

Characteristics of the Stem-Leaf Transitional Zone in Some Species of Caesalpinioideae (Leguminosae)

Abdel Samai Moustafa SHAHEEN

Botany Department, Aswan Faculty of Science, South Valley University - EGYPT

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Abstract: The vascular supply of the proximal, middle, and distal parts of the petiole were studied in 11 caesalpinoid species with the aim of documenting any changes in vascular anatomy that occurred within and between the petioles. The characters that proved to be taxonomically useful include vascular trace shape, pericyclic fibre forms, number of abaxial and adaxial vascular bundles, number and relative position of secondary vascular bundles, accessory vascular bundle status, the tendency of abaxial vascular bundles to divide, distribution of sclerenchyma, distribution of cluster crystals, and type of petiole trichomes. There is variation between studied species in the number of abaxial, adaxial, and secondary bundles, as seen in transection of the petiole. There are also differences between leaf trace structure of the proximal, middle, and distal regions of the petioles within each examined species. *Senna italica* Mill. and *Bauhinia variegata* L. show an abnormality in their leaf trace structure, having accessory bundles (concentric bundles) in the core of the trace. This study supports the moving of *Ceratonia* L. from the tribe Cassieae to the tribe Detarieae. Most of the characters give valuable taxonomic evidence reliable for delimiting the species investigated (especially between *Cassia* L. and *Senna* (Cav.) H.S.Irwin & Barneby) at the generic and specific levels, as well as their phylogenetic relationships.

Key Words: Abaxial, adaxial, accessory, secondary vascular bundles, indumentum, pericyclic fibre

Introduction

The Caesalpinioideae includes 171 genera and about 2250 species of tropical and sub-tropical trees and shrubs (Lewis et al., 2005). Polhill and Raven (1981) listed 5 tribes, namely Cercideae, Caesalpinieae, Cassieae, Amheristeae, and Detarieae, as the components of the Caesalpinioideae. As a result of phylogenetic analysis, Bruneau et al. (2001) and, more recently, Herendeen et al. (2003) listed only 4 tribes, Cassieae, Detarieae, Caesalpinieae, and Cercideae, as the components of the Caesalpinioideae.

Anatomical data have been used to good effect at all levels of the taxonomic hierarchy, as well as for identification and assessment of the taxonomic relationships among taxa of the flowering plants (Stuessy, 1990). The distribution of sclerenchyma and the nature of the mesophyll tissue proved highly informative regarding relationships at the generic level in Australian and South African Restionaceae (Briggs & Johnson, 1979); floral anatomy helped resolve the delimitation of the Bignoniaceae and Scrophulariaceae

(Armstrong, 1985). Pericarp structure has reinforced the distinctiveness of the surianoid taxa from the Simaroubaceae and has supported the inclusion of *Stylobasium* Desf. within the segregate family Surianaceae (Fernando and Quinn, 1992). Shaheen (2002) concluded that anatomical patterns of seeds of native and naturalised *Solanum* L. are useful for distinguishing 3 groups among the genus *Solanum*.

Petioles provide many useful anatomical characters. Agbaywa and Ndukwu (2004) reported variations in the number of bi-collateral vascular bundles in petioles of the species of *Cucurbita* L. Many useful petiole anatomical characters have been used in the delimitation and assessment of the identification of the genus *Ficus* L. (Saquaro, 2005), in *Musa* L. (Ennos et al., 2000), *Hevea brasiliensis* Wind. ex A.Juss (Maria & Rodriigo, 2003), 2 members of the family Leguminosae, *Bauhinia* L. and *Hardwickia* Roxb. (Seetharam & Kotresha, 1998), and 10 mimosoid species (Shaheen, 2006).

In the present study, the petiole anatomy of 11 caesalpinoid species were studied in cross sections

through the proximal, middle, and distal regions, with the aim of documenting differences in vascular anatomy within and between the petioles. This study also attempted to reveal additional characteristics for caesalpinoid species, which might be useful for identification and assessment of the taxonomic relationships among species studied at the generic and specific levels, as well as for assessing their phylogenetic relationships.

Materials and Methods

Mature, fresh, and dry material of 11 species belonging to *Cassia* L. (4 species), *Senna* (Cav.) H.S.Irwin & Barneby (4 species), *Delonix* L. (1 species), *Ceratonia* L.

(1 species), and *Bauhinia* L. (1 species) were collected (Table 1). The specimens represented horticultural species as well as some species native to Egypt (Boulos, 1999). *Senna occidentalis* (L.) Link is considered endemic to Egyptian Nubia (Shaheen, 2004). The identification of the local horticultural species was achieved by the morphological comparison to herbarium specimens kept at the ASW Herbarium (Aswan Faculty of Science Herbarium, South Valley University, Egypt). For fresh specimens, the most important collections are cited and are being kept in the ASW Herbarium. The voucher information for these specimens is listed in Table 1.

Three samples per species and 3 petioles per sample were examined. The petiole was reconstituted by boiling

Table 1. Collection data with the sources of the species studied.

No.	Tribe/Species	Source - collecting number-collector name
1	<i>Cassieae</i> , subtribe: (<i>Cassilinae</i>) <i>Cassia nodosa</i> Buch-Ham ex Roxb. (= <i>Cassia javanica</i> L. var. <i>indochiensis</i> Gagne)	I & III-11075-11076-A.M.Shaheen
2	<i>Cassia fistula</i> L.	IV-11077- A.M.Shaheen
3	<i>Cassia javanica</i> L.	I - 11078 - A.M.Shaheen
4	<i>Cassia spectabilis</i> L. (= <i>Cassia speciosa</i> Schhard.) (= <i>Cassia carnaval</i> Sperg.) (= <i>Senna spectabilis</i> (DC.) H.S.Irwin & Barneby) (= <i>Senna macranthera</i> (Collad.) H.S.Irwin & Barneby)	II - 11083 - A.M.Shaheen
5	<i>Senna siamea</i> (Lam) H.S.Irwin & Barneby (= <i>Cassia siama</i> Lam.)	I - 11079 - A.M.Shaheen
6	<i>Senna alexandrina</i> Mill, Barneby (= <i>Cassia acutifolia</i> Forssk)	I & II - 11080, 11081 - A.M.Shaheen
7	<i>Senna italica</i> Mill. (= <i>Cassia italica</i> (Mill.) Sprengel)	II - 521 - A.M.Shaheen
8	<i>Senna occidentalis</i> (L.) Link (= <i>Cassia occidentalis</i> L.)	II - 522 - A.M.Shaheen
9	<i>Detarieae</i> <i>Ceratonia siliqua</i> L.	II - 4244 - I. Springuel
10	<i>Caesalpinieae</i> <i>Delonix elata</i> (L.) Gamble (= <i>Poinciana elata</i> L., Gent.)	II - 6677 - L. Boulus
11	<i>Cercideae</i> <i>Bauhinia variegata</i> L.	III - 11082 - A.M.Shaheen

I: Plant Island, Aswan, Egypt.

