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TRICHOMES AS BIOMARKERS FOR THE IDENTIFICATION OF A FEW MEDICINAL PLANTS OF THE FAMILY MALVACEAE

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ABSTRACT

In the present work 19 medicinal plants belonging to the family Malvaceae have been subjected to an intensive study on their micromorphological characters such as stellate trichomes and glands present in leaves and other vegetative parts to find out their utility as biomarkers in raw materials. The hairs offered a variety of structures such as unicellular, bicellular, typical stellate hairs with 3 to 9 arms and flat discs. Substantial variations were found in number of arms, thickness of walls and lumen, base of stellate hair, its pitting and the cells of epidermis surrounding the hairs. The glands also varied from unicellular to multicellular flask shaped structures. Based on these characters a key to identify all the 19 plants were prepared. The utility of these characters as biomarkers in pharmacognosy and in taxonomy is discussed.

Keywords: Micromorphological characters, Trichomes, Stellate hairs, Glands, Medicinals, Malvaceae. Bala.

INTRODUCTION

Micromorphological characters including the trichomes and glands of plants have assumed great taxonomical significance recently as viable taxonomic markers. These characters which would be of help in identifying plants even in vegetative state will be of great use for field taxonomists in identifying plants in the absence of flowers and fruits which are available only in certain seasons of the year. It was Solereder [1] and Metcalfe and Chalk [2] who made some significant contributions to the micromorphological characters of plants. Plant morphologists had used many micromorphological characteristics as foliar trichomes to resolve the taxonomic conflicts and thus these characters have played an important role in plant taxonomy [3, 4]. In case of medicinal plants, trichome characters act as biomarkers to identify the plant even in the raw material or powder form [5, 6].

The family Malvaceae is a rich storehouse of medicinal plants. They are the following.

1. **Bala.** This is one of the very important rasayana drugs, held in great repute by Ayurvedic physicians for the treatment of rheumatism and neurological disorders like hemiplegia, facial paralysis and sciatica, general debility and uterine disorders. It is demulcent, aphrodisiac and produces strength and beauty to body. Different plants belonging to Malvaceae are useds as bala. Bala is of different types such as Common Bala (*S. cordifolia* Linn.), Nagbala (*Sida acuta* Burm.F., *S. spinosa* Linn. and *S. veronicifolia* Lam.), Atibala (*Urena lobata Linn., U. sinuata* Linn. *Abutilon indicum* Linn. and *A. hirtum* Sweet.) and Mahabala (S. *rhombifolia* var *rhomboidea* Mast) [7].

2.**Japa** *(Hibiscus rosa-sinensis* Linn.). Leaves and flowers of this plant are used for promoting the growth and color of hair and in treating ulcers.

3. Red sorrel (*H. sabdariffa* Linn.). The tender leaves and fleshy calyces of flowers are used to prepare cooling and refreshing drinks. Infusions of the leaves or calyces (Rosella/ Hibiscus tea) are regarded as diuretic, cholerectic, febrifugal and hypotensive, decreasing the viscosity of the blood and stimulating intestinal peristalsis. Pharmacognosists in Senegal recommend red sorrel extract for lowering blood pressure and in later scientists confirmed the hypotensive activity of the calyces and found them antispasmodic, anthelmintic and antibacterial as well.

4. Palash (*Thespesia populnea* Sol.)**,** is a reputed remedy for skin diseases. It is also useful in dysentery, piles, haemorrhoids and urinary infections. In addition a number of related plants also are used in traditional medicinal practices like Ayurveda and in folklore. They are the following.

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5. Kakrai (*Abutilon glaucum* Sweet). The leaves of this plant are used for piles and the roots in place of *Abutilon indicum.*

6. Jungli-Bala. (*Sida rhombifolia* var.*retusa* Borss). This plant is used like Bala for rheumatism and neurological complaints like epilepsy. It is also used in diarrhoea and as a diuretic and antipyretic.

7. Jungli Bhindi (*Hibiscus solandra* L'Her, Syn.*Hibiscus lobatus* O. Kuntze). Paste of fresh plant is applied on breast to cure mastitis [8]. Whole plant is used in debility and spermatorhoea in Madhya Pradesh [9].

