

## Anatomical structures of vegetative and reproductive organs of *Senna occidentalis* (Caesalpinaceae)

Mohamed Abdel Aziz Ahmed NASSAR, Hassan Ramadan Hassan RAMADAN, Hend Mohammad Saad IBRAHIM\*

Department of Agricultural Botany, Faculty of Agriculture, Cairo University, Gamaa St., Giza, Egypt

Received: 30.11.2011

Accepted: 21.11.2012

Published Online: 15.05.2013

Printed: 30.05.2013

**Abstract:** The current investigation is concerned with the histological features of *Senna occidentalis* (L.) Link (coffee senna plant). The anatomical structure of different vegetative and reproductive organs was investigated weekly or fortnightly, according to the investigated organ, throughout the growing season. Organs studied included the main root, main stem (represented by shoot apex, apical, and median internodes), different foliage leaves developed on the main stem and on lateral shoots (including lamina and petiole), epidermal peel and trichomes, flower buds, fruits, and seeds. Histological features of various organs of the coffee senna plant were analysed microscopically and photomicrographed. Moreover, ultrastructural features of the seed coat were investigated by scanning electron microscope.

**Key words:** *Senna*, Caesalpinaceae, anatomy, vegetative organs, reproductive organs

### 1. Introduction

*Senna occidentalis* (L.) Link, commonly known as coffee senna, is typically a shrub native to America that is widespread in warm areas of the world, except for Australasia. Coffee senna is used as a flowering shrub for landscape purposes (Hussein, 2003). It is also used as a coffee substitute and has some medicinal uses. When seeds are brewed into the coffee-like beverage it is used to treat asthma. Previous pharmacological investigations showed that *Senna occidentalis* leaf extracts have broad spectrum antibacterial, antimalarial, antifungal (Caceves et al., 1991; Perez & Suarez, 1997; Tona et al., 1999), antimutagenic, antiplasmodial, anticarcinogenic, and hepatoprotective activity (Sharma et al., 2000; Tona et al., 2004; Vashishtha et al., 2009; Yadav et al., 2009). It is also used for stomach disorders, rheumatism, and the treatment of liver diseases (Sara et al., 1994; Jafri et al., 1999). The leaves are widely used as a leaf vegetable and are eaten either raw or mixed with coconut, chilli, and onion (Selvam, 2007; Vashishtha et al., 2009). In addition, coffee senna has been used to reduce the number of mosquitoes indoors at night (Paisson & Jaenson, 1999) and for the control of a large variety of insects (Dweivedi & Kumar, 1998).

Most of the *Senna* Mill. taxa, including the species under study, belonged to the genus *Cassia* L. until reassigned by Irwin and Barneby (1982) to follow the genus *Senna*; however, this process is not entirely complete, and some

corrections may still take place. Thus, any new information about *Senna* plants should be welcomed.

Previously, morphology of the vegetative and reproductive growth of the coffee senna plant was investigated throughout the consecutive stages of its life span (Nassar et al., 2011). Accordingly, the present study aimed to shed more light on the anatomical structure of vegetative and reproductive organs of the plant during successive stages of its life span in order to complement the phyto-graphic study of the coffee senna plant initiated by Nassar et al. (2011).

Most of the available literature deals with the anatomical structure of leguminous seeds, especially seed coat, in general, and some *Senna* species in particular. These studies include Bhalla and Slattery (1984) on clover; Miller et al. (1999), Ma et al. (2004), Moise et al. (2005), Shao et al. (2006), and Miller et al. (2010) on soybean; Souza and Marcos-Filho (2001) on some members of Fabaceae; Srivastava et al. (2006) on *Cassia angustifolia* M.Vahl.; Rodriguez-Pontes (2007) on *Senna corymbosa* (Lam.) Irwin and Barneby; and Fritz et al. (2009) on some *Senna* species. Moreover, Sahai (1999, 2001) studied seed coat sculpture in some *Cassia* species by scanning electron microscope (SEM). However, fewer investigators studied the anatomical characteristics of the family Caesalpinaceae (Essau, 1977; Metcalfe & Chalk, 1979; Fahn, 1990). Some investigators studied the anatomical structure of the

\* Correspondence: hendmoh@gmail.com

vegetative organs of some *Senna* species (Ibrahim, 1996; Kotresha & Seethram, 2000; Shaheen, 2007; Kumar, 2009; Ogundipe et al., 2009; Saheed & Illoh, 2010).

Obviously, new information about different botanical aspects of this species, which is of great interest from a medicinal point of view, is required.

## 2. Materials and methods

The present investigation was carried out in the wire greenhouse of the Cairo University Agricultural Botany Department, Faculty of Agriculture, Giza, Egypt, during the 2009 growing season to investigate the anatomical structure of the vegetative and reproductive organs of the coffee senna plant over its life span.

Seeds of coffee senna were sown on 19 March 2009 to provide the experimental plant materials. The experiment included 3 replicates; each was represented by 1 plot. The plot was 3 × 3 m with 5 ridges 60 cm apart. Seeds were sown in hills spaced 30 cm apart. The plants were thinned to 1 plant per hill. All field practices were carried out as recommended for this experimental plant in this particular location.

Microtechniques were carried out in the laboratory of the Cairo University Agricultural Botany Department, Faculty of Agriculture. A full microscopic study was performed to investigate the histological structure of the coffee senna plant. Samples were taken periodically (weekly or fortnightly) throughout the growing season. Specimens represented different plant organs and included:

- 1- The main root through its basal portion.
- 2- The main stem, represented by the shoot apex, apical internode, and median internode.
- 3- Different compound foliage leaves developed on the main stem and lateral branches, represented by the petiole and lamina of the middle leaflets pair.
- 4- Leaf epidermal peels (to examine the type of stomata).
- 5- Flower buds.
- 6- Mature green pods and seeds.

The microtechnique procedures given by Nassar and El-Sahhar (1998) were followed. Specimens were fixed at least 48 h in FAA (10 mL of formalin, 5 mL of glacial acetic acid, 50 mL of ethyl alcohol 95%, and 35 mL of distilled water). After, fixation materials were washed in 50% ethyl alcohol at least twice and then dehydrated in a normal butyl alcohol series before embedding in paraffin wax (melting point 56–58 °C). Cross and longitudinal sections were cut on a rotary microtome to a thickness of 20 µm and were stained with either safranin/light green or a crystal violet/erythrosine combination before mounting in Canada balsam and covering. Epidermal peels were stained with an aqueous solution of erythrosine, mounted in Canada balsam, and covered. Slides were analysed microscopically and photomicrographed.

## 2.1. Scanning electron microscope

Ultrastructural features of seed surfaces were examined by scanning electron microscopy at 25 kV. Mature dried seeds were mounted with SP1 supplies, conducting carbon paints on copper stubs, and then coated with a thin layer of gold palladium using Edwards sputter coater unit S 150 B. The seeds were examined by SEM (model: JEOL-JSM-T100) at the Cairo University Applied Center for Entomonematodes, Agricultural Experiments and Researches Station, Faculty of Agriculture, Giza, Egypt.

