenchymatous fibres differentiate the two species of **Barleria** from other species of **Justicieae**. Therefore, separation of Barleria from other genera of **Justicieae** (Engler and Diels, 1936) can be supported on the basis of this anatomical character. The length and length/diameter ratio of vessel members, inclinations of end walls of vessel members, absence of libriform and septate fibres, high proportion of solitary pores in transverse section are also distinctive features.

Andrographis paniculata of the subtribe **Andrographideae** is also distinct for the presence of septate fibres, low proportion of solitary pores, semi ring porous wood and also by the ray type (heterogeneous type I of Kribs, 1935). The following classification will indicate these features :—

I. Pore chains scarce and very short; septate fibres absent; rays mostly uniseriate, mainly of upright cells; high proportion of solitary pores;

(i) Wood semi ring porous ... **Barleria prionitis**

(ii) Wood diffuse porous ... **B. cristata**

II. Pore chains common and long :

A. Rays mostly uniseriate, juvenile, consisting mainly of upright cells, septate fibres present; a pore chain having upto 16 pores .. **Andrographis paniculata.**

B. Multiseriate rays equal to or more than the uniseriate rays in frequency; septate fibres absent.

(i) Ray-height varies considerably, low to indefinitely high, short multiseriate portions alternated by long uniseriate portions; semi-ring porous or ring porous —

(a) Pores mostly solitary; distinctly ring porous wood ... ... **Ecbolium linneanum**

(b) Pores commonly in chains; wood semi-ring porous ... ... **Adhatoda vasica**

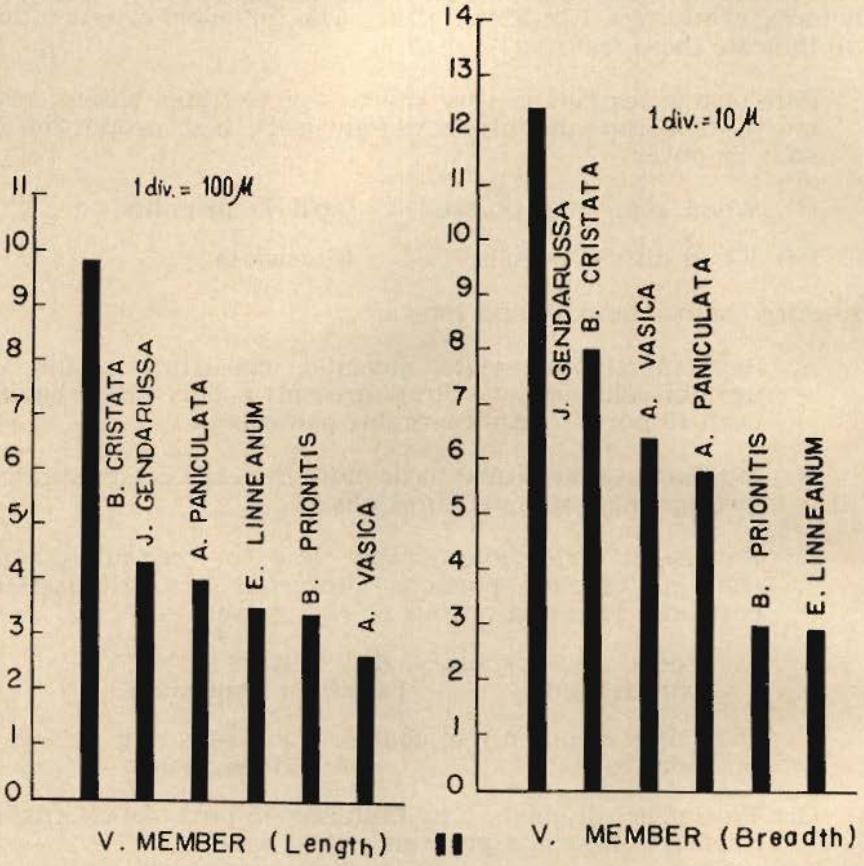
(ii) Rays generally high; long multiseriate portions alternated by short uniseriate portions; wood diffuse porous ... ... **Justicia gendarussa.**

A tentative plan of evolution:

The subtribe **Barlerieae** appears to be the most primitive from the nature of wood rays (heterogeneous type I, comparatively long vessel members (Fig. 11) with generally highest L/D ratio, highest proportion of solitary pores, generally diffuse of apotracheal parenchyma, comparatively narrow fibres and vessels, presence of angular pores and distinctly diffuse porous wood. But the inclination of end-walls suggest advance nature. It appears that **Barlerieae, Eujusticieae** and **Andrographideae** are three separate lines of evolution of which **Barlerieae** is widely apart.

Within the subtribe **Eujusticieae**, it becomes evident that **Ecbolium linneanum** is the most primitive species for its most inclined end walls of vessel members, very narrow vessels, very high proportion of solitary pores. But this species shows a combination of very advanced and very primitive characters. Advanced characters are circular pores and ring porous wood. **Adhatoda**

vasica is probably the most evolved species, for its shortest vessel members with low L/D ratio, the least inclined end walls of vessel



members, uniformly alternate intervascular pitting, shortest fibres, semi-ring porous wood and high frequency of pore aggregates. Presence of angular pores are primitive character. **Justicia gendarussa** shows a combination of some advanced and some comparatively primitive characters like **Ecbolium linneanum.** For example, short and broad vessel members and high proportion of pore aggregates are advanced characters. But the inclined end walls of vessel members, diffuse porous wood and comparatively long fibres occurrence of angular pores and heterogeneous type I ray are primitive characters. Any how, **Justicia** are also an advanced genus within this subtribe. **Andrographis paniculata,** an annual herb, also shows a peculiar combination of advanced and primitive characters. The length and breadth of vessel members represent the species as a moderately advanced one. Preponderance of uniseriate rays, semi ring porous wood and comparatively high frequency of pore aggregates support its advanced nature. But the highly inclined end walls of vessel members occurance of mostly opposite pits represent the genus as a comparatively primitive one. This combination of advanced and primitive characters may be a case of

paedomorphosis (Carlsquist, 1962). Whatever may be the explanation, these combinations of characters may be related to distinct lines of evolution, in which some particular characters may lag in the progressive changes.