8. Sticky Fanpetals (*Sida glutinosa* Cav.). The whole plant is neuroprotective and antioxidant in nature [10].

9. Yellow sticky melon (*Pavonia zeylanica* Cav.) roots and leaves are used against inflammation, hemorrhage and dysentery and as an emollient.

10. Sleepy Hibiscus/Wax Mallow (*Malvaviscus arboreus* Cav.)Traditionally the fruit and flowers have been used to treat diarrhea. The flowers and fruit make a good herbal tea.

In the present work, all the 19 medicinal species mentioned above, belonging to the family Malvaceae, have been studied for their micromorphological characters such as trichomes and glands in their leaves with a view to identify the pharmacognostic biomarkers which will enable to identify the drug plant as they are or in powder form.

MATERIALS AND METHODS

All the plants were collected from in and around Vadodara, Gujarat, India. For studying the trichomes, epidermal peels were taken out manually with the help of blade and stained with safranin [11]. Stained peel was mounted in glycerin and then observed under the compound microscope. Sections of leaves were taken and stained with safranin and mounted in glycerin. The measurements on the cells and other ingredients were done using stage and ocular micrometers and Camera-lucida diagrams prepared.

RESULTS

All the plants screened contained different types of trichomes. The length, breadth and other characters of the trichomes of 19 medicinal species of Malvaceae are presented in Table 1 and Figs 1-3. Fifteen plants possessed typical stellate trichomes having 3 or more arms radiating from the centre. Typical unicellular tapering trichome was characteristic to *Sida acuta* and *Pavonia zeylanica,* while one or two-armed trichomes were seen in *Malvaviscus arboreus* and *Urena sinuata. Thespesia populnea* had a unique stellate trichome which was multicellular flat and disc shaped. In all the types of stellate hairs there were many characteristic variations highly useful in identifying the species in question. In addition many of the plants contained various types of glands also. The characteristics of the trichomes and glands of all the plants screened are explained below.

1. *Abutilon indicum* Linn. Here the stellate hairs were short and 2-3 armed though 8-9-armed stellate hairs were also seen. Many a times the stellate hairs are attached at the base to a large parenchymatous cell containing a sphaeraphide. The walls of arms were thick and lumen inside was very narrow. In addition 2-celled glands (with a round swollen glandular apical cell and a colourless basal sell) also are seen (Fig.1).

2. *Abutilon hirtum* Sweet. Here the stellate hairs were 2-3 armed though 7-9 armed hairs also are seen. The walls of arms were thin and lumen inside was broad. There were no glands on the leaf surface (Fig.1).

3. *Abutilon glaucum* Sweet. Both small and big stellate hairs were seen here. Hairs were mostly 2-3 armed. But the arms had a constricted base. Often in T.S. of leaf, the base of stellate hairs were found attached to a large parenchymatous cell containing a sphaeraphide. Glands had a 1-celled stalk and 3-4 celled head (Fig.1).

4. *Hibiscus rosa-sinensis* Linn. Here the trichomes were small and had arms of varying length in that some arms were very short while others were longer. The arms were having very thick walls and very thin lumen. Unicellular glands also were present (Fig.1).

5. *H. sabdariffa* Linn. Here the stellate hairs were two-armed. The arms were very long with thin walls and large lumen containing some viscous inclusions. Glands having a unicellular colorless stalk and multicellular (4-5 celled) head also were present (Fig.1).

6. *H. solandra* L'Her. The arms of stellate hairs here are joined at two levels. The hairs having 6 arms were having 3 arms joined at the base wherein the angles between two arms were 120^{o} to form a tier below and the other three arms joined similarly to form a tier below . Similarly hairs having 8 arms were with 4 arms joined in the form of a cross in a tier and other 4 arms forming a similar tier below. The epidermal cells surrounding the base of stellate hairs also were thick and the walls between the base of hair and surrounding cells were pitted. The walls of hairs were longitudinally stratified (Fig. 2).

7. *H. shizopetalous* Hook.F. Here the stellate hairs similar to that of *Hibiscus rosa-sinesis* in general appearance, but were 5-7 armed, having a broad lumen and firmly fixed in a ring of thickened epidermal cells (as in *H. solandra*) to which there were pit connections (Fig.1).

8. *Malvaviscus arboreus* Cav. This plant possessed very long unicellular (1050 µm) trichome as well as 2-armed stellate hairs in which each arm resembling a unicellular

trichome. The base of hairs was pitted (Fig.2).