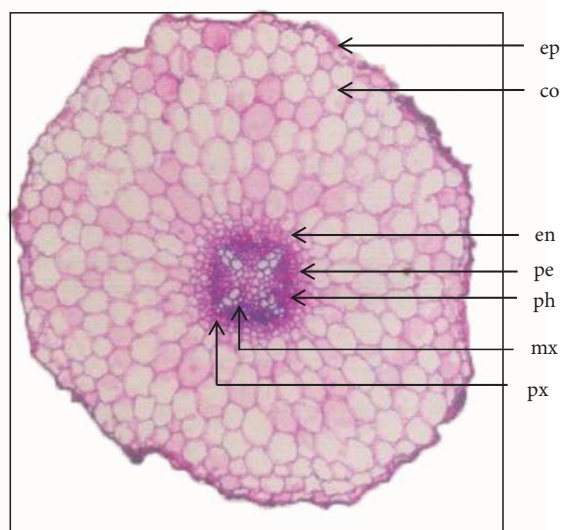
SEM-micrographs at various magnifications were used to elucidate the seed morphological features of the studied species. Depending upon the size of seeds, magnification power between 35 and 3000× was selected with the aim of showing the finest possible detail in seed structure. Terms provided by Murley (1951) and modified by Stearn (1983) were used.

## 3. Results and discussion

### 3.1. Structure of the main root

The histological structure of the *Senna occidentalis* root system was investigated through transverse sections of the main root at different stages of growth.

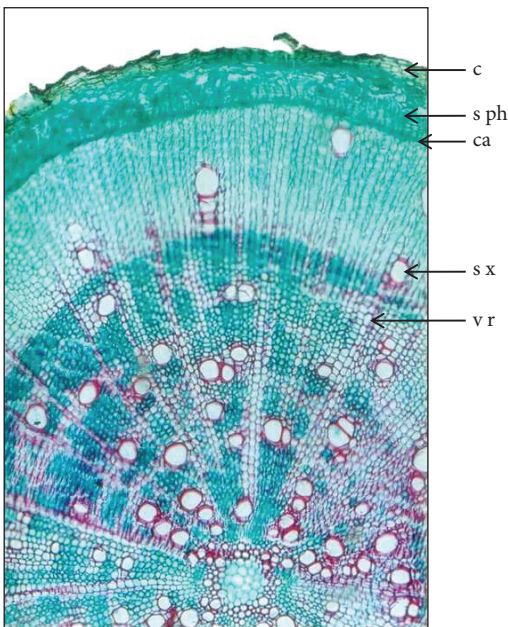
The transverse section through the main root of a 7-day-old seedling (Figure 1) shows an obvious distinction among the 3 main tissue systems: the epidermis, cortex, and vascular tissue systems. The main root, at 7 days, is in the primary stage of growth. The epidermal layer consists of uniseriate cells that are irregular in shape with thin



**Figure 1.** Transverse section of the main root of *Senna occidentalis* at 7 days, showing its primary structure (40×). co = cortex, en = endodermis, ep = epidermis, mx = metaxylem, pe = pericycle, ph = phloem, and px = protoxylem.

walls that lack cuticle and stomata. Some of the epidermal cells extend out in the form of tubular, unicellular root hairs. The cortex is composed of about 7 layers of thin-walled, irregular parenchyma cells with conspicuous intercellular spaces. Moving inward, cell size increases, and it reaches its maximum diameter in the middle of the cortex before decreasing again. The innermost layer of the cortex is the endodermis, a uniseriate layer of small, rectangular-shaped cells that form a distinct layer surrounding the stele. The Casparian strips are hardly observed in transverse sections. The pericycle is a single layer of thin-walled parenchymatous cells just within the endodermis and peripheral to the vascular tissues. As seen in transverse sections, the vascular system of the root is radial. Xylem and phloem occur in separate patches arranged on alternate radii and are intervened by small parenchyma cells. The parenchyma found in between the xylem and phloem patches is known as conjunctive tissue. The vascular bundle is tetrarch since 4 patches of xylem alternate with a similar number of phloem patches. The xylem is exarch, or protoxylem (about 3–4 vessels), and is situated towards the periphery; metaxylem (about 4–5 vessels) lies towards the centre, thus showing a centripetal mode of differentiation from the procambium. The phloem is characterised by the absence of fibres and the presence of sieve tubes, companion cells, and phloem parenchyma. The pith is narrow (a few cells).

The main root of 10-week-old plants (Figure 2) shows an increase in the diameter of the root due to increasing

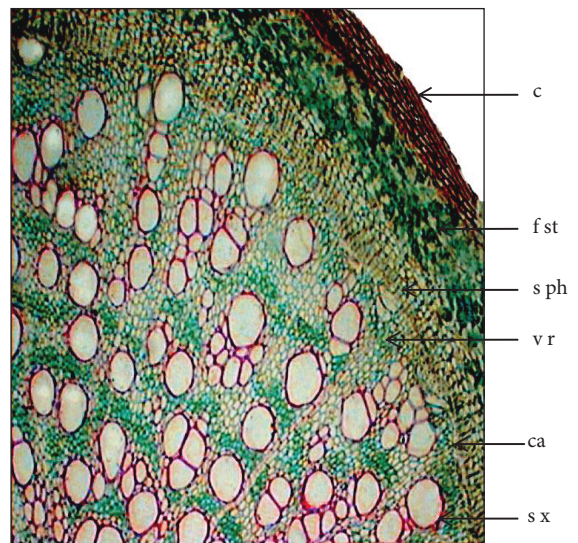


**Figure 2.** Transverse section of the main root of *Senna occidentalis* at 10 weeks, showing its secondary structure (40×). c = cork, ca = cambium, s ph = secondary phloem, s x = secondary xylem, and v r = vascular ray.

secondary growth. The intensive increase of vascular tissue circumference results in the rupture of the epidermis and cortex and an increase in a continuous, well-defined periderm from the pericycle. The periderm tissue is well-developed at this stage of secondary growth. It is composed of 7–10 layers of rectangular cells; 2–4 of these layers are phellem (cork), 2–3 layers towards the centre form the phellogen, and the inner 6 layers constitute the phelloderm. The secondary phloem strands are found opposite the secondary xylem rows or groups and are composed of sieve tubes and their companion cells in addition to phloem parenchyma. The vascular cambium consists of 4–5 layers. Vessels of the secondary xylem are in radial rows of 6–10 vessels (uniseriate) or 14–16 vessels in radial clusters (groups). The vessels of secondary xylem are surrounded by groups of fibres and parenchyma cells. The primary xylem occupies the centre of the root.

When plants are 16 weeks old (Figure 3) the secondary thickening is more developed, and the root comprises mainly a vascular cylinder surrounded by a periderm. The periderm tissue is composed of 5–7 layers of phellem, 2–3 layers of phellogen (cork cambium), and 4–6 layers of phelloderm. The amount of secondary elements increases, and there is more xylem than phloem.

The previously given histological information regarding the main root of *Senna occidentalis* is mostly in accordance with that of the main root of *S. occidentalis* as described by Kumar (2009). It disagrees, however, with some of his description. Kumar (2009) described the vascular bundle as diarch while this study found it to be tetrarch.



**Figure 3.** Transverse section of the main root of *Senna occidentalis* at 16 weeks, showing an advanced stage of secondary growth (64×). c = cork, ca = cambium, f st = fibre strand, s ph = secondary phloem, s x = secondary xylem, and v r = vascular ray.