II: Aswan Faculty of Science Herbarium (ASW).

III: Parks at Aswan Faculty of Science.

IV: Garden of Agricultural Museum, Dokki, Giza, Egypt.

in water with a few drops of Teepol for several minutes. Segments of 2-3 mm were then cut from proximal, middle, and distal regions of the petioles, placed in labelled tubes, and passed through an ethanol series (60%, 95%, and 100%) for 3-4 h. A microtome at Ain Shams University, Cairo, was used to cut sections. The sections were mounted in a drop of 5% aqueous acetone, attached by strongly heating slides over a Bunsen burner and were then stained with 0.05% toluidine blue (pH 4.4) for 2 min, rinsed thoroughly in tap water, dried on a hot plate, and mounted.

A Zeiss drawing apparatus on a Leitz microscope was used to draw representative sections. Photographs were taken under a Leitz Deplane photomicroscope in the Aswan Faculty of Science, Department of Botany, Egypt.

Results

Generally, the petiole nodes are trilacunar, each lateral leaf trace giving rise to a vascular bundle entering a stipule. In the pulvinus, the 3 leaf traces fuse to form a closed cylinder in some species, whereas in other species the leaf traces break up into 1 abaxial (central bundle), 1 adaxial (central bundle), and 2 secondary lateral (or proximal) bundles (Robbertse, 1973).

General description

Cassia nodosa (Figure 1a-c, Table 2)

The leaf trace is already divided into 7 bundles in the proximal region: 1 larger abaxial, 2 smaller adaxial, and 4 secondary vascular bundles. The petiole trace is more irregular in shape and pericyclic fibres are present in separate regions outside the phloem of each bundle. The cortical sclerenchyma is absent. Cluster crystals are confined to the cortical and pith cells. Multi-cellular trichomes are present.

In the medial region, the petiole trace is less irregular in shape, with fewer vascular bundles: 3 smaller abaxial, 3 larger adaxial (weakly divided) and 2 secondary vascular bundles (lateral to the adaxial bundles). Pericyclic fibres, cluster crystals, and multi-cellular trichomes are present in the proximal region.

In the distal region, the trace has a structure as in the medial region, except that all 4 secondary bundles lie above the adaxial bundles and the associated pericyclic fibres have joined into a continuous cylinder. Cluster crystals are confined only to the pith cells.

Cassia fistula (Figure 1d-f, Table 2)

In the proximal region, the petiole trace has a more irregular shape; it is already divided into 8 smaller abaxial, 1 larger adaxial, and 4 secondary vascular bundles (proximal). Pericyclic fibres are present in separate regions outside the phloem of each of these bundles. The cortical sclerenchyma is absent. Cluster crystals are confined to the pith cells only. Trichomes are fully absent.

In the medial region, the petiole trace is heart-like in shape, with additional, partly divided secondary vascular bundles (4 bundles) in the cortex in the adaxial position, and pericyclic fibres are present in separate regions outside the phloem of each of these bundles. The cortical sclerenchyma is present as lightly lignified cells adjacent to the epidermis. Cluster crystals are confined to the pith cells only. Trichomes are absent.

In the distal region the trace has a structure as in the medial region, except that all secondary vascular bundles (5) lie above the adaxial bundle. Neither the 2 major bundles nor their associated pericyclic fibres have joined into a continuous cylinder.

Cassia javanica (Figure 2a-c, Table 2)

The petiole trace in the proximal region is divided into 5 bundles: 1 abaxial, 2 adaxial, and 2 secondary vascular bundles. Pericyclic fibres are present in separate regions outside the phloem of each of these bundles. The cortical sclerenchyma is absent. Cluster crystals are confined only to the cortical cells. Dense multi-cellular trichomes are present.

In the medial region, the petiole trace is heart-like in shape. The abaxial and adaxial bundles have united into a more or less complete vascular cylinder with a continuous layer of pericyclic fibres, above which lies an arc of 2 or 3 secondary vascular bundles. The cortical sclerenchyma is still absent. Cluster crystals are distributed through cells of the cortex and pith. Trichomes are absent.

In the distal region, the petiole trace consists of 3 abaxial, 1 adaxial, and 2 secondary vascular bundles (lateral to the adaxial bundle). All of these bundles lie within a continuous layer of pericyclic fibres. There are another 2 secondary bundles on the adaxial position, each with their own cap of pericyclic fibres. The cortical sclerenchyma and trichomes are absent. Cluster crystals are confined only to the cortical cells.

