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TRADITIONAL AND AYURVEDIC MEDICINAL IMPORTANCE OF AGASTHYA LEAVES [*SESBANIA GRANDIFLORA* (L) PERS.] W.R.T. ITS PHARMACOGNOSTIC AND PHYSICOCHEMICAL EVALUATION

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ABSTRACT
Agasthya [*Sesbania grandiflora* (L) Pers.] a traditionally revered Ayurvedic medicinal plant especially in Southern India. It goes as a principal ingredient in preparation of Ayurvedic medicinal formulations. Classical texts of Ayurveda have attributed wide ranging therapeutic indications to the herb. Agasthya leaves are known to possess anthehelmintic, alixeteric properties aperient, tonic, diuretic, and laxative properties. Further, they have been documented as therapeutically useful in Kaphaja disorders, pruritis, skin disorders, night blindness, epilepsy, gout, ophthalmia nasal catarrh and headache. The leaves contain a non-poisonous saponin. The present study is an attempt to decipher the rationality of the traditional use of leaves with an Ayurvedic perspective vis-à-vis pharmacognostical and physicochemical evaluation of the plant.
Keywords: Agasthya, Ayurveda**,** pharmacognostical, physicochemical evaluation.

INTRODUCTION

In Ayurvedic system, whole plant of Agasthya is medicinally valued. It goes as a principal ingredient in preparation of Ayurvedic medicinal formulations. Classical texts of Ayurveda have attributed wide ranging therapeutic indications to the herb. Agasthya leaves contain a non-poisonous saponin and are known to possess anthehelmintic, alixeteric, aperient, tonic, diuretic, and laxative properties. Further, they have been documented as therapeutically useful in Kaphaja disorders, pruritis, skin disorders, night blindness, epilepsy, gout, ophthalmia nasal catarrh and headache. Agasthya is traditionally revered plant in India. Traditional Indian practices have a close relation to general health behavior. As a matter of one such traditional practice, South Indians fast proportional to their age on the 11th day of the lunar cycle (known as 'ekadashi') and on the next day they break the fast with a curry prepared with Agasthya leaves, red gram, cococut gratings and chutney (ground paste) of Amla (Indian Gooseberry).This is believed to have restorative and rejuvenative effect. Further leaves and flowers of Agasthya are also considered sacred and offered to the God, in rituals especially during the auspicious day of Uttana dwadasi [12th day of Lunar cycle in Kartika masa (Oct-Nov)]. Therefore, with rich traditional and Ayurvedic medicinal importance of the leaf, an attempt has been made to conduct pharmacognostic and physicochemical evaluation vis-à-vis traditional and medicinal uses.

Brief Description of Agasthya

Agasthya is botanically equated to *Sesbania grandiflora* (L) Pers. belonging to the Family, Fabaceae. It is a species of tropical climate, short lived, quick growing and soft wooded tree. It grows up to 6-9 meters high and is cultivated in various parts of South India. Flowers are fleshy with large showy white, pink or crimson petals. They are 7.5 to 10cms long with short axillary racemes (Figure 1). Leaves are long, abruptly pinnate; leaflets 41-61 linear, oblong, and glabrous. It is known as Swamp pea in English, Agathi and Agasthya in Sanskrit, Agase in Kannada. Tender leaves, flowers and pods are used as vegetable and considered excellent source of vitamin C and calcium[1]. Seeds are rich in protein. The dried leaves are used as tea and is considered to have antibiotic, antihelminthic, antitumour and contraceptive properties. A poultice made from the leaf juice is effective in bruises[1,2]. Whole plant is used in Ayurvedic formulations like Grahani kapata rasa, Ratnagiri rasa and Pittakasantaka rasa etc[3]. The leaf is widely used in detoxification process of Ayurvedic metallo mineral drug, Manashila (AS_2S_2)[4]. Agathi leaves contains moisture-73.1gms, protein- 8.4 gms, fat-1.4 gms, minerals- 3.1 gms, crude fibers- 2.2gms, carbohydrates- 11.8 mg, energy-93 mg, calcium-1130 mg, phosphorus-80 mg, iron- 3.9 mg[5].