Origin of **Barleria** from a separate stock, remote from the common stock of origin of **Andrographideae** and **Eujusticieae**, as was surmised from the cytological study (Datta and Maiti, 1968) is supported by the present findings also. Both **Andrographideae** and **Eujusticieae** are probably equally evolved.

SUMMARY

The present anatomical study corroborates the conclusions reached by cytological study. Separation of **Barleria** from the Justicieae by Engler and Diels has been justified.

Barleria seem to be the most primitive genus of all studied here. **Andrographideae** and **Eujusticieae** represent separate lines of evolution, probably arose from a common stock. **Adhatoda vasica** represents the most advanced characters of secondary xylem in the present study within the **Justicieae**.

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EXPLANATION OF FIGURES

Figs. 1 - 10. Sections through wood of Justicieae (Acanthaceae) 1, BARLERIA PRIONITIS L. - transection showing semi-ring porous nature. 2 & 3, B. CRISTATA L. var DICHOTOMA - transverse and tangential sections showing diffuse porous wood and heterogeneous rays. 4, ANDROGRAPHIS PANICULATA Nees. transection showing semi-ring. 5 & 6, ADHATODA VASICA Nees. - transverse and tangential sections showing semi ring porous nature and short uniseriate constriction of rays. 7 & 8, ECBOLIUM LINNEANUM Kurz. - transverse and tangential sections showing distinct ring and heterogeneous ray. 9 & 10, JUSTICIA GENDARUSSA Burm. - transverse and tangential sections showing long radial rows of pores and rays with long uniseriate portions between multiseriate ones.

Fig. 11: - Histogram showing lene 15 aid diam. of vessel member.

TABLE 1

Species	Wood pore	Ratio of Sol : Ch : Cl	Axial paren.	Ray type	Vessel members					Fibre	
					L (/u)	D (/u)	L/D ratio		Incl. Pits	L (/u)	D (/u)
Barlerieae											
B. prionitis	D Cir. ang. (rare)	22 : 6 : 1	uni. dif. pat.	hetero I (juv.)	345.90 (±17.36)	29.95 (±1.90)	11.54	25.56	ro. to ov., alt.	1030.3 (±30.25)	11.12 (± 1.14)
B. cristata	D Cir. & ang.	8 : 3 : 1	dif. pat.	hetero I (juv.)	975.02 (±38. 7)	79.98 (±1.72)	12.19	22.36	ro., alt.	703.80 (± 38.86)	15.74 (± 1.22)
Andrographideae											
A. paniculata	S. R. ov.	2 : 3 : 1	dif. & pat.	hetero I (juv.)	402.05 (±21.50)	57.83 (±1.29)	6.96	41.10	ov., opp. alt.	655.32 (± 29.02)	26.66 (± 1.29)
Eujusticieae											
Adhatoda vasica	S. R. ov. & ang.	1 : 7 : 1	dif.	hetero I	261.44 (±8.16)	64.09 (±2.86)	4.07	19.00	ro. to. ov., alt.	442.36 (± 15.86)	17.58 (± 0.97)
Eujusticieae											
Ecbolium linneanum	R. cir.	15 : 6 : 1	dif.	hetero I	349.00 (±14.23)	29.04 (±0.70)	12.00	40.50	ro., alt.	474.20 (± 18.86)	13.78 (±0.40)
Justicia gendarussa	D. ov. & ang.	1 : 6 : 1	uni., dif.,	hetero I	431.72 (±19.35)	123.84 (±5.16)	3.48	28.14	ro., opp. to alt.	1014.37 (± 35.90)	30.10 (± 1.09)

L = length, D = diameter, Incl. = inclination of vessel member ends; paren. = parenchyma;

sol. = solitary pores, Ch. = pore chains, Cl. = pore clusters; ro. = roun, ov. oval,

alt. = alternate, opp. = opposite. D = diffuse porous, S. R. = semi ring porous, R = ring porous,

cir. = circular to oval pores, ang. = angular pores, uni. = unilayered vasi centric,

dif. = diffuse apotracheal, pat. = patches of apotracheal parenchyma;

hetero = heterogeneous ray,

juv. = showing juvenile characters.

RESUMEN

El presente estudio anatómico corrobora las conclusiones obtenidas mediante las investigaciones citológicas. Se encuentra justificada la separación de **Barleria** de las **Justicieae**, propuesta por Engler y Diels.

Barleria parece ser el género más primitivo de todos los estudiados aquí. **Andrographideae** y **Eujusticieae** representan líneas de evolución separadas, aunque probablemente tienen un origen común.

En el presente estudio se encontró que dentro de las **Justicieae, Adhatoda vasica** presentó los caracteres más avanzados de xilema secundario.