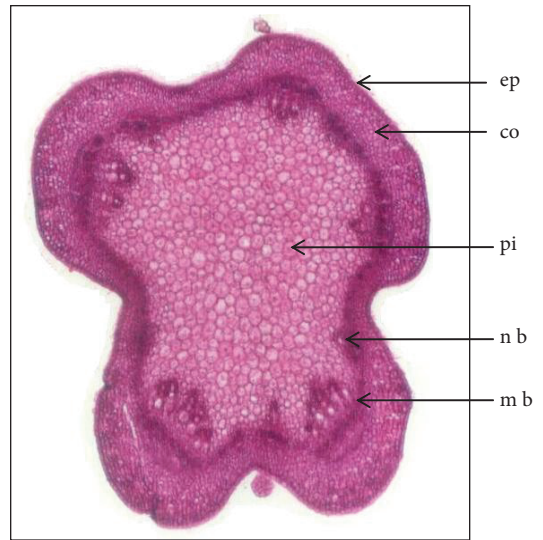
### 3.2. Structure of the main stem

#### 3.2.1. The shoot apex

The dome-shaped, apical meristem of *Senna occidentalis* is composed of a 2-layered tunica (30.4  $\mu\text{m}$ ) overlying the corpus (139.6  $\mu\text{m}$ ) (Figure 4). The shoot apex above the first discernible leaf primordium averages 145  $\mu\text{m}$  in height and 160  $\mu\text{m}$  in diameter. The average number of cells across this region is 16–18. The distance below the tip where the procambium first differentiates is 169.2  $\mu\text{m}$ . The average height of the shoot apex above the first vascular tissue to differentiate is 312.5  $\mu\text{m}$ , and the diameter of pith across this particular region is about 375  $\mu\text{m}$ . Vacuolation is observed at 380  $\mu\text{m}$  below the promeristem.

#### 3.2.2. The internode directly below the shoot apex

The main stem of *Senna occidentalis* below the shoot apex at 4 weeks (Figure 5) is pentagonal in outline. The epidermis is composed of uniseriate, nearly square parenchymatous cells with a thin layer of cuticle and glandular trichomes emerging from the epidermal cells. The cortex consists of 7–10 layers of cells; the outer 2–3 layers are collenchymatous cells underlying the epidermis, and the remainder are polygonal parenchymatous cells with small intercellular spaces. The starch sheath is hardly recognisable. The vascular collateral bundles are arranged in a ring. There are 5 major bundles at the ridges and 5 minor ones occupying the distance (in the furrows) between any 2 of them. Each major bundle has 20–25 vessels, while minor bundles have 3–4 vessels. The cambial zone consists of 2–3 layers of thin-walled rectangular cells. The phloem is composed of sieve tubes, companion cells, and phloem parenchyma. The pith occupies a large portion in the centre of the section and consists of polygonal parenchymatous cells with relatively small intercellular

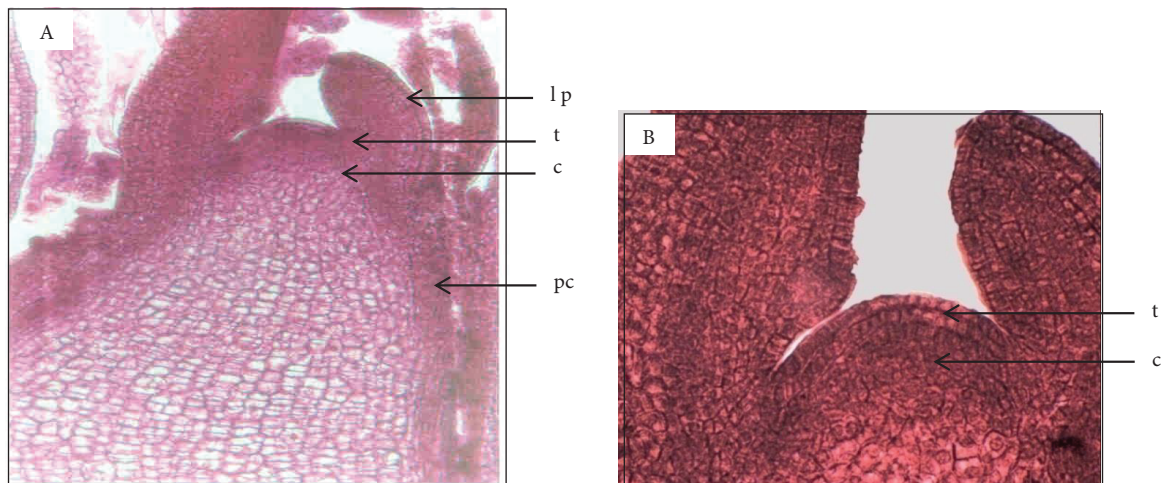


**Figure 5.** Transverse section of internode directly below the shoot apex of *Senna occidentalis* at 4 weeks (50 $\times$ ). co = cortex, ep = epidermis, m b = major bundle, n b = minor bundle, and pi = pith.

spaces. The pith is connected with the cortex through medullary rays that are 5–7 rows wide.

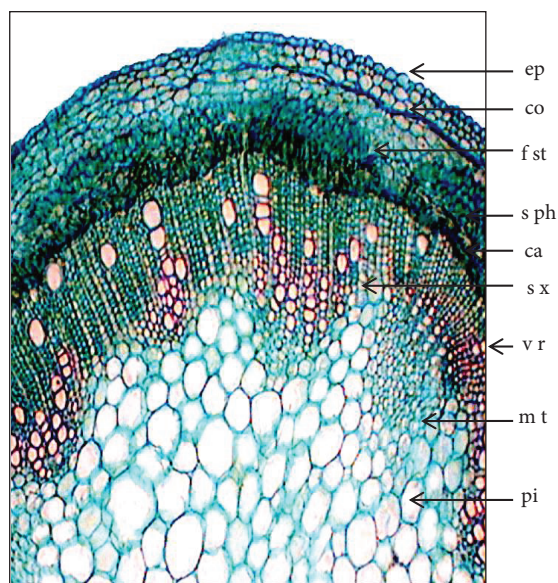
#### 3.2.3. The median internode of the main stem

The middle part of the *Senna occidentalis* main stem at 6 weeks (Figure 6) loses its characteristic outline since ridges and furrows disappear and become circular. The epidermis is composed of a single layer of radially and tangentially elongated cells covered with a thick cuticle. The cortex consists of 6–8 layers of cells. The outer 2–3 layers are collenchymatous cells beneath the epidermis, and the remainder are thin-walled parenchymatous cells with



**Figure 4.** Longitudinal median section of shoot apex of *Senna occidentalis* showing the 2-layered tunica and corpus, differentiation of the procambium, and different stages of leaf development. A = whole section (52 $\times$ ) and B = magnified portion of A (400 $\times$ ); c = corpus, lp = leaf primordium, pc = procambium, and t = tunica.