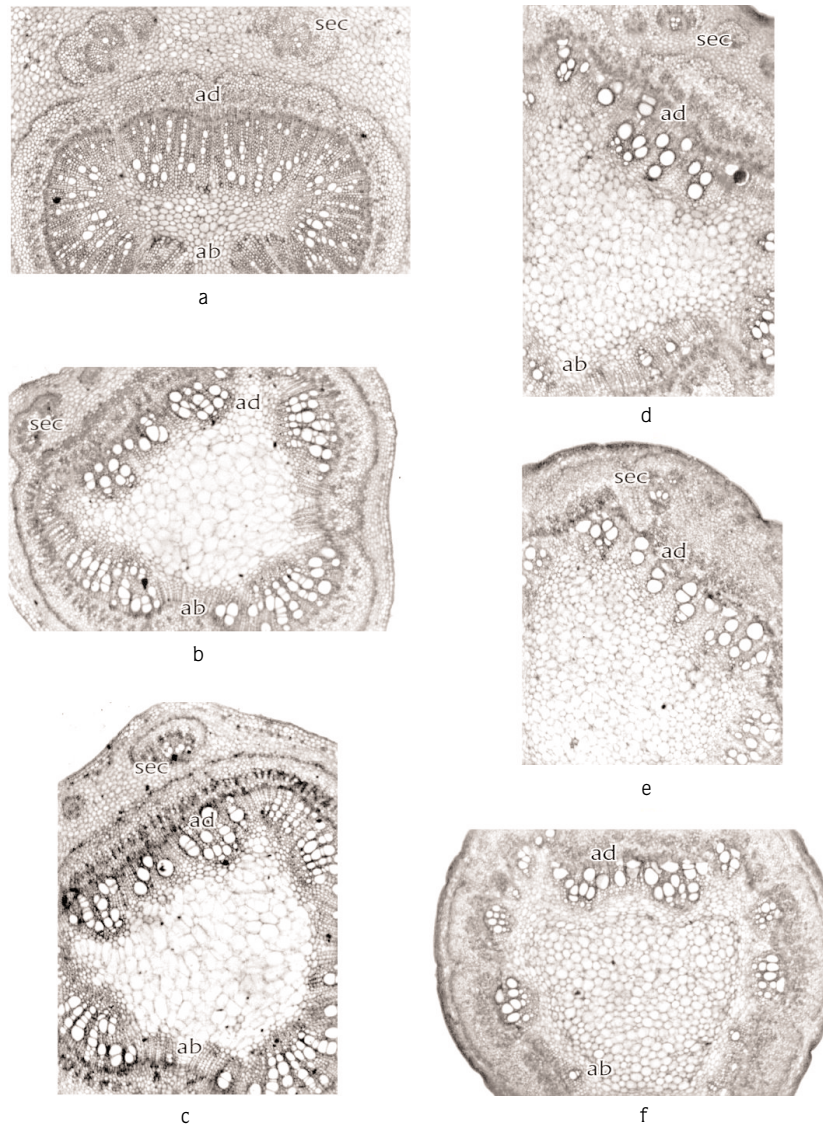


Figure 1. Transverse sections of the petiole of *Cassia nodosa* (a-c) and *C. fistula* (d-f). Abaxial bundles (ab); adaxial bundles (ad); secondary bundle (sec).

***Cassia spectabilis* (Figure 2d-f, Table 2)**

The leaf trace is ovate in the proximal region, but already divided into 3 abaxial fragments and 1 adaxial fragment. A pair of secondary vascular bundles is now apparent at the other side of the leaf trace. The pericyclic fibres are present in a narrow band that is separate above each bundle. The cortical sclerenchyma is absent. Cluster crystals are present in the cortical and pith cells. Dense multi-cellular trichomes are present.

In the medial region the division between the abaxial (5 bundles) and adaxial (1 large flattened) bundle is much

more pronounced, and the 2 secondary vascular bundles are still apparent at the other side of the leaf trace. Pericyclic fibres are present, as before, above each bundle. The cortical sclerenchyma is still absent. Cluster crystals are confined only to pith cells. Dense multi-cellular trichomes are present.

In the distal region, the abaxial and adaxial bundles have fused to form a single ovate petiole trace. One pair of secondary vascular bundles remains in the cortex within the pronounced ridges on the petiole. The pericyclic fibres form a continuous layer around the main

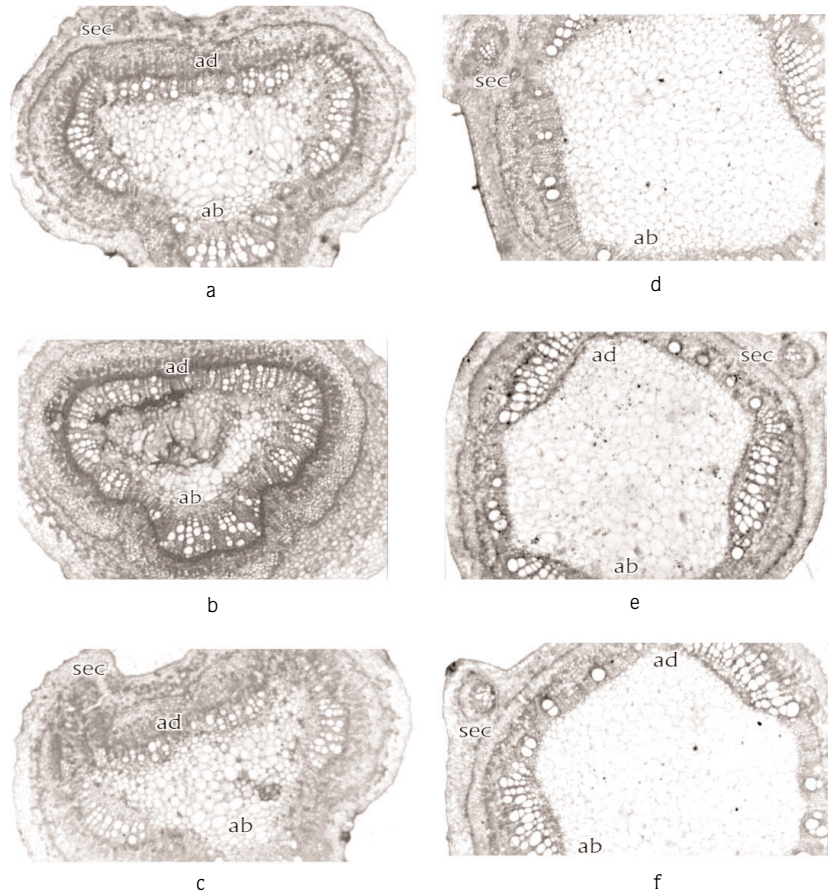


Figure 2. Transverse sections of the petiole of *Cassia javanica* (a-c) and *C. spectabilis* (d-f). Abaxial bundles (ab); adaxial bundles (ad); secondary bundle (sec).

trace and a band of fibres is also present outside the phloem of each secondary vascular bundle. The cortical sclerenchyma is still absent. Cluster crystals are confined only to the pith cells. Dense multi-cellular trichomes are present.

Senna siamea (Figure 3a-c, Table 2)

In the proximal region, the petiole trace consists of 3 abaxial, 2 adaxial, and a pair of secondary vascular bundles, which lie at the edge of the adaxial bundle. The pericyclic fibres are now broken into discrete caps on the 5 bundles. The xylem is divided into separate bundles, but the phloem is continuous between them in the abaxial bundles. The cortical sclerenchyma and trichomes are absent. Cluster crystals are confined to the pith cells.

In the medial region, the petiole trace consists of 3 abaxial (weakly divided) and 1 adaxial bundle, each with its own segment of pericyclic fibres. Here, the xylem has

a tendency to divide into 4 bundles, but the phloem is continuous between them. The cortical sclerenchyma and trichomes are absent. Cluster crystals are as in the proximal region.

In the distal region, the trace has a structure as in the medial one, except that there are extra abaxial bundles (5 bundles). The xylem is divided into separate bundles, but the phloem and the pericyclic fibres are in a continuous ring. A pair of secondary vascular bundles lie laterally to the adaxial bundle. The cortical sclerenchyma and trichomes are absent. Cluster crystals are confined to the pith cells.