Figure 1: Flowering twig with leaflets of *Sesbania grandiflora* (L.) Pers

MATERIALS AND METHODS

S. grandiflora leaves were collected in the surroundings of Bangalore, identified through flora and specimen deposited in RRCBI, Herbarium, NADRI, Bangalore (RRCBI-Mus/06). Microscopical studies were carried out

by free hand sections of the leaflets cleared with chloral hydrate solution, water and stained with safranin[6]. Microphotographs of free hand sections were obtained under Trinocular microscope with Nikon Digital camera (Nikon coolpix 4500). Stomatal index, vein islet number, palisade ratio and stomatal number were determined[7]. Leaf sample was Shade dried, coarsely powdered and used for Physico- chemical, Phytochemical, fluorescence and TLC Studies[8-10].

RESULTS

Powder Microscopic Studies

Leaflets of *S. grandiflora* are dorsiventral, upper and lower epidermis are single layered covered by thin cuticle. Upper epidermis is followed by 2-3 layers of narrowly arranged angular collenchymatous cells and 1-4 layers of closely arranged parenchymatous cells in the form of neck of the round bottom flask (Figure 2, 5). Centre portion of the midrib region is occupied by well developed vascular bundle which is conjoint, collateral, closed with metaxylem facing towards the lower region and protoxylem facing towards the upper region (Figure 2, 4). Lower region of the leaf let is planoconvex in structure consisting of 2-3 layers of angular collenchymas followed by 1-3 layers of parenchyma. About 1-4 layers of collenchymatous cells are arranged near the vascular bundle region (Figure 3). Microscopically laminar region shows single layered upper and lower epidermis, 2 layered palisade parenchyma and loosely arranged spongy parenchyma cells (Figure 6). Stomata are of anisocytic, present on both surfaces but more towards the lower surface (Figure 7).

The quantitative values of the upper and lower surface of stomatal index is 15-20-26 per sq mm, and 16-22-28 per sq mm respectively. The palisade ratio is 4.20-5.50 per sq mm. The vein islet number is 20-30 per sq mm and the upper and lower surface Stomatal number is 4-6-8 per sq mm,5-7-9 per sq mm respectively.

Diagnostic characters of Agathi

- Presence of 1to 4 layered angular collenchymatous tissue near the vascular bundle towards the lower region.
- Presence of prominent 2-3 layered narrowly arranged angular collenchymatous cells and 1-4 layers of closely arranged parenchymatous cells in the form of neck of the round bottom flask towards the upper epidermal layer.
- Presence of anisocytic type of stomata on both surfaces of the leaf let, more towards the lower surface.
- Presence of conjoint, collateral, closed Vascular bundle in the midrib region of the leaf let.

Physicochemical Studies

The results of physicochemical studies are presented in table 1. Further, the sample tested positive for saponins which is evident by the percentage of chloroform and petroleum ether extracts. The sample also contained traces of tannins. The swelling index is 9ml and the foaming index is <100. The sample contains inorganic constituents like bicarbonates (HCo_3), sulphates (SO_4), chlorides (Cl_3), calcium (Ca), Sodium (Na), calcium phosphate ($CaPO_4$), iron (Fe) and magnesium (Mg).

Fluorescence Studies

The fluorescence behavior of the powder with different reagents under day light and short ultra violet light showed distinct characteristic features. The details are depicted in table 2.

Thin layer chromatography studies (Table 3)

Thin layer Cromatography study has been carried out with petroleum ether, Chloroform and ethanol extracts by using solvents such as benzene: ethanol (19:1), Chloroform: methanol (19:1) and Toluene: ethyl acetate (90:10) respectively. Plates viewed under ordinary light, UV long wave 365nm and UV short wave 254nm. Rf values of the various spots were calculated. The Rf value of the petroleum ether extract has shown nine spots 0.08, 0.13, 0.2, 0.5, 0.58, 0.71, 0.8, 0.86, 0.94; similarly Chloroform extract 0.05, 0.19, 0.28, 0.35 and the ethanol extract 0.05, 0.14, 0.29, 0.54, 0.77 .