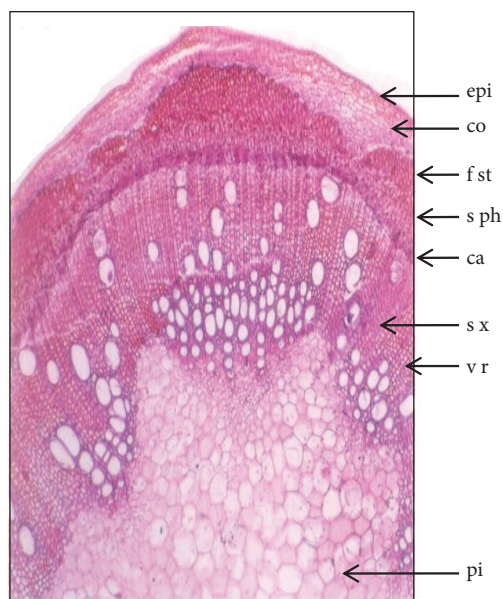




**Figure 6.** Transverse section of the median internode of the main stem of *Senna occidentalis* at 6 weeks (64 $\times$ ). ca = cambium, co = cortex, ep = epidermis, f st = fibre strand, m t = mechanical tissue, pi = pith, s ph = secondary phloem, s x = secondary xylem, and v r = vascular ray.

intercellular spaces. Chlorenchymatous cells are found in 2 regions; either immediately below the epidermis (outer) or below the layers of collenchymatous of cells (inner). The starch sheath is barely observable. Solitary and druse crystals are present in some cortical cells. The stele consists of 28–30 collateral bundles, which are arranged in a ring and vary in size; major bundles of 40–50 vessels and minor ones of 17–20 vessels. Xylem elements contain vessels, fibres, and paratracheal parenchyma. The phloem consists of 4–5 strands including sieve tubes, companion cells, and phloem parenchyma. Some phloem tissue cells contain druse crystals. The cambial zone is composed of 2–3 tiers of thin-walled rectangular cells separating phloem and xylem. A fibrous group abuts the outside of each bundle, and the interfascicular cambium provides new bundles. The pith is formed of polygonal parenchymatous cells, which increase gradually in size towards the centre and have small, triangular intercellular spaces among them. The pith contains druse crystals in some cells. Medullary rays comprise of 4–8 lignified parenchyma cells that separate the bundles and connect the cortex and pith.

When plants are 10 weeks (Figure 7) the secondary thickening proceeds, and secondary growth is present in a continuous cylindrical form, especially in the secondary xylem. The vascular cylinder is surrounded by an interrupted ring of extraxylary fibres. The vascular bundles are arranged in a ring. They vary in size and are embedded in lignified parenchyma cells. Vascular rays consist of uniseriate and multiseriate lignified cells. The secondary



**Figure 7.** Transverse section of median internode of the main stem of *Senna occidentalis* at 10 weeks (40 $\times$ ). ca = cambium, co = cortex, epi = epidermis, f st = fibre strand, p = pith, s ph = secondary phloem, s x = secondary xylem, and v r = vascular ray.

phloem contains sieve tubes, companion cells, and phloem parenchyma. Phloem fibres are absent. The xylem vessels are embedded in lignified parenchymatous cells and present either singly or in radial groups consisting of 5–8 vessels in major vascular bundles and 4–5 vessels in minor ones. The secondary xylem elements contain vessels, fibres, and paratracheal parenchyma. The pith consists of slightly polygonal parenchymatous cells that contain druse crystals.

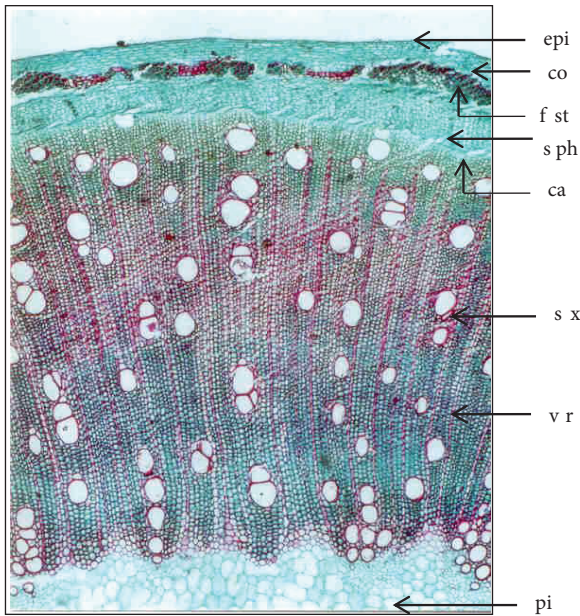
When plants are 16 weeks old (Figure 8) the epidermal cells are pressed and covered with a thick layer of cuticle. It is free from trichomes and ragged in places where the subepidermal cork is formed. The secondary thickening continues, and secondary growth is present in a continuous cylindrical form, especially in the secondary xylem. The vascular cylinder is surrounded by an interrupted ring of extraxylary fibres. The primary xylem is found abutting the pith, and it is difficult to recognise the primary phloem.

The aforementioned description of the histological structure of the main stem is in agreement with Metcalfe and Chalk (1979) regarding stems of some genera of Caesalpiniaceae. Ibrahim (1996) and Kumar (2009) on some *Senna* and *Cassia* species are also in harmony with the present findings.

### 3.3. Structure of the leaf

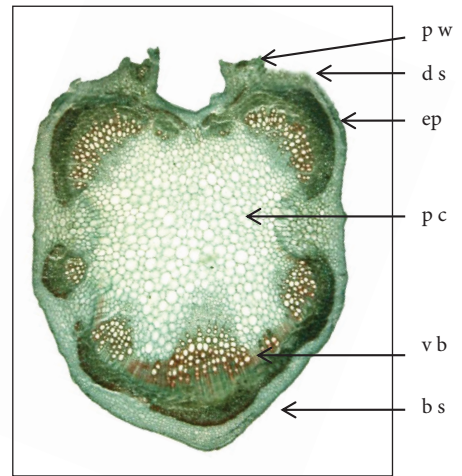
#### 3.3.1. Leaf petiole

The petiole of *Senna occidentalis* (Figure 9) is surrounded by a uniseriate epidermis of nearly rectangular cells that



**Figure 8.** Transverse section of median internode of the main stem of *Senna occidentalis* at 16 weeks (flowering onset) (52×). ca = cambium, co = cortex, ep = epidermis, f st = fibre strand, pi = pith, s ph = secondary phloem, s x = secondary xylem, and v r = vascular ray.

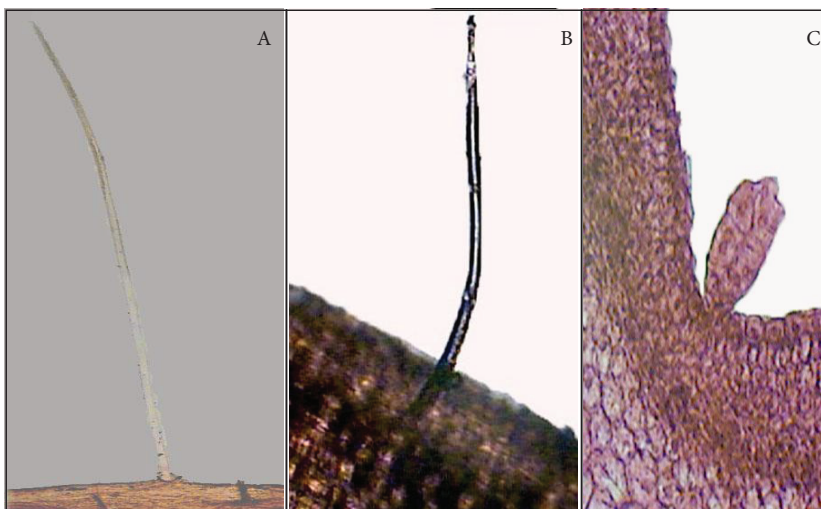
are both radially and tangentially elongated with a thin cuticle layer, and bear trichomes (Figure 10), mostly of the unicellular type (i.e. only 1, very long cell). There are rarely trichomes of the multicellular type (i.e. many cells, which are usually short). The petiole is obovate with 2 ridges and an adaxial wide groove. The cortex is formed by 2 types of cells: an outer chlorenchyma and an inner parenchyma. The main petiolar vasculature in the



**Figure 9.** Transverse section of *Senna occidentalis* petiole (90×). b s = abaxial side, d s = adaxial side, ep = epidermis, p c = parenchyma cells, p w = petiole wing, and vb = median vascular bundle.

form of 7 separate vascular bundles: 3 dorsals, 2 laterals, and 2 ventrals (3 + 2 + 2). There are 2 ridge bundles. A dissected ring of sclerenchymatous cells surrounds the main petiolar vasculature. The pith is wide and formed of parenchymatous cells. Druse crystals are present.