Senna alexandrina (Figure 3d-f, Table 2)

In the proximal region, the leaf trace is already divided into 5 bundles: 1 larger abaxial, 3 much smaller adaxial, and 2 secondary vascular bundles

Table 2. Characteristics of the petiole anatomy of investigated caesalpinoid species. P = proximal region, m = medial region, d = distal region, and v.b = vascular bundles.

Species/ characters	No. Abaxial v.b			No. Adaxial v.b			No. Secondary v.b			Pericycle form			Cluster crystals			Trichome type		
	P	m	d	p	m	d	p	m	d	p	m	d	p	m	d	p	m	d
<i>Cassia</i>																		
<i>C. nodosa</i>	1	3	3	2	3	3	4	2	4	s	s	c	cp	cp	p	M	M	M
<i>C. fistula</i>	8	8	8	1	1	1	4	4	5	s	s	s	pi	pi	pi	-	-	-
<i>C. javanica</i>	1	1	3	2	2	1	2	2	4	s	s	c	co	cp	co	M	M	M
<i>C. spectabilis</i>	3	5	5	1	1	1	2	2	2	s	s	c	cp	pi	pi	M	M	M
<i>Senna</i>																		
<i>S. siamea</i>	3	3	5	1	1	1	2	2	2	s	s	c	pi	pi	pi	-	-	-
<i>S. alexandrina</i>	1	1	1	3	3	3	2	2	2	s	s	s	pi	pi	pi	u	u	u
<i>S. italica</i>	3	3	4	1	1	1	2	2	3	c	c	c	cp	pi	cp	u	u	u
<i>S. occidentalis</i>	5	8	8	1	1	1	2	2	2	c	c	c	cp	cp	cp	-	-	-
<i>Ceratonia</i>																		
<i>C. siliqua</i>	1	1	1	1	1	1	-	2	3	s	s	s	co	co	co	-	-	g
<i>Delonix</i>																		
<i>D. elata</i>	1	1	1	1	1	1	-	2	5	s	s	s	-	-	-	-	-	-
<i>Bauhinia</i>																		
<i>B. variegata</i>	1	1	2	1	1	-	2	2	1	co	s	s	co	co	co	u	u	u

Key to attributes in Table 2

Pericyclic form: s = form a separate band above each bundle, c = form a continuous layer surrounded the main trace.

Cluster crystals: cp = confined to cortical and pith cells, pi = confined to pith cells only, co = confined to cortical cells only, - = absent

Trichome type: M = multi-cellular, u = unicellular, g = glandular, - = absent.

(above the adaxial ones). Pericyclic fibres are present in separate regions outside the phloem of each of these bundles. The cortical sclerenchyma is absent. Cluster crystals are confined to the pith cells. Unicellular trichomes are present.

In the medial and distal regions, the leaf trace has a structure as in the proximal region, except that the 2 secondary bundles lie within the ridge of the main trace and the cortical sclerenchyma is present as lightly lignified cells adjacent to the outer cortex.

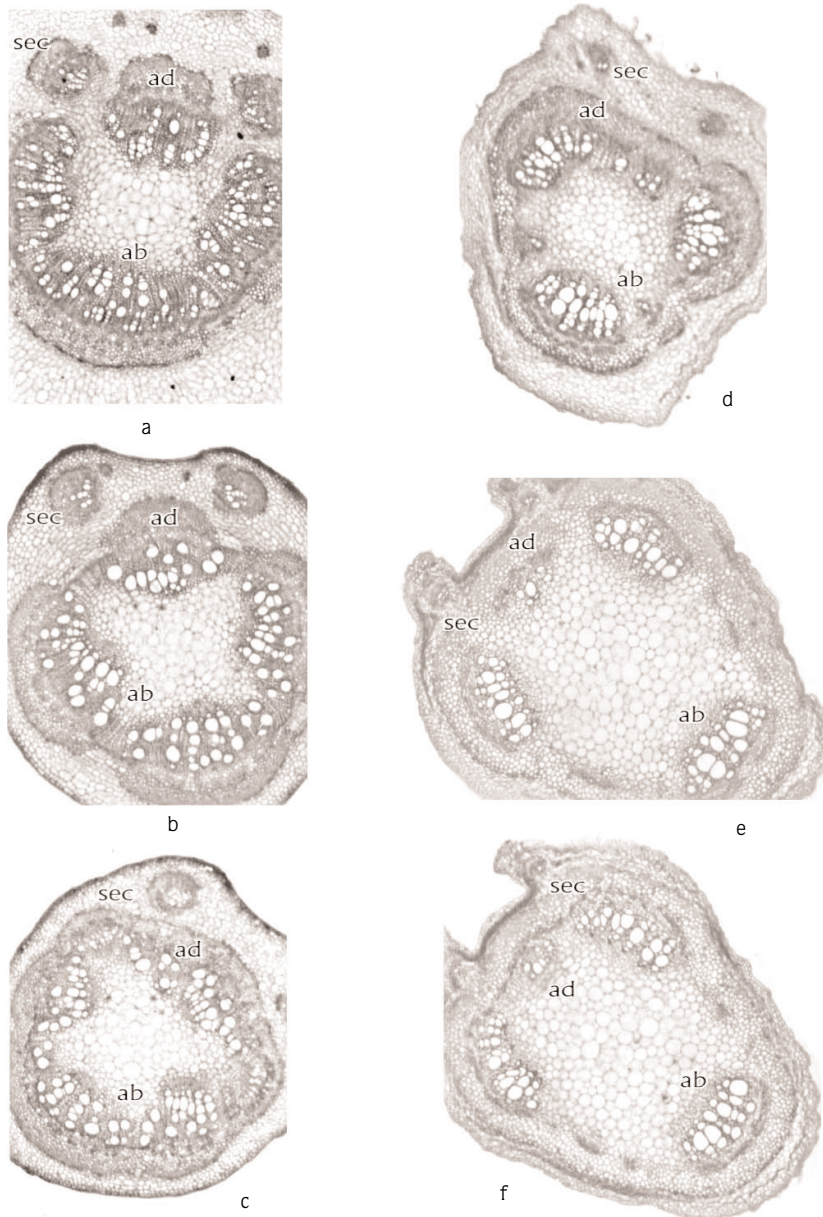
Senna italica (Figure 4 a-c, Table 2)

In the proximal region, the leaf trace is markedly divided, consisting of 3 abaxial bundles, 1 adaxial bundle, and a pair of secondary bundles, which lie laterally to the adaxial one. Pericyclic fibres are present as a continuous layer outside the abaxial bundles (3 bundles), as a separate layer outside the phloem of the rest of the bundles. The cortical sclerenchyma is absent. Cluster

crystals are present in the cortical and piths cells. Unicellular trichomes are present.

