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Anatomical studies on Balā - An Ayurvedic drug and its varieties

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ABSTRACT
The root and petiolar anatomy of Balā, an important Ayurvedic drug and its varieties (Sida spp.) has been studied in detail to identify the original drug from the adulterants. The diagnostic key for distinguishing the different varieties on the basis of the anatomical features of these drugs is also presented in detail.

INTRODUCTION
Balā is a reputed drug in Ayurveda is used for the treatment of rheumatism and it forms a chief ingredient of several important Ayurvedic preparations like Ksirābala, Dhanvantaram, Balaristam, Rasadhti kasayan, Asvagandhadhi leham etc. Root is the official part of the drug and is reported to be cool, sweet, demulcent, aphrodisiac and tonic. It produces strength, imparts beauty to the body and cures vataartha, raktapitta, consumption, polyuria and ulcers. The drug is also useful in neurological disorders like hemiplegia, facial paralysis and sciatica, general debility, headache, opthalmia, dysuria, leucorrhoea, tuberculosis, diabetes, fever and uterine disorders (1).

In Bhavaprakasa nighantu, 4 varieties namely balā, athabalā, nagabalā and mahabalā are mentioned. The other two varieties are rājabalā mentioned in Rajanighantu and bhānimabalā in Ousadhinighantu are not in vogue or in practice. Of the four varieties, balā is the most widely used one. This has been equated with Sida cordifolia Linn. of Malvaceae (2 - 7). Ayurvedic formulary of India (8) has also accepted this and is widely used for source of balā in northern parts of India. But the traditional physicians of Kerala have adopted Sida rhombifolia ssp. retusa for this drug (9 - 11).

The other species of Sida ie. Sida acuta, Sida rhombifolia ssp. rhombifolia and Sida cordata are also adulterated or substituted with this drug. These species are common weeds found in abundance in many gardens and wastelands. The roots of these plants may simulate each other in dried condition and this may lead to either unintentional or intentional adulteration.

The present study puts forth a set of anatomical parameters of roots and petioles of some Sida species, which can be employed to distinguish the original drug as mentioned in the classical Ayurvedic drugs from the other adulterants. This study throws light on the need to properly identify the plant species to achieve standardization of Ayurvedic preparations which uses Balā as a key ingredient.

MATERIALS AND METHODS
Materials for the present study were collected from different places. They are as follows:
1. Sida cordifolia, Sida acuta, Sida cordata were collected from Cholayil Medicinal Plants Conservation Park (CMPCP), Velagapuram, Thiruvallur district, near Chennai and authenticated by regional floras (12 - 16).
2. † Sida rhombifolia ssp. retusa was collected from Kerala.
4. †† Sida rhombifolia ssp. rhombifolia var. rhombifolia from Korattur, Chennai and the material was referred and identified with the help of Auroville herbarium collections (Personal communication - Dr. Walter), Pondicherry.

† Huang Hua Ren Shu (in Flora of China 12: 270-275, 2007) treated this species as Sida alnifolia var. alnifolia along with other 3 varieties under Sida alnifolia Linn.
†† Sida rhombifolia ssp. rhombifolia var. rhombifolia - This is very rare plant. Hence, this plant was not taken for root anatomy study with the objective of conserving the live plant. However the petiole anatomical study was carried out.

Fresh root and petiole samples of the above species were washed and fixed in FAA for 24 h and dehydrated, paraffin infiltrated and embedded in wax through customary techniques (17 - 18). Serial transsections were obtained at 10-12μm thickness with rotary microtome and the sections were stained with Toluidine blue (19). Photomicrographs were taken with Nikon E400 microscope unit using 4X objective lens.

Observation
Anatomical descriptions of the root
1. Sida cordifolia Linn.
Macroscopic characters
Roots 5-15cm long and 0.75 –1cm thickness with few lateral roots of smaller size; tap root branched at the tip; outer surface is buff to grey-yellow; odourless; taste slightly bitter.

Microscopic characters (Fig 2:1)
In transection of young root measuring about 4-5mm in diameter is roughly circular in outline with small fissures. Outermost zone consist of radial bands of rectangular, tangentially elongated, thin-walled cork cells about 4-5 rows. Secondary phloem composed of phloem fibres in wedge-shaped patches with thin walled parenchyma in
between. Phloem rays thin-walled tangentially elongated towards secondary cortex; a few rosette crystals of calcium oxalate found scattered in phloem parenchyma. The secondary xylem is very prominent forming dense solid cylinder occupying the major area of the root. Growth rings are fairly distinct and demarcated by broad zone. Vessels circular, wide, arranged in ring porous with pores solitary and in short radial multiples. It consists of prominent wide radial bands of rays, predominantly bi-seriate and fairly thick-walled fibres. Starch grains are abundant in xylem ray cells.