The above description of the petiole is generally in agreement with Ibrahim (1996), Shaheen (2007), Ogundipe et al. (2009), and Saheed and Illoh (2010) regarding leaf petioles and trichomes of *Senna occidentalis*. It is also in harmony with the description given by Albert and Sharma (2013) regarding trichome types in several *Bauhinia* species, some closely related members of leguminosae.



**Figure 10.** Photograph showing types of trichomes of *Senna occidentalis*. A = unicellular trichome on the leaf petiole (132×), B = multicellular trichome on the leaf petiole (64×), and C = glandular trichome on the apical internode (64×).



**3.3.2. The leaf blade (lamina)**

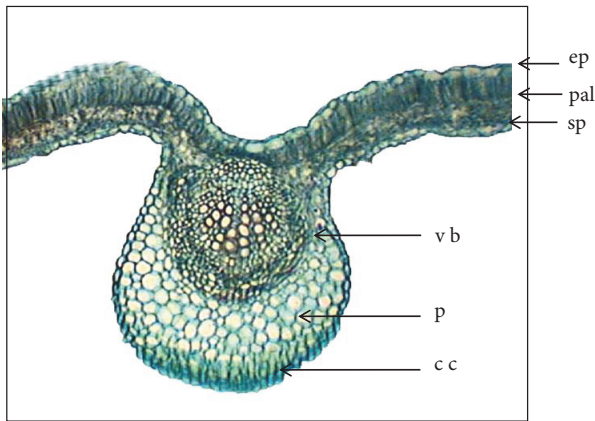
The anatomical structure of leaflet blades representing mature foliage compound leaves of *Senna occidentalis* at different stages of plant growth was also investigated (Figure 11). Nearly all investigated compound leaves have the same structure. The mesophyll is of the dorsiventral type; the palisade tissue is located on the adaxial side of the blade and the spongy tissue on the abaxial one. Upper epidermal cells are thin-walled, tangentially elongated, and covered with a thin layer of cuticle. Lower epidermal cells are radially elongated. Stomata (Figure 12) are more numerous on the lower epidermis than on the upper. It is composed of both paracytic and anisocytic types. In the paracytic type each guard cell is accompanied by one or more subsidiary cells, the longitudinal axes of which are parallel to those of the guard cells and aperture. In the anisocytic type the guard cells are surrounded by 3 unequally sized subsidiary cells

(Fahn, 1990). The midrib region has a shallow furrow adaxially, and the lower epidermis is convex. Palisade tissue consists of 1–2 adaxial layers, is full of chloroplasts, and occupies one-half of the entire thickness of the mesophyll. It is worth noting that palisade tissue is extended in the midrib region. The spongy tissue is composed of 4–5 layers of loosely arranged chlorenchymatous cells with many wide intercellular spaces. Mechanical tissue in the form of annular collenchyma cells situated abaxially in the midrib region and sclerenchymatous cells, which surround the midvein vascular supply in the form of a continuous ring, thicken at the adaxial side. Thus, the included bundle, the principle one, is not directly embedded in the mesophyll as the smaller ones are. The midrib includes a large main bundle. The xylem is located on the adaxial side of the midvein and consists of about 6–8 parallel rows, each with 3–5 vessels. The phloem is found at the abaxial side of the bundle and is composed of sieve tubes, companion cells, and phloem parenchyma. Solitary and druse crystals are present.

The previously mentioned structure of *Senna occidentalis* leaf blade is in harmony with Ibrahim (1996), Kotresha and Seetharam (2000), Kumar (2009), Ogundipe et al. (2009), and Saheed and Illoh (2010) regarding leaf blades of *Senna occidentalis*. The above description of stomata types present in *Senna occidentalis* agrees with Albert and Sharma (2013) regarding some *Bauhinia* species, a closely related legume.

**3.4. Structure of the flower bud**

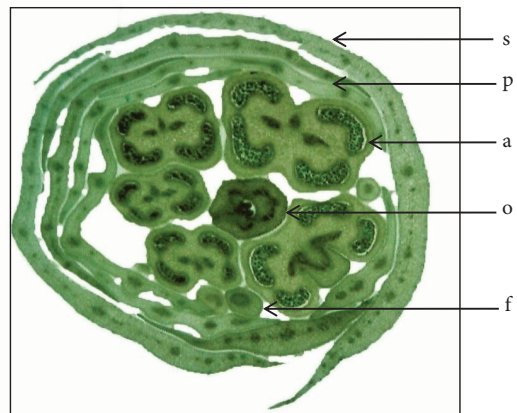
Figure 13 reveals that there is no bract around the flower of *Senna occidentalis*; a bract exists but abscises earlier during flowering. The calyx consists of 5 free sepals (polysepalous). Each sepal consists of 2 epidermal layers. The upper epidermis has a uniseriate layer of barrel-shaped cells covered with a thin cuticle layer and



**Figure 11.** Transverse section of mature leaf blade of *Senna occidentalis* through the midrib at 16 weeks (64×). c c = collenchyma cells, ep = epidermis, pal = palisade tissue, p c = parenchyma cells, sp = spongy tissue, and v b = vascular bundle.



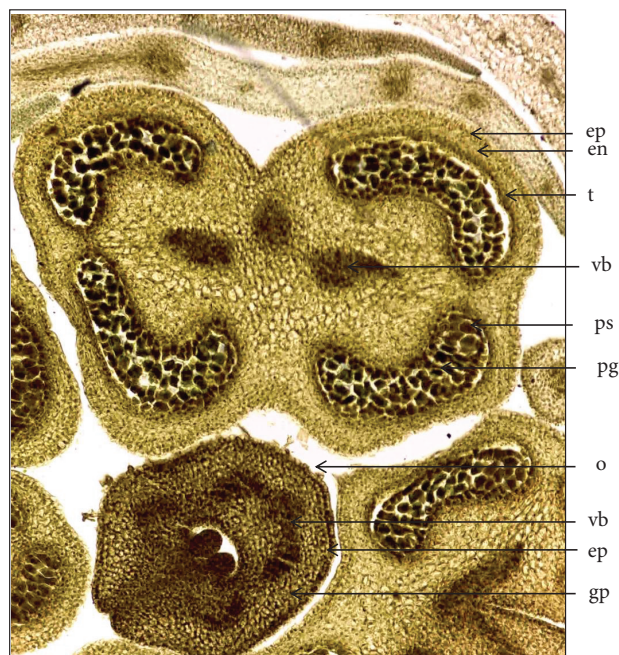
**Figure 12.** Epidermal peel showing the paracytic and anisocytic stomata types developing through the leaf blade of *Senna occidentalis* (400×). as = anisocytic stoma, ep = epidermis, g c = guard cell, pa = paracytic stoma, and s c = subsidiary cell.