Table 1: Physicochemical Analysis

S.No.	Parameters	Results
1	% Foreign matter (w/w)	< 2
2	% Loss on drying at 110^0C. (w/w)	9.51
3	% Ash content (w/w)	8.65
4	% Water soluble ash (w/w)	1.74
5	% Acid insoluble ash (w/w)	0.26
6	% Extractive values: (w/w)	
	a. Petroleum ether	6.92
	b. Chloroform	2.46
	c. Ethanol	22.68
7	% Solubility at room temp. (w/w)	
	a. Ethanol	24.86
	b. Water	41.71
8	% Extractable matter (Hot) (w/w)	51.10
9	% Tannins (w/w)	Traces
10	Swelling index (w/w)	9ml
11	Foaming index (w/w)	< 100
12	Organic constituents (Qualitative)	Steroid, saponin
13	Inorganic constituents (Qualitative)	HCO_3, SO_4, Cl_3, Ca, Mg, Na, $CaPO_4$, Fe

Table 2: Fluorescence Studies

[Powder (P) + reagent]	Ordinary light	U.V. Long wave 365 nm	U.V. Short wave 254 nm
Powder	G	Fl. W	G
P + Water	G	Dark Y	G
P + 1N. HCl	Br	Br	Dark G
P + 1N. NaOH	G	Mustard	Dark G
P+1N. NaOH In MeOH	Dark G	Light lemon Y	Dark G
P + 50% KOH	Dark G	Pinkish O	Deep G
P + 50% H2SO4	Deep G	Br	G
P + Con. H2SO4	Bl	Fl G	G Bl
P + 50% HNO3	Br	Br	G
P + Con. HNO3	Br	Light Br	Br G
P + Acetic acid	Bl Br	O	Dark G
P + Iodine water	G	Dark Br	G

(G: Green; Bl: Black; Br: Brown; Y: Yellow Fl: Fluorescent; O: Orange ; B: Blue; Cr: Cream; W: White)

Table 3: Thin Layer Chromatographic (TLC) Studies

S.N	Extractives	Adsorbent	Solvent system	Spraying reagent	Rf. Values
1	Petroleum-either 60-80°C	Silica gel 60 F 254 pre coated sheets	Benzene:Ethanol (19:1)	10%H2SO4 in Methanol	0.08, 0.13, 0.2, 0.5, 0.58, 0.71, 0.8, 0.86, 0.94.
2	Chloroform	Silica gel 60 F 254 pre coated sheets	Chloroform: Methanol (19:1)	10%H2SO4 in Methanol	0.05, 0.19, 0.28, 0.35.
3	Ethanol	Silica gel 60 F 254 pre coated sheets	Toluene:Ethyl acetate (90:10)	10%H2SO4 in Methanol	0.05, 0.14, 0.29, 0.54, 0.77.