In the medial region, the petiole trace consists of a cylinder of some 4-5 bundles within a continuous layer of pericyclic fibres, and 2 or 3 secondary bundles lie in the outer cortex on the adaxial side, each with its own fibre cap. The cortical sclerenchyma is absent. Cluster crystals are confined to the pith cells. Unicellular trichomes are present.

The leaf trace in the distal region is highly distinctive. Here, there is a U-shaped group of abaxial bundles (4 bundles), 1 adaxial bundle, and 4 secondary bundles (laterally). In addition, a solid core of vascular tissue lies above the adaxial bundle, to which we apply the term accessory bundle. This bundle is distinguishable from the usual adaxial bundle in that it is concentric, with the phloem completely surrounding a fluted xylem mass, and the whole bundle is enclosed within a sheath of



Figures 3. Transverse sections of the petiole of *Senna siamea* (a-c) and *S. alexandriana* (d-f). Abaxial bundles (ab); adaxial bundles (ad); secondary bundle (sec).

sclerenchyma. Cluster crystals are present in the cortical and pith cells. Unicellular trichomes are present, while the cortical sclerenchyma is absent.

Senna occidentalis (Figure 4 d-f, Table 2)

In the proximal region, the leaf trace is cylindrical, being strongly divided into 5 large abaxial bundles, 1 very small adaxial bundle, and a pair of quite large secondary vascular bundles that lie laterally to the adaxial bundle.

Each bundle has a separate fibre cap. The cortical sclerenchyma is absent. Cluster crystals are present in the cortical and pith cells. Trichomes are absent.

In the medial region, the structure is complicated by the presence of extra abaxial bundles. The structure present in the proximal region is maintained, except that there are 8 abaxial bundles associated with a continuous band of pericyclic fibres, as well as a very small adaxial

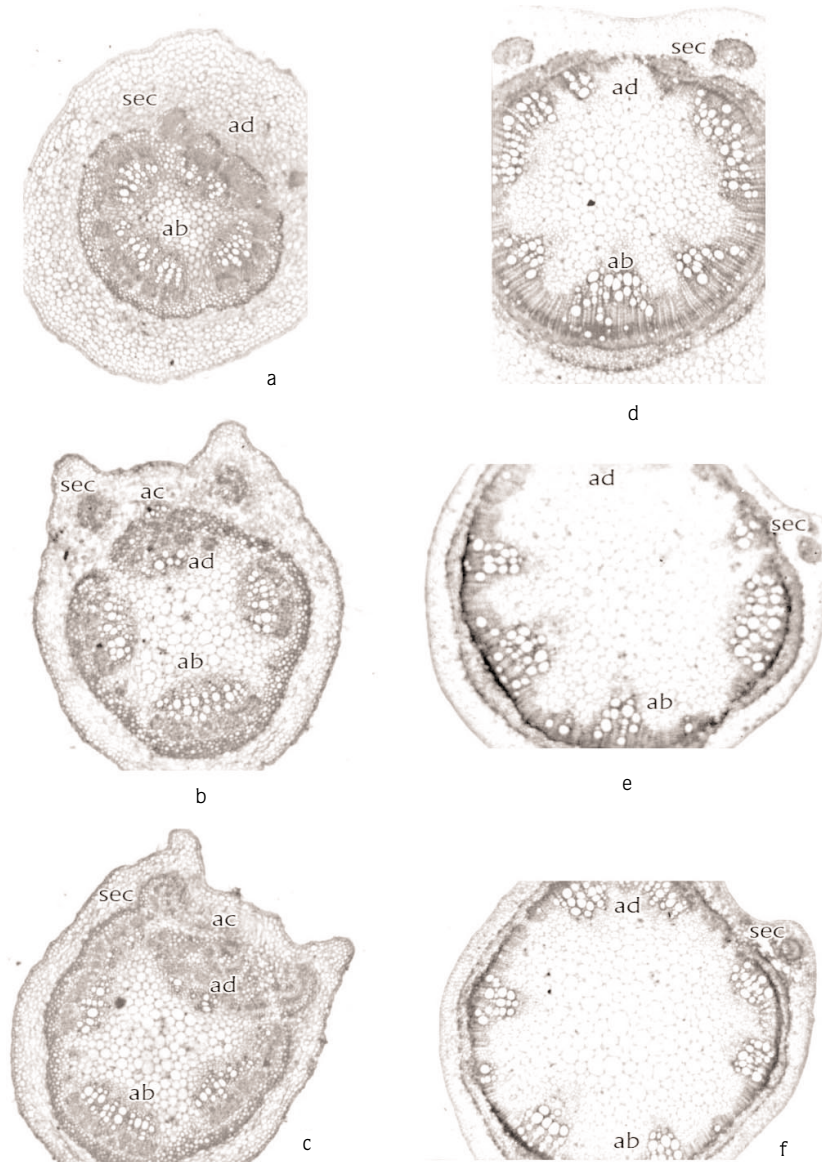


Figure 4. Transverse sections of petiole of *Senna italica* (a-c) and *S. occidentalis* (d-f). Abaxial bundles (ab); adaxial bundles (ad); secondary bundle (sec).

bundle. Two clear secondary vascular bundles lie laterally to the adaxial one at the edge of the main trace.

In the distal region, the petiole trace is still cylindrical, being divided into 8 much larger abaxial bundles and 1 much smaller adaxial bundle. The main trace is associated with a continuous band of pericyclic fibres. There are 2 clear secondary vascular bundles that lie laterally to the adaxial bundle. The cortical sclerenchyma and trichomes are absent. Cluster crystals are present in the cortical and pith cells.

Ceratonia siliqua (Figure 5a-c, Table 2)

In the proximal region, the leaf trace is irregular, being strongly divided into a more flattened abaxial and a more largely flattened adaxial bundle. There are no secondary vascular bundles. The cortical sclerenchyma is present as heavily lignified cells adjacent to the epidermis and distributed through the bundles and pith cells. Cluster crystals are confined to the cortical cells. Trichomes are absent.

In the medial region, the petiole trace consists of 1 very flat cup-shaped abaxial bundle, 1 adaxial bundle, and 2 secondary bundles. Each bundle has its own cap of pericyclic fibres. Cortical fibres are present as before. Cluster crystals are present in cortical and pith cells. Trichomes are absent.