**Macroscopic characters**

Root consists about 5-7mm thick, slender, main tap root with small numerous rootlets, dark yellow-brown in colour. Surface is rough due to rootlets scar and peeled cork.

**Microscopic characters** (Fig. 2:2)

Cross section of root about 1-2mm in diameter shows circular in outline with prominent exfoliated cork layers. Cork consists of 4-7 rows of rectangular, tangentially elongated, thin-walled parenchymatous cells. Inner cortex consists of scattered phloem fibres in between the thin-walled parenchymatous. Secondary xylem consists of dense solid cylinder. Vessels predominantly solitary or less frequently short radial multiples, wide and polygonal shape in outline. Xylem fibres are thick-walled and wider lumen. Xylem rays are 1-2 seriate and starch grains absent.

3. *Sida rhombifolia* Linn. ssp. *rhombifolia*

**Macroscopic**

Root may occur as entire or cut pieces of varying lengths, 7-8mm in thickness, with wavy lateral roots comparatively thinner than main roots having numerous rootlets, brownish yellow, surface rough due to scars of small rootlets and lenticels; fracture, hard and splintery.

**Microscopic** (Fig. 2:4)

A thin root about 3-4mm shows circular in outline with exfoliated cork, consisting of 3-7 rows of narrow, rectangular, tangentially elongated, thin-walled parenchymatous cells. Secondary phloem composed of phloem fibres in wedge-shaped patches with thin walled parenchyma in between; phloem rays thin walled, tangentially elongated towards secondary cortex. A few rosette crystals of calcium oxalate found scattered in phloem parenchyma. Secondary xylem composed of vessels, fibres, parenchyma and rays. Vessels arranged in long radial multiples rows. Xylem rays 2-3 seriates, filled with starch grains.


**Macroscopic characters**

Root consists of a short stout taproot system attaining a diameter of about 1cm and often long and flexuous with fairly thick lateral roots. The roots have a pale yellow colour; outer surface is fairly smooth except for the small filmy strips of exfoliating cork. The bark has thickness of about 1 to 11/2 mm and can be easily peeled off in fresh condition or by wetting.

**Microscopic characters** (Fig. 2:5)

A young root of 2-3mm thick shows circular in outline with exfoliated cork cells. It consisting of 5-6 rows of rectangular or tangentially elongated, thin walled cells followed by 2 to 3 layers of cork cambium towards inner side. Secondary cortex inner to cork cambium consists of 2 to 3 rows of tangentially elongated cells. Inner to the cortex, phloem elements, composed with thin-walled parenchyma alternate with phloem fibres. A few rosette crystals of calcium oxalate found scattered in phloem parenchyma. Secondary xylem is in dense solid cylinder occupying major part. Vessels are diffuse porous with pores solitary, wider, circular and/or in short radial multiples with abundant distribution. Xylem rays mostly bi-seriate with abundant starch.

5. *Sida cordata* (Burm.f.) Borss. (Syn. - *Sida veronicifolia* Lam.)

**Macroscopic characters**

Tap-root is very long, slender about 4-6mm in diameter having many small rootlets, yellowish brown in colour, outer surface is smooth.

**Microscopic characters** (Fig. 2:3)

About 3-4mm thick root shows circular outline with less frequent exfoliation of cork layers. Cork consisting about 4-5 rows of tangentially elongated, rectangular cells. The outermost cortex consist fairly distinct parenchymatous cells, which are loosely arranged with intercellular spaces. The inner cortex bounded internally by patches of phloem fibres in between the parenchymatous cells. Phloem rays broadly enter towards the cork. A few rosette crystals of calcium oxalate found scattered in phloem parenchyma. Secondary xylem is very prominent, dense solid cylinder. Vessels circular, wider and diffuse porous with pores solitary and in short radial multiples. Xylem rays prominent, 2-3 seriate with absence of starch grains.

**Anatomical description of the Petiole**

The petiolar anatomy has been studied based on the topographic arrangement of the vascular bundles.