9. *Pavonia zeylanica* Cav. Here the trichomes were unicellular and long (560 µm) with a broad base, in that they appeared conical. In addition there were two types of glands were seen; one having a multicellular globose head and a multicellular uniseriate 3-4 celled stalk and another which is uniseriate and flask shaped (Fig.2) .

10. *Sida cordifolia* Linn. Here the stellate hairs were mostly 2 to 3 armed though 7-armed hairs also are seen. They arose from a ring of thickened epidermal cells and were comparatively long. The lumen was very narrow (walls very thick) but was swollen at the base enclosing an oval lumen. The arms of 2-celled trichomes were curved about each other giving a horn-shaped appearance. In addition, flask shaped glands with a broad apical cell also were seen (Fig.2).

11. *S. acuta* Burm.F. This plant possessed typical unicellular pointed trichome resembling an arm of stellate hair. The walls were moderately thin and stratified. Very few 4-5 armed stellate hairs also are seen (Fig.2).

12. *S. glutinosa* Cav. Here the trichomes were of mixed type and were; 1. unicellular acicular or curved at the tip, 2. 4-armed cross-shaped stellate hairs with long arms and 3. 5-7 armed hairs. The lumen was very narrow (Fig.3).

13. *S. rhombifolia* var.*retusa* Borss. Here the majority of stellate hairs were having 2 or three long arms. Very small stellate hairs with 5 arms also were seen. Here the stem also was found having abundant stellate hairs with 4-5 transversely septate arms (Fig.3).

14. S. *rhombifolia* var *rhomboidea* Mast. This plant contained typical 5-armed stellate hairs. But they were larger than the former plant and the hairs had a broad conical flask-shape base (Fig.3).

15. *S. spinosa* Linn. The stellate hairs in this plant were more similar to that of *S. rhombifolia* var.*retusa* in that they were 2 or 3 armed. But here the leaves contained multicellular flask-shaped as well as 2-celled glands (Fig.2).

16. *S. veronicifolia* Lam. In this plant there were long unicellular acicular trichomes and small 5-armed stellate hairs. In both the types, the bases of trichomes were pitted with the surrounding thin or thick walled epidermal cells (Fig.3).

17. *Urena lobata* Linn. The stellate hairs here were, large, 5-armed and fitted in a ring of thick walled epidermal cells. The cross walls between the base of arm and the cells surrounding were pitted. There were also glands having unicellular stalks and 1-celled globose heads (Fig.3).

18. *U. sinuata* Linn. had one or two armed stellate trichomes. There were no basal ring of thick walled cells nor pits at the base of arms. The glands had a 3-5 celled head and one celled stalk (Fig.3).

19. *Thespesia populnea* Sol. possessed a unique multi-armed (up to 30 arms) stellate hair in which the arms were joined by the margins to form a disc shaped structure occurring closely appressed to leaf surface. The lengths of arms were unequal. In addition there were unicellular thick-walled acicular trichomes (Fig.3).

Key 1. The key, for identification of all the nineteen medicinal species of the Malvaceae studied, is given below:

1. Flat disc-shaped multicellular stellate hairs- cells fused throughout, appressed to the surfaces of leaves- *Thespesia populnea*
1. Hairs more or less erect or slanting, arms free except for the base.
2. Hairs predominantly unicellular.
3. Only unicellular trichomes present- *Pavonia zeylanica*
3. Unicellular trichomes along with hairs with more arms
4. Unicellular trichomes along with 2-armed hairs
5. Arms of trichomes more than 1050 µm long, base pitted - *Malvaviscus arboreus*
5. Arms of trichomes shorter, less than 400 µm long, Base not pitted – *Urena sinuata*
4. Unicellular trichomes along with 4, 5 or more arms
6. 4-Armed cruciform hairs and unicellular hairs curved at tip - *Sida glutinosa*
6. 4-Armed cruciform hairs absent and unicellular hairs straight.
7. Arms less than 400 µm long, base of arms pitted with the surrounding thick walled cells epidermis– *Sida veronicifolia*
7. Arms more than 500 µm long, no ring of thick walled cells At the base - *Sida acuta.*
2. Hairs 2 or more armed (Unicellular hairs absent).
8. 2- Armed hairs predominant
9. Two armed hairs only - *Hibiscus sabdarifa*
9. Two armed hairs along with hairs having more arms.
10. Trichomes arise from a ring of thick walled epidermal cells to which pit connections are seen, lumen very narrow - *Sida cordifolia*
10. No ring of cells at base, lumen broad
11. Glands present