**Figure 13.** Transverse section of floral bud of *Senna occidentalis* at early flowering (90×). a = anther, f = filament, o = ovary, p = petal, and s = sepal.



stomata in between 8–10 layers of ground chlorenchyma; there are obvious intercellular spaces. There are 25 small vascular bundles that extend through the ground tissue. The corolla consists of 5 free petals (polypetalous). Each petal consists of 2 epidermal layers with nearly square parenchyma cells and 3–5 layers of ground parenchyma in between. There are 5–9 traces extending through the ground tissue. The androecium consists of 10 stamens (3 large, 4 medium-sized, and 3 small) arranged in 2 whorls. Filaments are straight, shorter than or not more than twice as long as the basifixed anthers, which are dehiscent by apical pores or short slits. The anther is situated on the slender filament (basifixed) of a single vascular bundle that culminates blindly in the connective tissue situated in between the 2 anther lobes (2 lobes, 4 locules). The outermost wall of the anther (Figure 14) is the epidermis. Just beneath the epidermis there is an endothecium that possesses strips or ridges of secondary wall material. The innermost layer is composed of multinucleate cells (tapetum). Pollen grains have sculptures and 3-germ splits (tricolporate) on their walls (Perveen & Qaiser, 1998). The gynoecium is monocarpellary, the placentation marginal, campylotropous ovule; the style is short, and the stigma is small and terminal. The ovary wall consists of an outer epidermis with nearly square cells, followed by 10–12 layers of ground parenchyma that contain 5–7 vascular bundles. The inner epidermis has large barrel-shaped



**Figure 14.** Transverse section of floral bud of *Senna occidentalis* showing a magnified view of the anther and ovary (100×). en = endothecium; ep = epidermis, gp = ground parenchyma, o = ovary, pg = pollen grain, ps = pollen sac, t = tapetum, and vb = vascular bundle.

cells; the ovary is unilocular with numerous ovules. The previous description of the flower bud structure agrees with Subrahmanyam (1995).

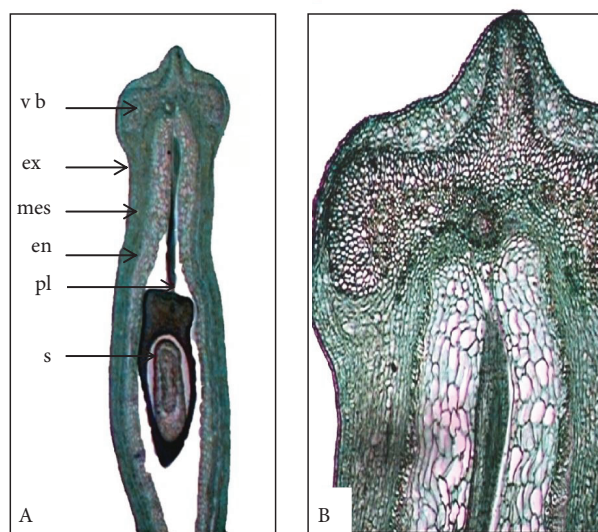
### 3.5. Structure of the fruit

Figure 15 represents the structure of *Senna occidentalis* fruit. The exocarp consists of an epidermis only; the outer epidermis has uniseriate, barrel-shaped cells with stomata and is covered with thick layer of cutin. The ground tissue is mesocarp and consists of 10 layers of irregular, thin-walled parenchyma cells lying next to the exocarp. The component parenchymatous cells of these layers are characterised by their rather thick and slightly lignified walls. They are tangentially elongated. The inner epidermis is endocarp and consists of uniseriate, cylindrical parenchyma cells. There are 4 large vascular bundles embedded in the ground parenchyma, and they are surrounded by a sheath of fibres and 15 layers of parenchyma cells where the 2 valves separate when the fruit ripens (abscission tissue).

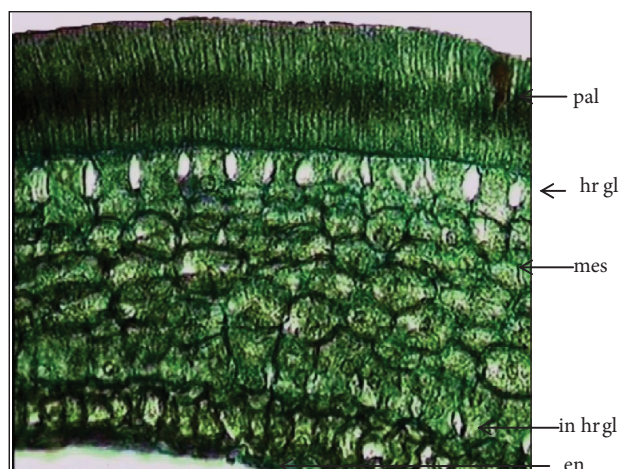
The above description of *Senna occidentalis* fruit is in general agreement with Esau (1977) and Fahn (1990).

### 3.6. Structure of the seed

The seed coat develops from the integuments that surround the ovule prior to fertilisation. The seed coat of *Senna occidentalis* (Figure 16) differentiates into 4 distinct layers. The outermost layer is the waxy cuticle, which represents the first barrier for imbibition. The next layer, the epidermis, is formed of thick-walled, elongated palisade cells called macrosclereids with the long axis oriented perpendicularly to the surface. A light refractive, apparently denser region distinguished by optical



**Figure 15.** Transverse section of the green fruit (pod) of *Senna occidentalis* showing the structure of the fruit and the seed. A = almost whole section (60×) and B = magnified portion of A (132×). en = endocarp, ex = exocarp, mes = mesocarp, pl = placentum, s = seed, and vb = vascular bundle.

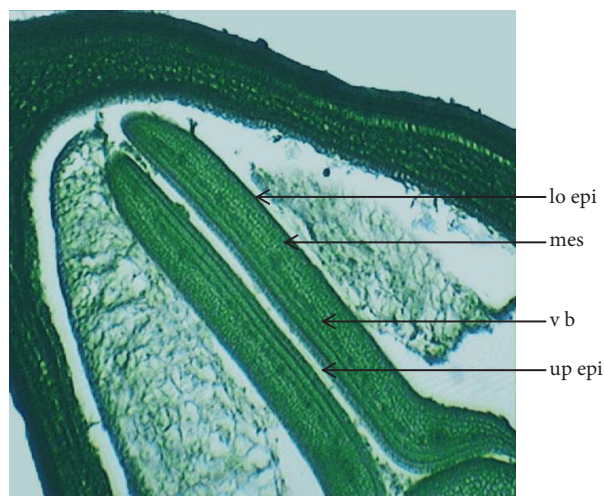


**Figure 16.** Transverse section of the green seed of *Senna occidentalis* showing the structure of the seed coat (360×). en = endosperm, hr gl = hourglass cells, in hr gl = inner hourglass cells, mes = mesophyll, and pal = palisade.

microscopes is called linea lucida or light line. Only 1 palisade layer is found throughout the testa, except under the hilum where 2 can occur; the external layer is called the counter-palisade and originates from the funiculus. A single layer of cells forms the hypodermis; the cells are also called hourglass cells, pillar cells, or osteosclereids. They are larger than adjacent cell layers and are separated by wide intercellular spaces, except under the hilum cleft, where they are absent. The fourth layer of the seed coat is the interior parenchyma, which is formed by 6–7 layers of thin-walled, protoplast-free, tangentially elongated parenchyma cells. The innermost layers are largely pressed and uniformly distributed throughout the whole testa, except in the area of the hilum, where a smaller number of layers can be distinguished. Inward from the mesophyll layer, irregularly arranged intercellular spaces indicate the presence of inner hourglass (endotestal) cells. These are followed by 3–4 layers of thin-walled cells that are considered a portion of the endosperm.