1. *Sida cordifolia* Linn. (Fig. 3:1)

In transverse section of petiole measuring about 1mm diameter shows triangular shape in outline. A small groove is present on the adaxial side. Epidermis is single layer of compactly arranged barrel shaped cell and covered with abundant stellate trichomes. Hypodermis is represented by 3-4 layers of collenchymatous tissue occur just below the epidermis. The ground tissue made up of parenchymatous cells with distinct intercellular spaces. A few rosette type of calcium oxalate crystals found scattered in the ground tissue. Three vascular bundles present in the ground tissue and all are similar in size and shape and vascular system open type. They arranged as one dorsal (lower) and two laterals. Thus, the arrangement is expressed as 1 + 2.

2. *Sida rhombifolia* Linn. ssp. *retusa* (Linn.) Borss. (Fig. 3:2)

Petiole about 0.7mm in diameter and consist single layer of epidermis with compactly arranged cubical cells and covered by abundant stellate trichomes. 2-3 layers of collenchymatous cell present just below the epidermis. Five vascular bundles present in the ground tissue and in different size and vascular system open type. They arranged as one dorsal (lower) and two ventrals and these are small in size compare to others. Thus, the arrangement is expressed as 1 + 2 + 2.

3. *Sida rhombifolia* Linn. ssp. *rhombifolia* (Fig. 3:3)
Petiole measuring about 1mm in diameter and shows single layer of epidermis with barrel shaped cells covered by stellate trichomes and less distribution. 1-2 layers of collenchymatous cells present just below the epidermis. A few rosette type of calcium oxalate crystals found scattered in the ground tissue. Four vascular bundles arranged as one dorsal, two laterals and one ventral. But these bundles are fused by thick-walled sclerenchymatous cells and form closed type of vascular system. Middle region of the vascular bundle made up of thin-walled parenchymatous cells. The arrangement of vascular bundles expressed as 1 + 2 + 1.

4. *Sida rhombifolia* Linn. *ssp. rhombifolia var. rhombifolia* (Fig. 3:4)

Petiole about 0.7mm in diameter and consist single layer of epidermis with compactly arranged cubical cells and covered by stellate trichomes and less distribution. 3-4 layers of collenchymatous cells present just below the epidermis. A few rosette type of calcium oxalate crystals found scattered in the ground tissue. Four vascular bundles arranged as one dorsal, two laterals and one ventral and open system. Dorsal and lateral bundles fused by sclerenchymatous cells and form girdle shape. The arrangement of vascular bundles expressed as 1 + 2 + 1.

5. *Sida acuta* Burm.f. (Fig. 3:5)

Cross section of petiole measuring about 0.5mm diameter shows wavy margin in outline with stellate trichomes. Hypodermis and ground tissue form few layers only. Four vascular bundles arranged as one dorsal, two laterals and one ventral and these bundles fused by thick-walled sclerenchymatous cells and form closed type as in *Sida rhombifolia ssp. rhombifolia*. Middle region of the vascular bundle made up of thick-walled sclerenchymatous cells. The arrangement of vascular bundles expressed as 1 + 2 + 1.

6. *Sida cordata* (Burm.f.) Borss. (Fig. 3:6)

In transverse section of petiole measuring about 0.8mm diameter shows triangular shape in outline. A small groove is present on the adaxial side. A few simple, unicellular elongate trichomes present in the adaxial groove. Well-developed hypodermis and ground tissues present. Vascular system open type and four vascular bundles are arranged as one dorsal, two laterals and one ventral and open. The arrangement of vascular bundles expressed as 1 + 2 + 1.

**DISCUSSION**

Balā is one of the most important drugs used in the various formulations of Ayurveda and other Indigenous medicines. Many species of *Sida* are used as balā and/or its varieties. In North India, *Sida cordifolia* is used as balā. But in Kerala, *Sida rhombifolia ssp. retusa* have adopted. The other species of *Sida* may adulterate or substitute with the original balā. It is easy to distinguish these species when they bear flowers and fruits. However, the crude drug in the market is very difficult to identify (the original from the adulterant) due to lack of flowers and fruits. The anatomical features of root and petiole of these species are specific and these can be employed for the identification of drug of choice (original drug).

**The following anatomical features are suggested to diagnose the root samples**

1. *Sida cordifolia* Linn.

Growth rings are fairly distinct, vessels circular in outline, wider, arranged in ring porous with pores solitary and in short radial multiples. Xylem rays predominantly bi-seriate with abundant starch grains.