12. Multicellular flask-shaped as well as 2-celled glandsPresent along with stellate hairs- *Sida spinosa.*
12. Flask shaped gland absent. Smaller glands present.
13. Glands 2-celled, Arms of stellate hairs reach up to 8-9- *Abutilon indicum*
13. Glands 1-celled stalk and 3-4 celled head, Arms of stellate hairs mostly 2-3 - *Abutilon glaucum.*
11. Glands absent.
14. Lumen narrow, base of stellate hair not constricted - *S. rhombifolia var.retusa*
14. Lumen broad, base of stellate hair constricted - *Abutilon hirtum*
8. Hairs 3 or many armed (no 2-armed hairs)
15. - Armed hairs
16. Hairs 3, 6 and 8-armed- *H. solandra*
16. Hairs 3 or 4- armed, lumen very narrow - *Hibiscus rosa- sinensis*
15. Hairs 5 or more armed (never 1,2or 3-armed)
17. Arms of stellate hair unequal, Lumen broad, base of arms pitted - *Hibiscus schizopetalous*
17. Arms of stellate hair equal
18. Trichomes arise from a ring of thick walled epidermal cells, base of arms pitted - *Urena lobata*
18. Base of trichomes surrounded by normal epidermal cells, no ring of thick walled epidermal cells.
- *S. rhombifolia* var *rhomboidea*