The seed of *Senna occidentalis* (Figure 17) shows that the embryo consists of 2 cotyledons and the axis, which has 4 parts: the radical, hypocotyl, epicotyl, and plumule. The cotyledons are parallel to the long axis of the embryo. The cotyledon consists of 2 epidermal layers of nearly square parenchyma cells. The mesophyll tissue is composed of 6–7 layers of thin-walled parenchyma cells with small intercellular spaces. The vascular bundles are embedded in the mesophyll tissue.

The previously mentioned structures of the *Senna occidentalis* seed coat and seed are in agreement with the findings of Bhalla and Slattery (1984) in clover (*Trifolium alexandrinum*) seeds, Serrato-Valenti et al. (1993) in *Stylosanthes scabra* seed (Leguminosae; Papilionoideae),



**Figure 17.** Transverse section of the immature seed of *Senna occidentalis* showing the structure of the cotyledons (64×). lo epi = lower epidermis, mes = mesophyll, up epi = upper epidermis, and v b = vascular bundle.

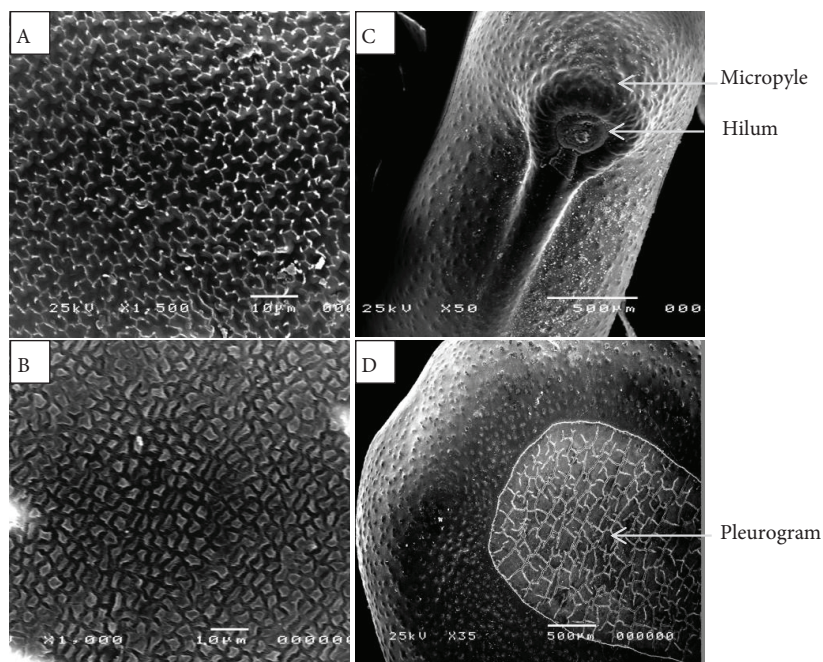
the description of the ultrastructure of seed and seed testa from 12 *Cassia* species in Sahai (2001), the studies of Souza and Marcos-Filho (2001) in Fabaceae, Srivastava et al. (2006) in *Cassia angustifolia* seeds, Rodriguez-Pontes (2007) in *Senna corymbosa* seed development, and Fritz et al. (2009) in seed testa of some *Senna* species. Similarly they are in general accordance with the studies of Moise et al. (2005), Miller et al. (1999, 2010), Ma et al. (2004), and Shao et al. (2006) on soybean (*Glycine max*) seed and seed coat. Recently, the description of the seed coat structure of *Senna occidentalis* found harmony with the work of Büyükkartal et al. (2013) on 2 varieties of *Vicia sativa*, a Fabaceae member.

### 3.7. Scanning electron microscopic investigations

SEM is proving to be an especially suitable tool for studying seed surfaces. It helps to detect the minute differences in seed coat patterns, which might help to define species characters. The morphological description of seed surface features by SEM successfully provided taxonomic evidence for the identification and classification of species and establishes phylogenetic relationships between them (Prasad & Singh, 1978).

The seed was subjected to scanning electron microscopy to provide more information about its surface features (Figure 18). The seed coat shows that the seed is ovate and almost round. The hilum dimensions are  $272.72 \times 227.27 \mu\text{m}$ , and it is oblate. The epidermal cells of the seed coat are irregular and tuberculate. The anticlinal walls are flat and irregular. The outer periclinal cell walls are flat with smooth, rounded projections. The micropyle is adjacent to the hilum.





**Figure 18.** Seed surfaces of *Senna occidentalis* as detected by SEM. A and B = seed surface sculpture pattern, C = hilum shape and micropyle position, and D = pleurogram.

The above description of *Senna occidentalis* seed structure is in accordance with Sahai et al. (1997) and Sahai (1999).

In summary, this study introduces a full anatomical map for *Senna occidentalis* (L.) Link from seed to seed and completes a phyto-graphic study for this taxonomically debated and economically and medicinally important

plant. The study showed that *Senna occidentalis* (L.) Link is a typical Caesalpiniaceae member of the genus *Senna* Mill. and is more distinguished from the genus *Cassia* L. to which it formerly belonged. The study delimits ambiguity concerning this species and is intended as an effort to guide researchers in the exploration of different aspects of the science related to this species.

## References

- Albert S & Sharma B (2013). Comparative foliar micromorphological studies of some *Bauhinia* (Leguminosae) species. *Turkish Journal of Botany* 37: 276-281.
- Arya V, Yadav S, Kumar S & Yadav JP (2010). Antimicrobial activity of *Cassia occidentalis* L. leaf against various human pathogenic microbes. *Life Science and Medical Research* 9: 1-11.
- Bhalla PL & Slattery HD (1984). Callose deposits make clover seeds impermeable to water. *Annals of Botany* 53: 125-128.
- Büyükkartal HN, Çölgeçen H, Pınar NM & Erdoğan N (2013). Seed coat ultrastructure of hard-seeded and soft-seeded varieties of *Vicia sativa*. *Turkish Journal of Botany* 37: 270-275.
- Caceves A, Lopez BR, Giron MA & Logemann H (1991). Plants used in Guatemala for the treatment of dermatophytic infections 1. Screening for antimycotic activity of 44 plant extracts. *Journal of Ethnopharmacology* 31: 263-276.
- Dweivedi SC & Kumar R (1998). Evaluation of *Cassia occidentalis* leaf extract on development and damage caused by *Trogoderma granarium*, Khapra beetle. *Journal of Ecotoxicology and Environmental Monitoring* 8: 55-58.
- Esau K (1977). *Anatomy of Seed Plants*, Second Edition. New York, USA: John Wiley and Sons.
- Fahn A (1990). *Plant Anatomy*, Fourth Edition. Oxford, UK: Pergamon Press.
- Fritz E, Olzant SM & Sautcel J (2009). Line of breakage in the testa of seeds of various species of the genus *Senna*. *Scientia Pharmaceutica* 77: 265.
- Hussein MMM (2003). Growth of *Senna occidentalis* (L.) Link in sandy soil as affected by fertilization and some amendments. *Cairo University, Bulletin of Faculty of Agriculture*, 54: 189-216.
- Irwin HS & Barneby RC (1982). The American *Cassiinae*, a synoptic revision of Leguminosae, tribe *Cassieae*, subtribe *Cassiinae* in the New World. *Memoirs of the New York Botanical Garden* 35: 1-918.
- Jafri MA, Subhani MJ, Javed K & Sigh S (1999). Hepatoprotective activity of leaves of *Cassia occidentalis* against paracetamol and ethyl alcohol intoxication in rats. *Journal of Ethnopharmacology* 68: 355-361.