Vessels polygonal in outline, wider, predominantly in solitary or very less frequent short radial multiples. Xylem fibres are thick-walled and wider lumen. Xylem rays are 1-2 seriate and starch grains absent.

3. *Sida rhombifolia* Linn. *ssp. rhombifolia*

Vessels circular in outline, moderately wide, arranged in long radial multiples. Xylem rays 2-3 seriates, filled with less starch grains.


Vessels circular in outline, wider, arranged in diffuse porous with pores solitary, in short radial multiples with abundant distribution. Xylem rays mostly bi-seriate with abundant starch.

5. *Sida cordata* (Burm.f.) Borss.

Vessels circular in outline, wider, arranged in diffuse porous with pores solitary and in short radial multiples with less abundant compared to *Sida acuta*. Xylem rays prominent, 2-3 seriate with absence of starch grains.

**The following petiolar anatomical features are suggested to diagnose the Sida spp.**

1. *Sida cordifolia* Linn.

Triangular shape in outline. Vascular system open type and three vascular bundles similar in size and shape and arranged as 1 + 2.

2. *Sida rhombifolia Linn. ssp. retusa* (Linn.) Borss.

Vascular system open type and five vascular bundles different in size and arranged as one dorsal, two laterals and two ventrals and these are small in size compare to others and arrangement expressed as 1 + 2 + 2.

3. *Sida rhombifolia Linn. ssp. rhombifolia*

Vascular system closed type, four bundles arranged as one dorsal, two laterals and one ventral, these bundles fused by thick-walled sclerenchymatous cells. Middle region of the vascular bundle made up of thin-walled parenchymatous cells and arrangement expressed as 1 + 2 + 1.

4. *Sida rhombifolia Linn. ssp. rhombifolia var. rhombifolia*

Vascular system open type, four vascular bundles arranged as one dorsal, two laterals and one ventral. Dorsal and lateral bundles fused by sclerenchymatous cells and form girdle shape and arrangement expressed as 1 + 2 + 1.

5. *Sida acuta Burm.f.*

Vascular system closed type, four vascular bundles arranged as one dorsal, two laterals and one ventral and these bundles fused by thick-walled sclerenchymatous cells and middle region of the vascular bundle made up of thick-walled sclerenchymatous cells and arrangement expressed as 1 + 2 + 1.


Vascular system open type, four vascular bundles arranged as one dorsal, two laterals and one ventral and the arrangement expressed as 1 + 2 + 1.

**A key for identification based on root anatomical features of five Sida species.**

1a. Growth rings present. Vessels ring porous.
Fig. 1 - Morphology of Sida spp.
Fig. 2 - Root Anatomy of *Sida spp.*

*Sida cordifolia*  
*Sida rhombifolia ssp. retusa*

*Sida cordata*

*Sida rhombifolia ssp. rhombifolia*  
*Sida acuta*
Fig. 3 - Petiolar Anatomy of *Sida* spp.

*Sida cordifolia*
1b. Growth rings absent. Vessels diffuse porous
2a. Vessels polygonal in outline and predominantly solitary. *Sida rhombifolia* ssp. retusa
2b. Vessels circular in outline and not as above
3a. Vessels arranged in long radial multiples
*Sida rhombifolia* ssp. rhombifolia
3b. Vessels not as above
4a. Starch grains absent in xylem rays, Vessels less abundant *Sida cordata*
4b. Starch grains present in xylem rays, Vessels abundant.

*Sida acuta*
2. A key identification based on petiolar anatomical features of six *Sida* species.
1a. Vascular system open type
2a. Dorsal and lateral vascular bundles fused
*Sida rhombifolia* ssp. rhombifolia var. rhombifolia
2b. Vascular bundles not as above
3a. Vascular bundles 5 nos
*Sida rhombifolia* ssp. retusa
3b. Vascular bundles not as above
4a. Vascular bundles 4 nos

*Sida cordata*

Sida cordata
4b. Vascular bundles 3 nos

Sida cordifolia
1b. Vascular system closed type
5a. Middle region of the vascular bundle made up of thin-walled parenchymatous cells

Sida rhombifolia
5b. Middle region of the vascular bundle made up of thick-walled sclerenchymatous cells

Sida acuta

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REFERENCES

5. A.C. Dey, Indian Medicinal Plants, Dehra Dun; 21 - 22 (1980).

LEGEND