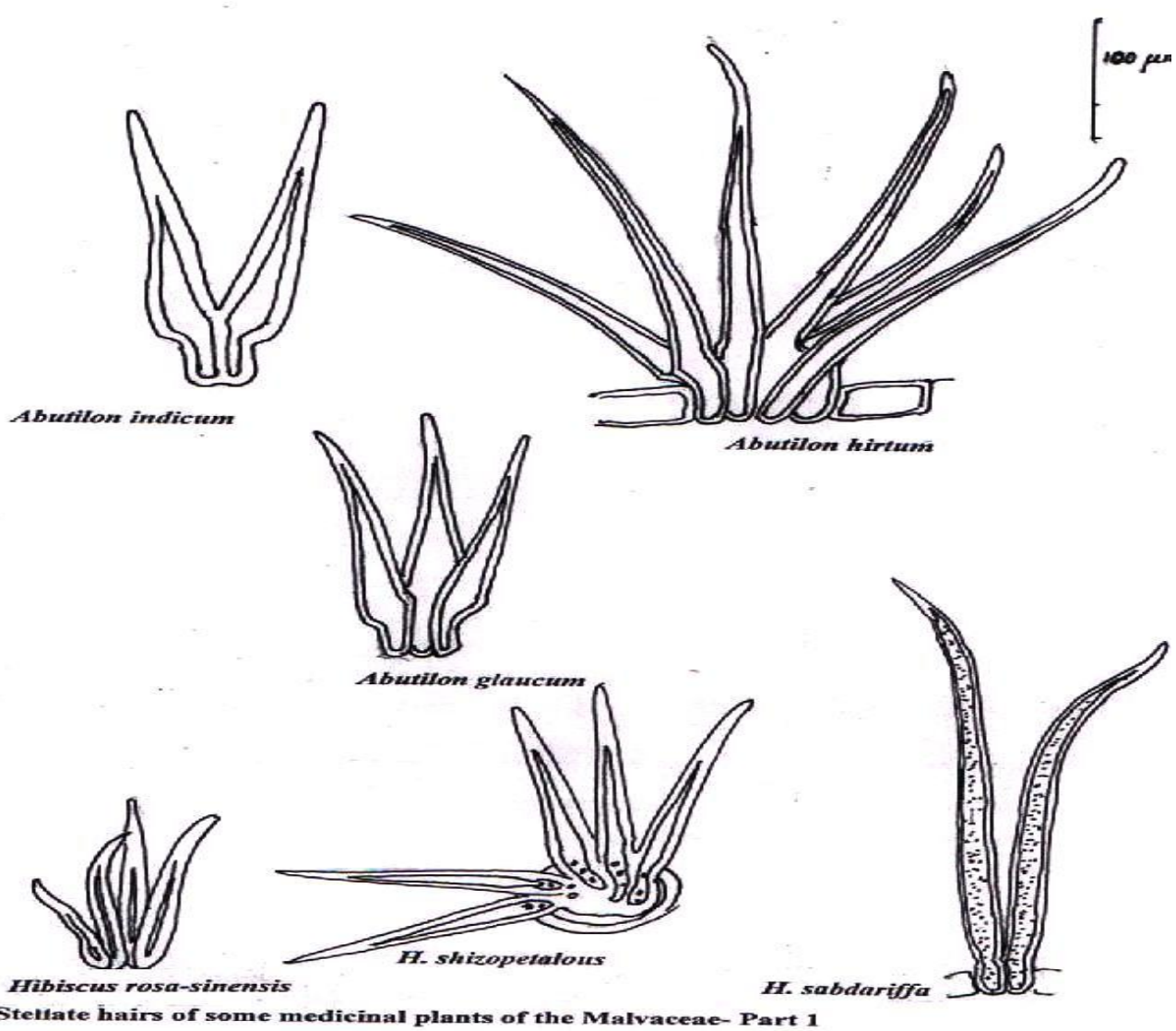

Fig.1.Stellate hairs of some medicinal plants of the Malvaceae- Part 1

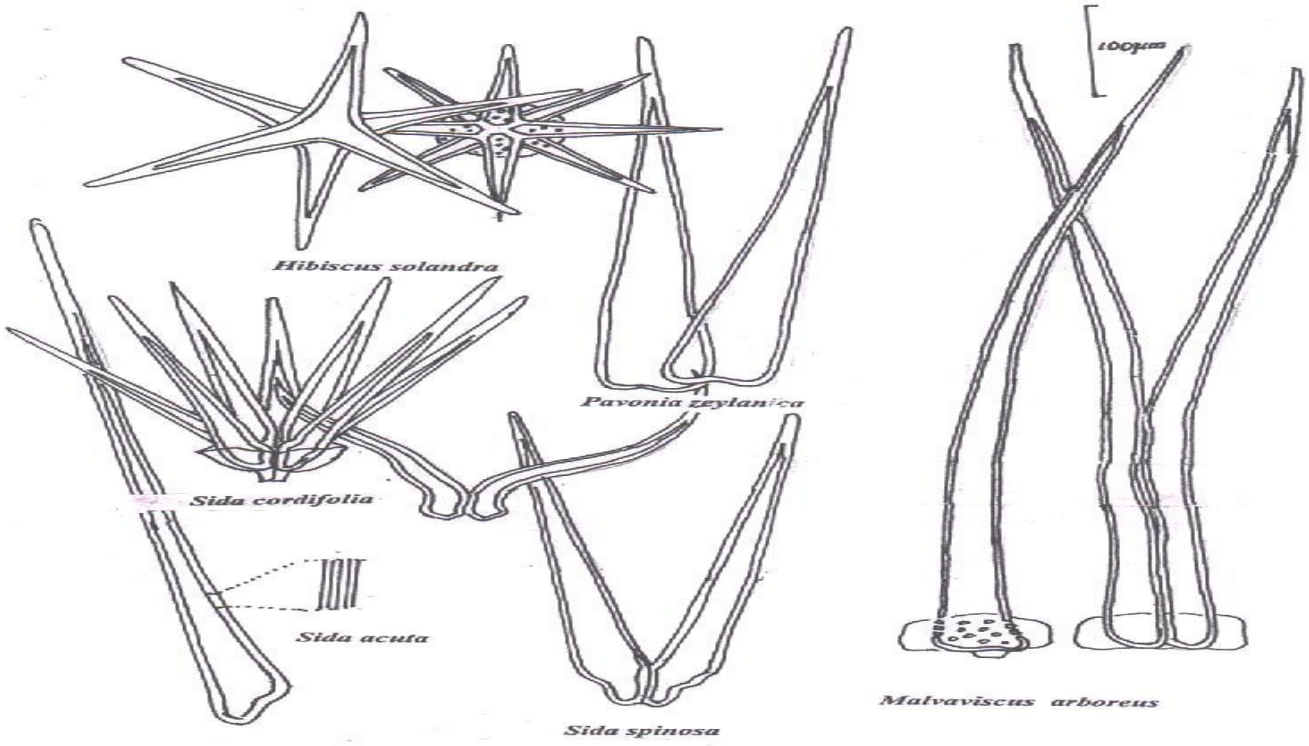

Fig.2.Stellate hairs of some medicinal plants of the Malvaceae- Part 2