In the distal region, the trace has a structure as in the medial region, except that all 3 secondary vascular bundles lie laterally to the adaxial one. Trichomes here are glandular.

Delonix elata (Figure 5d-f, Table 2)

In the proximal region, the leaf trace is irregular in shape and the structure is very simple, being divided into only 2 bundles: 1 large flattened abaxial (cup-shaped) and 1 smaller adaxial bundle. The division between the abaxial and adaxial bundles becomes clearer and each bundle has its own narrow band of pericyclic fibres. There are no secondary vascular bundles. The cortical sclerenchyma, cluster crystals, and trichomes are absent.

In the medial region, the trace is less irregular in shape, with an additional partly divided secondary vascular bundle occurring at the edge of the abaxial bundles. The abaxial and secondary vascular bundles are associated with a continuous layer of pericyclic fibres. The cortical sclerenchyma, crystals, and trichomes are still absent.

In the distal region, the trace becomes more complicated, having extra secondary vascular bundles (5 bundles), which lie above and at the edge of the adaxial ones. The cortical sclerenchyma, crystal, and trichomes are still absent.

Bauhinia variegata (Figure 6a-c, Table 2)

In the proximal region, the petiole trace is box-like in shape, consisting of 3 bundles within a continuous layer of pericyclic fibres and phloem. A pair of secondary bundles is present laterally in the outer cortex. In addition, one accessory vascular bundle (concentric) is present at the core of the trace. Cluster crystals are confined to the cortical cells. Both the cortical sclerenchyma and multi-cellular trichomes are present.

In the medial region, the main trace is divided into one abaxial and one adaxial bundle of about equal size. The accessory bundle is still present at the core of the bundle. The cortical sclerenchyma is still present. Cluster crystals are confined to the cortical cells.

In the distal region, the trace has an abnormal structure, consisting of 2 abaxial, no adaxial, and a single secondary bundle (which lies at the edge of the abaxial bundle). In addition, a pair of accessory bundles is present at the core of the trace. The cortical sclerenchyma is still present. Cluster crystals are confined to the cortical cells. Multi-cellular trichomes are present.

Discussion

Petiole anatomy characteristics

Variation in the species studied occurs in the number of abaxial and adaxial bundles, and the number and relative position of the secondary vascular bundles, as seen in the transections of the petioles. Differences between the proximal, middle, and distal regions of the petioles, within and between the studied species, also are noted (Table 2).

Number of secondary vascular bundles

The number of secondary vascular bundles in the proximal, medial, and distal regions of the petiole is informative; 2 are consistently present in all species of *Senna*, 2-5 in all species of *Cassia*, 0-3 in *Ceratonia siliqua*, 0-5 in *Delonix elata*, and 1-2 in *Bauhinia variegata*. This character seems to be diagnostic for *Cassia* and *Senna* at the generic level. This variation in the number of secondary bundles among studied taxa may provide useful information for identification of the bulk of the species examined. In addition, the resemblance in the number of secondary bundles in the proximal region of *C. nodosa* and *C. fistula* may account for the close relationships between them, which was previously elucidated by Bentham (1871) and Hussein et al. (2002a, 2002b) on the basis of morphological attributes. In addition, the resemblance in the number of secondary bundles in the medial region of all investigated species of *Senna* and *Cassia* may account for the close relationship between them as a monophyletic group belonging to the tribe Casseae and sub-tribe Cassilinae. In addition, the unique number of secondary bundles of the medial region (2 bundles) of the petioles of all investigated taxa (except for *Cassia fistula*) accounts for the close relationship between all the species. These results agree with those published by Shaheen (2006), who reported variations in the number of secondary vascular bundles in the petioles of some mimosoid species. In this respect, Seetharam and Kotresha (1998) reported that there are differences

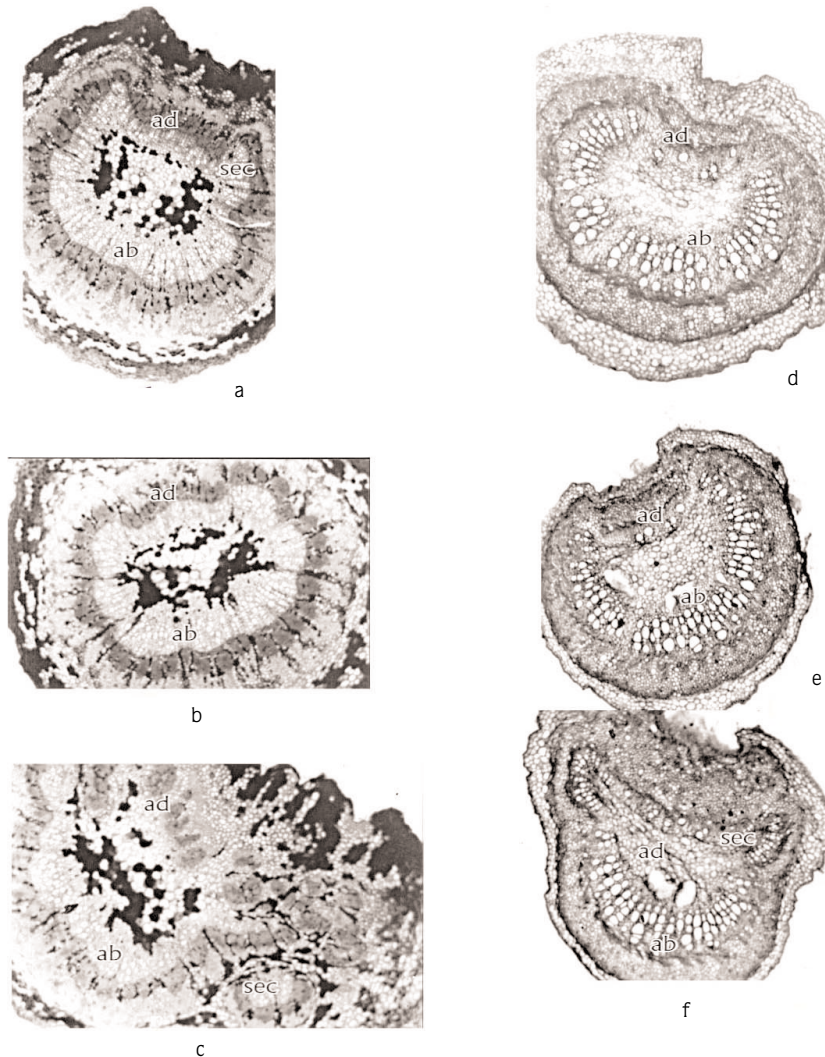


Figure 5. Transverse sections of petiole of *Ceratonia siliqua* (a-c) and *Delonix elata* (d-f). Abaxial bundles (ab); adaxial bundles (ad); secondary bundle (sec).

between *Bauhinia* and *Hardwickia* species in the number of secondary vascular bundles and their relative position.