- Kotresha K & Seetharam YN (2000). Epidermal micro-morphology of some species of *Cassia* L. (Caesalpinaceae). *Phytomorphology* 50: 229–237.
- Kumar A (2009). *Cassia occidentalis* Linn. Morphological and anatomical study. *Science 2.0*, ION Publications LLC.
- Ma F, Cholewa E, Mohamed T, Peterson CA & Gijzen (2004). Cracks in the palisade cuticle of soybean seed coats correlate with their permeability to water. *Annals of Botany* 94: 213–228.
- Metcalf CR & Chalk L (1979). *Anatomy of the Dicotyledons* Vol. 1. Oxford, UK: The Clarendon Press.
- Miller SS, Bowman LA, Gijzen M & Miki BLA (1999). Early development of the seed coat of soybean. *Annals of Botany* 84: 297–304.
- Miller SS, Jin Z, Schnell JA, Romero MC, Brown DCW & Johnson DA (2010). Hourglass cell development in the soybean seed coat. *Annals of Botany* 106: 235–242.
- Moise JA, Han S, Gudynaite-Savitch L, Johnson DA & Miki BLA (2005). Seed coats: structure, development, composition, and biotechnology. *In Vitro Cellular and Developmental Biology-Plant* 41: 620–644.
- Murley MR (1951). Seed of the Cruciferae. *American Midland Naturalist* 46: 1–18.
- Nassar MA & El-Sahhar KF (1998). *Botanical Preparations and Microscopy (Microtechnique)*. Giza, Egypt: Academic Bookshop.
- Nassar MAA, Ramadan HRH & Ibrahim HMS (2011). Morphological characteristics of vegetative and reproductive growth of *Senna occidentalis* (L.) Link (Caesalpinaceae). *Research Journal of Agriculture and Biological Sciences* 7: 260–270.
- Ogundipe OT, Kadiri AB & Adekanmbi OH (2009). Foliar epidermal morphology of some Nigerian species of *Senna* (Caesalpinaceae). *Indian Journal of Science and Technology* 2: 5–10.
- Paisson K & Jaenson TGT (1999). Plant products used as mosquito repellents in Guinea Bissau, West Africa. *Acta Tropica* 72: 39–52.
- Perez C & Suarez C (1997). Antifungal activity extracts against *Candida*. *American Journal of Chinese Medicine* 25: 181–184.
- Perveen A & Qaiser M (1998). Pollen flora of Pakistan - X. Leguminosae (subfamily: Caesalpinioideae). *Turkish Journal of Botany* 22: 145–150.
- Prasad T & Singh D (1978). Gametophytes and seed development in *Nicotiana physaloides*. *Journal of the Indian Botanical Society* 57: 78–83.
- Rodriguez-Pontes M (2007). Development of megagametophyte, embryo, and seed in *Senna corymbosa* (Lam.) H.S. Irwin and Barneby (Leguminosae- Caesalpinioideae). *Botanical Journal of the Linnean Society* 153: 169–179.
- Sahai K (1999). Structural diversity in the lens of the seeds of some *Cassia* L. (Caesalpinaceae) species and its taxonomic significance. *Phytomorphology* 49: 203–208.
- Sahai K (2001). Anatomical variability in seed coat of some *Cassia* L. (Caesalpinioideae) species with taxonomic significance. *Taiwania* 46: 158–66.
- Sahai K, Kaur H & Pal A (1997). Macro and micromorphological seed characteristics of some *Cassia* species and their taxonomic significance. *Phytomorphology* 43: 273–279.
- Saheed SA & Illoh HC (2010). A taxonomic study of some species in Cassiinae (Leguminosae) using leaf epidermal characters. *Notulae Botanicae Horti Agrobotici Cluj-Napoca* 38: 21–27.
- Sara FS, Dixit VK, Tripathi SC & Patnaik GK (1994). Antihepatotoxic activity of *Cassia occidentalis*. *International Journal of Pharmacognosy* 32: 178–183.
- Selvam V (2007). *Trees and Shrubs of the Maldives*. Bangkok, Thailand: Thammada Press Co., Ltd.
- Serrato-Valenti G, Cornara L, Ferrando M & Modenesi P (1993). Structural and histochemical features of *Stylosanthes scabra* (Leguminosae; Papilionoideae) seed coat as related to water entry. *Canadian Journal of Botany* 71: 834–840.
- Shaheen AM (2007). Characteristics of the stem-leaf transitional zone in some species of Caesalpinioideae (Leguminosae). *Turkish Journal of Botany* 31: 297–310.
- Shao S, Meyer CS, Ma F, Peterson CA & Bernards MA (2006). The outermost cuticle of soybean seeds: chemical composition and function during imbibitions. *Journal of Experimental Botany* 58: 1071–1082.
- Sharma N, Tripathi P, Athar M & Raisuddin S (2000). In vitro inhibition of carcinogen-induced mutagenicity by *Cassia occidentalis* and *Emblica officinalis*. *Drug and Chemical Toxicology* 23: 477–84.
- Souza FHD & Marcos-Filho J (2001). The seed coat as a modulator of seed-environment relationships in Fabaceae. *Revista Brasileira de Botanica* 24: 365–375.
- Srivastava M, Srivastava S, Khatoon S, Rawat AKS, Mehrotra S & Pushpangadan P (2006). Pharmacognostical evaluation of *Cassia angustifolia* seeds. *Pharmaceutical Biology* 44: 202–207.
- Stearn WT (1983). *Botanical Latin*, Third Edition. USA: David and Charles Inc.
- Subrahmanyam NS (1995). *Modern Plant Taxonomy*. Delhi, India: Vikas Publishing House PVT LTD.
- Tona L, Cimanga RK, Mesia K, Musuamba CT, De Bruyne T, Apers S, Hernans N, Miert SV, Pieters L, Totté J & Vlietinck AJ (2004). In vitro antiplasmodial activity of extracts and fractions from seven medicinal plants used in the Democratic Republic of Congo. *Journal of Ethnopharmacology* 93: 27–32.
- Tona L, Ngimbi MP, Tskala M, Mesia K, Cimanga K, Apers S, De Bruyne T, Pieters L, Tohe J & Vlietinck AJ (1999). Antimalarial activity of 20 crude extracts from nine African medicinal plants used in Kinshasa, Congo. *Journal of Ethnopharmacology* 68: 193–203.
- Vashishtha VM, John TJ & Kumar A (2009). Clinical and pathological features of acute toxicity due to *Cassia occidentalis* in vertebrates. *Indian Journal of Medical Research* 130: 23–30.
- Yadav JP, Arya V, Yadav S, Panghal M, Kumar S & Dhankhar S (2009). *Cassia occidentalis*: a review on its ethnobotany, phytochemical and pharmacological process. *Fitoterapia* 81: 223–230.