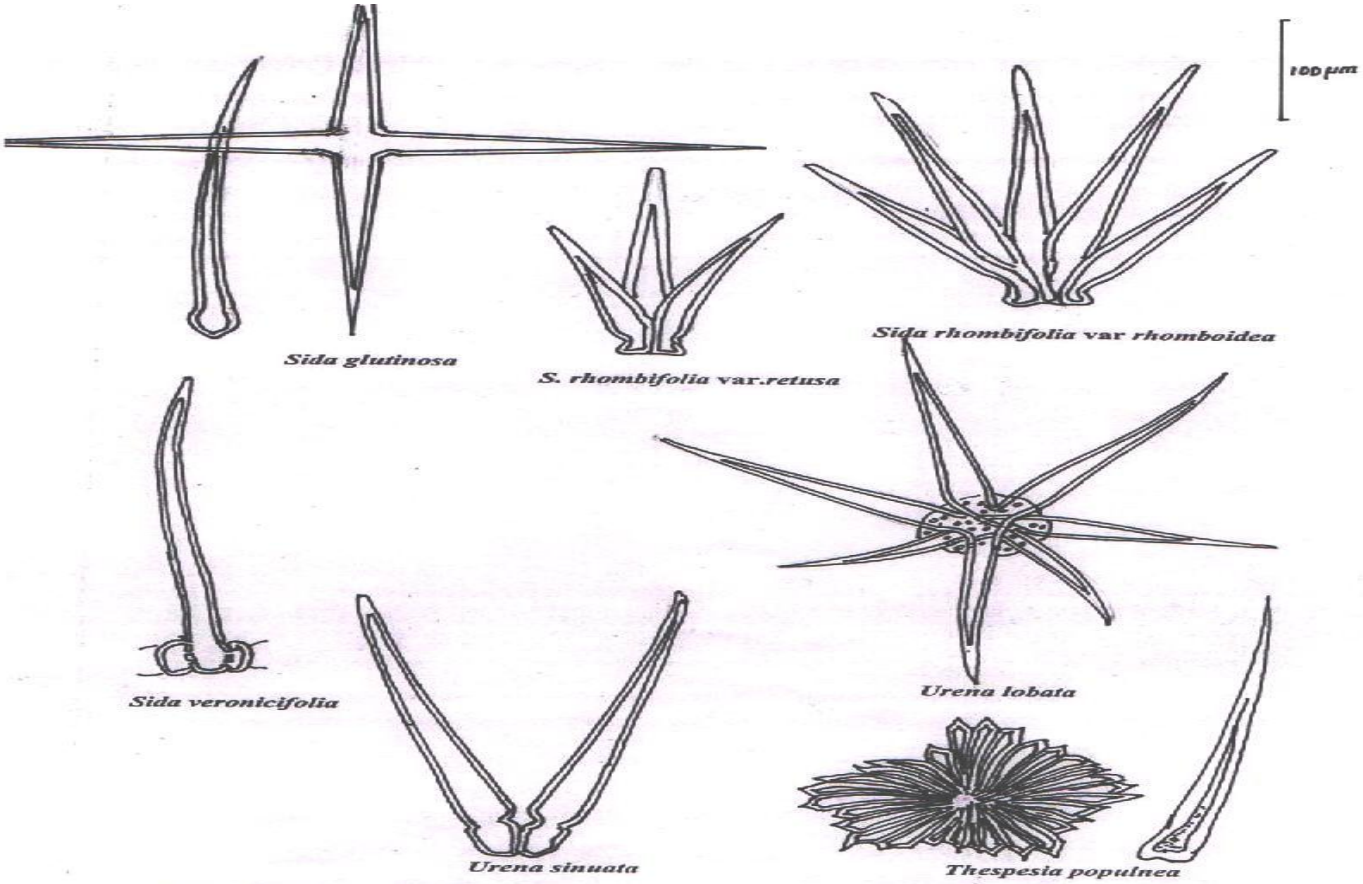

Fig.3.Stellate hairs of some medicinal plants of the Malvaceae- Part 3

Table 1. The nature, length, breadth and other characters of the trichomes of 19 medicinal plants of the Malvaceae