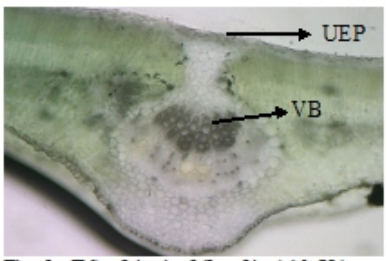

Fig. 2. T.S.of the leaf (Leaf let) 10xX4x Ground plan

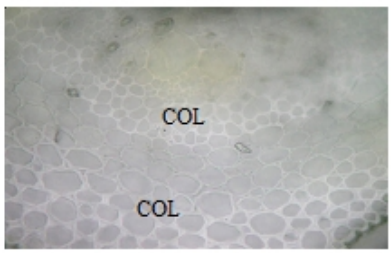

Fig 3. Lower Region enlarged 10xX40x

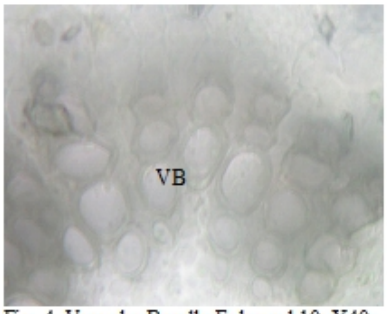

Fig. 4. Vascular Bundle Enlarged 10xX40x

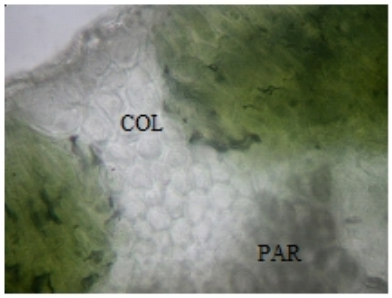

Fig 5. Upper region Enlarged 10xX40x

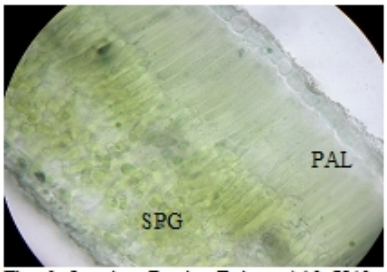

Fig .6. Laminar Region Enlarged 10xX40x

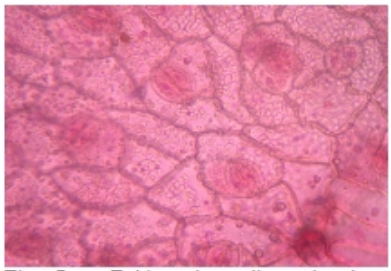
Fig. 7. Epidermal peeling showing stomata 10Xx40X(Anisocytic type of Stomata) Lower Region

Figure 2 - 7: Microscopical characteristics of *Sesbania grandiflora* (L.) PERS

COL: Collenchyma, PAR: Parenchyma, PAL: Palisade tissue, SPG: Spongy Parenchyma, UEP: Upper Epidermis, VB: Vascular Bundle

DISCUSSION

Traditional Importance

The plant Agasthya is a traditionally revered plant, used for internal consumption and as offerings in religious observances especially in South India. Among the most popular traditional use is intake of the plant with other ingredients while breaking a religious fast known as Ekadashi (11 day of the lunar cycle) fast. Indian religious customs include observance of fast on auspicious days. Ekadashi is one such day during which most of the South Indians fast for spiritual attainment. Fasting practices like this are a part of customs that are followed to achieve control over the mind and attain spiritual and academic gains. Although this custom is followed as a religious ritual, it has implications on healthy living. Calorie restriction twice every month in this way keeps the gastrointestinal tract and metabolism active. Further, according to Ayurveda fasting is a good method of health upkeep which is agreed upon even in the modern medical science. It is hence obvious that traditional Indian practices have been designed keeping in mind, health preservation. Fasting makes the body unfit for digestion of heavy to digest food articles, after the break of fast. Hence breaking of fast is usually done with a glucose rich fluid and then slowly solid food articles are introduced. Fasting also causes depletion of energy reserves. Hence during breaking of fast (after ekadashi) a curry prepared with leaves of Agasthya , red gram, and coconut gratings along with chutney (paste) of amla (Indian Gooseberry) rejuvenates the body. It is said that the word Agasthya to the plant is derived from the rishi of the same name. An interpretation of the name is to 'cleanse'. It is believed that Maharshi Agasthya (learned monks from ancient India) came to the south India and cleansed the society of all ills to make them civilized. Consistent with this analogy, leaves of plant Agasthya when taken orally cleanse the body of toxins and is rejuvenating. It restores physiological compromises as a result of fasting. A traditional use of the plant therefore by implication provides insights into probable presence of certain useful constituents possessing therapeutic potential. These uses could be taken as leads to pose research questions at basic as well as clinical evaluation of the plant.