Position of secondary vascular bundles in the medial region

There are 3 positions possible for secondary bundles in the medial region. These can be above the adaxial bundle in *Senna alexandrina*, laterally in 7 species (e.g., *Bauhinia variegata*), and both positions in *Cassia fistula*. The variation in the relative position of the secondary bundles may be useful for distinguishing some of the studied *Cassia* and *Senna* species. Shaheen (1995) reported only 2 positions (above adaxial and laterally) for

the secondary vascular bundles in the petioles of African and Australian *Acacia* species.

Number of abaxial vascular bundles

The number of abaxial bundles in the proximal, medial, and distal regions of the petiole varies from 1 to 8 in the investigated species of *Cassia* and *Senna* to only 1 in *Ceratonia*, *Delonix*, and *Bauhinia*. This character may be of limited usefulness for delimiting at the generic level, but it is very useful for identification at the species level; it is unique to *Senna siamea*, *Senna occidentalis*, *Senna italica*, *Cassia fistula*, *Cassia nodosa*, *Cassia spectabilis*, and *Bauhinia variegata*. In addition, the resemblance in

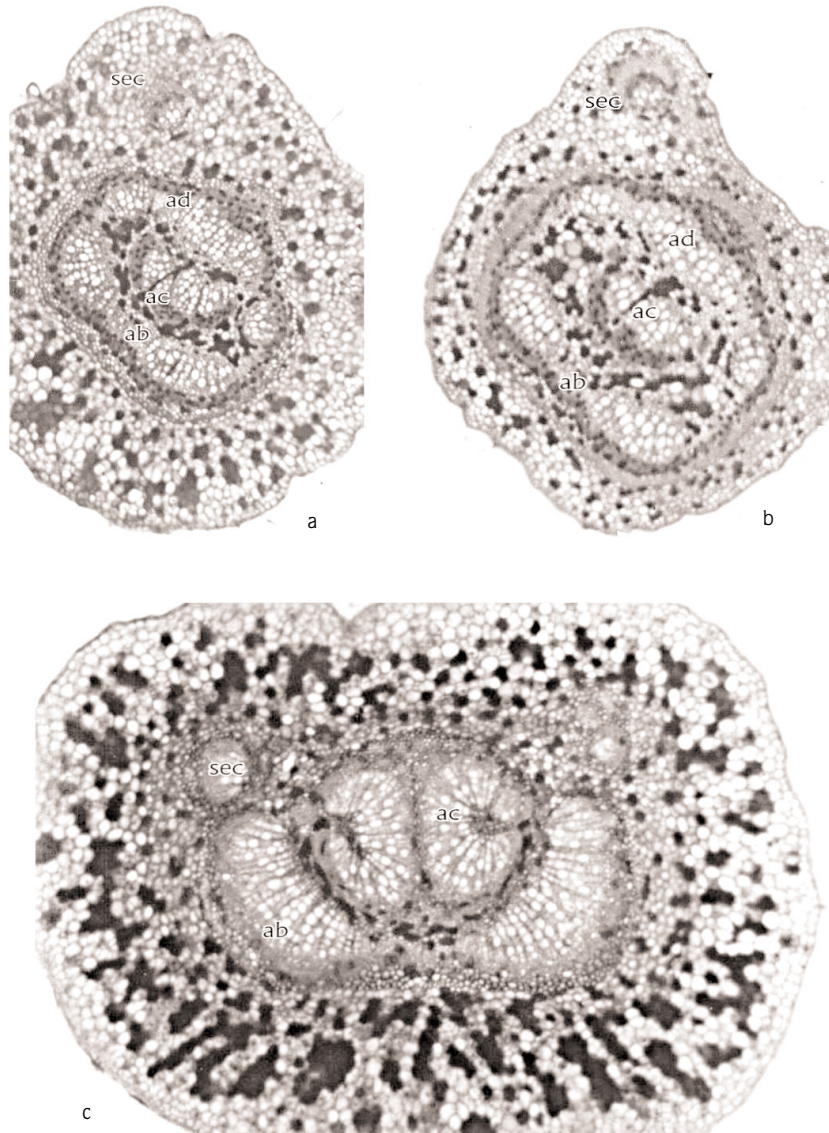


Figure 6. Transverse sections of petiole of *Bauhinia variegata* (a-c). Abaxial bundles (ab); adaxial bundle (ad); secondary bundles (sec); accessory bundles (ac).

the number of abaxial bundles in the proximal and medial regions of the petioles of *Senna alexandrina* and *Cassia javanica* may account for the close relationship between them, which was elucidated by Hussein et al. (2002b). These results agree with those published by Shaheen (1995, 2006), who reported that the number of abaxial bundles is informative and useful for the identification of Egyptian and some Australian *Acacias*, as well as some mimosoid species.

Number of adaxial vascular bundles

The number of adaxial bundles in the proximal, medial, and distal regions of the petioles varies from 1 to 3 in taxa of *Cassia* and *Senna* to only 1 (or absent) in *Ceratonia*, *Delonix*, and *Bauhinia*. This character seems to be useful for identification at the species level. It is characteristic for *Cassia nodosa*, *Cassia javanica*, and *Senna alexandrina*. It also seems to be useful at the generic level. In addition, the resemblance in the number

of adaxial bundles throughout the petiole of *Cassia fistula*, *Cassia spectabilis*, *Senna siamea*, *Senna italica*, and *Senna occidentalis* account for the close relationships between them as a monophyletic group belonging to the tribe Cassieae. Furthermore, the results support the moving of *Ceratonia* from the tribe Cassieae to Detarieae (Bruneau et al., 2001). In this respect, Shaheen (2006) proved the efficacy of this character in the delimitation and identification of some mimosoid species.

Abaxial bundle division

There is a tendency for the xylem and phloem to divide in certain regions of the petiole. In 6 species (mainly *Cassia* and *Senna*) the xylem and phloem are undivided, whereas they are divided in *Ceratonia*, *Delonix*, and *Bauhinia variegata*. These results support the moving of *Ceratonia* from the tribe Cassieae to Detarieae (Bruneau et al., 2001). Shaheen (2006) reported that in the petiole of some mimosoid species, the xylem is divided into separate bundles, but the phloem and pericyclic fibres are in a continuous ring. This character seems to be useful only for the identification of a few of the investigated species.