Sr.No.	Name of the species	Type	Length	Breadth
1	*Abutilon indicum* Linn.	2,3- 9 Armed	230 µm	14 µm
2	*Abutilon hirtum* Sweet	2,3,9 Armed	285 µm	14 µm
3	*Abutilon glaucum* Sweet.	2-3 Armed.	200 µm	13 µm
4	*Hibiscus rosa-sinensis* Linn.	3or 4 Armed	105 µm	15 µm
5	*H. sabdariffa* Linn.	2-Armed	360 µm	14 µm
6	*H. solandra* L'Her.	3/6 or 4/8 armed	240 µm	15 µm
7	*H. shizopetalous* Hook.F.	5-7 Armed	225 µm	21 µm
8	*Malvaviscus-arboreus* Cav.	1or 2-Armed	1050 µm	21 µm
9	*Pavonia zeylanica* Cav.	Unicellular tapering	560 µm	50 µm at base
10	*Sida cordifolia* Linn.	2/3 or 7-Armed	375 µm	14 µm
11	*S. acuta* Burm.F.	1 or 5-armed	675 µm	30 µm
12	*S. glutinosa* Cav.	1, 4 or 5-Armed hairs	465 µm	21 µm
13	*S. rhombifolia* var.*retusa* Borss.	2-3 Armed	300 µm	15 µm
14	S. *rhombifolia* var *rhomboidea* Mast.	5-Armed	450 µm	20 µm
15	*S. spinosa* Linn.	2 or 3 Armed.	340 µm	15 µm
16	*S. veronicifolia* Lam.	1 or 5-armed	300 µm	17.5 µm
17	*Urena lobata* Linn.	5-Armed	375 µm	20 µm
18	*U. sinuata* Linn.	1 or 2 armed	275 µm	15 µm
19	*Thespesia populnea* Sol.	Unicellular or many armed, disc shaped	120 µm	19.5 µm

DISCUSSION

From the above mentioned results it is absolutely clear that the trichomes from each plant are specific in their characters such as number of arms, thickness of walls and lumen, base of stellate hair, its pitting and the cells of epidermis surrounding the hairs. Glands also are diverse nature emphasizing their utility as biomarkers. The usefulness of these characters in identifying the medicinal species of the family Malvaceae is amply evident by the key (Key 1) prepared based on thm. This key can be used in confirming the identity of these species in a drug material or in a herbarium.

In addition, this study indicates the utility of the trichome character in identifying different medicinal herbs of the family Malvaceae which contains a number of important herbal drugs like various types of Bala, red sorrel, shoe flower, Palash etc. These characters can also aid in identifying the stem or the leaves in any useful part of a plant used as a drug or raw material and thus can serve as biomarkers for the plant in question. Even the very closely related species, as also of controversial taxonomic status [12], like *Urena lobata*, *Urena sinuata*, *Hibiscus rosa-sinensis* and *H. shizopetalous*, also can be distinguished by their trichome characters and this facility is not available with any other biomarker whether it is chemical or biochemical. This study gives additional data on the separate taxonomic status of *Urena lobata* from *Urena sinuata* and *Hibiscus shizopetalous* from *H. rosa-sinensis.* Micromorphological characters, thus, should be a welcome addition to any taxonomic description.

CONCLUSION

A glance at the characters exhibited by the different trichomes indicates the utility of the trichome character in identifying the various drug plants belonging to the family Malvaceae. These characters are present in the vegetative state and therefore, are available throughout the year for any student of pharmacognosy. These characters can also aid in identifying the stem or the leaves in any useful parts used as a drug or raw material and thus can serve as viable biomarkers. These characters also aid in resolving the taxonomic disputes between closely allied species as evidenced in the case of controversial taxa of the genera *Urena* and *Hibiscus.* Such facility is not available with any other biomarker whether it is chemical or biochemical. The micromorphological characters, thus, should be added to the taxonomic description of a plant species.

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