Documenting changes within the petiole trace structure of the same species

Three cases are recorded: no change in the structure of the leaf trace throughout the petiole (from proximal to distal) in *Senna alexandrina*; minor change in *Ceratonia siliqua*, *Delonix elata*, *Bauhinia variegata*, *Cassia fistula*, *C. specabilis*, and *Senna italica*; major change only in the petioles of *Cassia nodosa*, *C. javanica*, *Senna siamea*, and *Senna occidentalis*. This character seems to be of limited usefulness at the generic level and somewhat useful for the identification of some examined species. This result agrees with Shaheen (2006), who reported the efficacy of this character in the identification of some mimosoid species.

Pericyclic fibres form in the proximal regions

There are 2 cases: they form a continuous ring in *Senna italica* and *Senna occidentalis*, and present in a separate band above each bundle in the proximal and medial regions of the petioles in the remaining species. Hence, this character seems to be distinctive for the identification of *Senna occidentalis* and *S. italica*. The resemblance of this character between *Senna alexandrina* and *Senna italica* may account for the close relationship between them, which was previously elucidated by

Hussein et al. (2002b) on the basis of morphological attributes (morphological characters of the seeds).

Accessory vascular bundle status

The accessory bundle is a solid core of vascular tissue in the centre of the petiole, to which Shaheen (1995) applied the term accessory vascular bundle. There are 3 cases: it is present as a single bundle above the adaxial bundle in *Cassia italica* (distal region), as 1-2 bundles in the core of the trace in all regions of *Bauhinia variegata*, and is absent in the rest of the species. In this respect, Shaheen (1995) reported that this type of bundle is distinguishable from the usual adaxial bundle in that it is concentric, with the phloem completely surrounding a fluted xylem mass, and a whole bundle is enclosed within a sheath of the sclerenchyma. The presence of accessory bundles seems to be useful only at the species level; hence, it seems to be distinctive for recognition and identification of *Senna italica* and *Bauhinia variegata*.

Sclerenchyma distribution

There are 4 cases: it is present as lightly in one layer of cells, which is adjacent to the upper epidermis in the distal region of *Senna alexandrina*; heavy in one layer of cells adjacent to the epidermis in the medial and distal region of *Cassia fistula*; heavy in many cell layers of the cortical and pith in all regions of the petiole of *Ceratonia siliqua* and *Bauhinia variegata*; and absent in the rest of species. This character seems to be informative at the species level and useful for identification of some of the investigated taxa. This result agrees with Shaheen (2006), who reported the importance of this character in the identification of the some mimosoid species.

Cluster crystal distribution

There are 4 cases: confined to cortical and pith cells of petiole regions in species of *Cassia*, confined to cortical cells in *Ceratonia* and *Bauhinia*, confined to pith cells in *Senna* (except *S. occidentalis*), and completely absent in *Delonix elata*. Thus, the distribution of cluster crystals, as viewed under LM with high magnification, revealed a similar pattern among the closely related species of *Senna* as well as of *Cassia*, and seems to be very useful at the generic level. These results agree with those published by Shaheen (2006), who reported the importance of this character at the generic level in the study of the taxonomy of some mimosoid species. In this respect, Boughton (1986) reported that the distribution of some kinds of crystals in the outer mesophyll and pith cells of

phyllodes in some Australian *Acacia* is considered a useful taxonomic criterion for delimiting the infrageneric groups.

Trichome types

There are 3 cases: multi-cellular in all species of *Cassia* and *Bauhinia variegata*, unicellular in 2 species of *Senna*, glandular in *Ceratonia siliqua*, and absent in *Delonix elata*. Herein, and for the precise distinction of the species investigated, trichomes may be of taxonomic interest at the generic level. The result agrees with Shaheen (2006), who reported the efficacy of the petiole trichomes in the delimitation and the identification of some mimosoid species. In addition, the result shows that the taxonomical status of *Cassia spectabilis* is not stable; it has some petiole characteristics similar to *Cassia*, as well as *Senna*. In this respect, Tomlinson (1982, 1984) listed many genera for which trichomes features are diagnostic.

Vascular trace shape in the medial region

The shape of the vascular trace in the medial part of the petiole, as seen in transections, varies among the studied species: radially and symmetrically in 4 species (e.g., *Cassia nodosa*, *Senna alexandrina*), U-shaped in *Senna italica* and *Delonix elata*, heart-shaped in *Cassia javanica*, compressed in *Ceratonia siliqua*, box-shaped in *Bauhinia variegata*, and cup-shaped in *Senna siamea* and *Cassia specabilis*. Thus, this character seems to be useful for the identification of some species, such as *Cassia javanica*, *Ceratonia siliqua*, and *Bauhinia variegata*.

Conclusion

The variability, in terms of taxonomic significance, in leaf trace structure characteristics of the studied species was investigated using LM.

The number of secondary bundles in the proximal, medial, and distal regions of the petiole represents a fairly

consistent attribute among species of each genus, and thus it may constitute a reliable criterion for the distinctiveness between different genera. Moreover, the variation in the number of secondary bundles among *Cassia* and *Senna* species proved to be useful for the identification of some species in each genus.

Although the number of abaxial and adaxial bundles, in general, is frequently an inconsistent attribute, and they are of minor taxonomic significance, the number of abaxial bundles is diagnostic at the generic level and seems to be of great value at the species level.

Senna alexandrina shows no change throughout its anatomical petiole trace. In contrast, some species show minor change in leaf trace structure from the proximal to distal regions, while some others have major change along the petiole.

The petiole trace of *Cassia italica* and *Bauhinia variegata* have an abnormal complexity of the vascular trace, having accessory bundles (concentric) at the core of the trace in addition to the normal bundles.

The moving of *Ceratonia* from the tribe Cassieae to Detarieae has been supported by using some characters of this study, such as the number of adaxial bundles, abaxial bundle division, sclerenchyma distribution, cluster crystals distribution, trichome types, and vascular trace shape.

In conclusion, it could be stated that useful taxonomic evidence has been attained from studying the petiole trace structure and documenting the change in the petiole anatomy from proximal to distal regions of some caesalpinoid species. Many additional characteristics for caesalpinoids have been revealed. Some of these characteristics are useful for identification and assessment of the taxonomic relationships among species studied at the generic and species levels, as well as for assessing their phylogenetic relationships.

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