Physicochemical properties

Physicochemical studies revealed that loss on drying in the sample can be attributed to loss of water soluble constituents. Water soluble ash was negligible**.** Acid insoluble ash percentage which is generally due to the presence of silica was also negligible in the sample. Various extractives were observed and recorded. Increased proportion of ethanol extractives in the samples is indicative of a relatively predominant proportion of phytosteroids. The sample also contained traces of tannins. The swelling index of *S.grandiflora* was almost same as ispaghulla seed (>9 ml) [11] which indicates a very high polysaccharide and mucilage content of the plant. The foaming index of *S.grandiflora* is indicative of presence of saponins. Presence of saponins and tannins in the leaves are preliminary indicators of the rejuvenative potential of Sesbania leaves.

Thin layer Chromatography (TLC) Fluorescence Studies

TLC studies on the petroleum ether extract of the plant in benzene: ethanol (19:1) solvent system revealed presence of compounds corresponding to Rf values 0.08, 0.13, 0.2, 0.5, 0.58, 0.71, 0.8, 0.86, 0.94. These compounds among others could include lipid based compounds and fats which are known to play an important role in cellular protective mechanisms that play an important role in healing processes. Similarly, TLC of Chloroform extract of the plant in Chloroform: methanol (19:1) solvent system produced presence of compounds corresponding to Rf values 0.05, 0.19, 0.28, 0.35 chloroform extracts are specific to glycosides apart from alkaloids and other compounds including amino acids and proteins with free radical scavenging properties. Finally, TLC of Ethanol extract of the plant in Toluene: ethyl acetate (90:10) solvent system demonstrated presence of compounds corresponding to Rf values 0.05, 0.14, 0.29, 0.54, 0.77. This suggests presence of poly-phenols among other compounds. Many of the herbs used in Ayurveda system of medicine have known to possess antioxidant properties due to the presence of polyphenols. Further studies with HPTLC could bring out evidences for the specific compounds with properties consistent with the healing effects of the plant both systemically and at the cellular level. The UV and ordinary light Fluorescence depicted a specific pattern helping in establishing a parameter for specific identification of the plant.

Powder Microscopy

Studies on the leaves proved to be very useful in identification of unique diagnostic characters like presence of 1to 4 layered angular collenchymatous tissue near the vascular bundle towards the lower region, Presence of prominent 2-3 layered narrowly arranged angular collenchymatous cells presence of 1-4 layers of closely arranged parenchymatous cells in the form of neck of the round bottom flask towards the upper epidermal layer and identification of anisocytic type of stomata on both surfaces of the leaf let, more towards the lower surface and Presence of conjoint, collateral, closed Vascular bundle in the midrib region of the leaf let. Hence the powder microscopic studies of the leaf provides us diagnostic characters of the plant that can go a long way in pharmacognosy and authentic identification

Studies on *Sesbania grandiflora*

Extensive studies on the use of *Sesbania* species as forage have proved that the nutritive effects and weight gain achieved is significantly higher than other greens. Further they grow rapidly, fix extensive nitrogen and improve soil fertility[12] Leaves of *Sesbania grandiflora* have been reported to have potent antioxidant activity[13]. They have also been found to be rich in beta carotene[14].This might be the reason for wide ranging potential medicinal uses of these leaves. *Sesbania grandiflora* leaves have exhibited wide spectrum of anticonvulsant profile and anxiolytic activity[15]. A recent study has also shown potential anticancer and chemopreventive efficacy[16] potent antioxidant and cardioprotective properties[17].

SUMMARY AND CONCLUSION

Traditional uses of Agasthya provide a source for exploration of its potential role in therapeutics in the background of lifestyle related non communicable disorders assuming alarming proportions in India and across the globe. Therapeutic attributes of Agathya as per Ayurveda are not only consistent with its traditional usage but also provides immense information with respect to its therapeutic use in a wide range of disorders. With no effective answers to manifold life style disorders like Diabetes Mellitus, Cardiovascular Diseases, Major Depression and so on, the traditional utility of Agasthya as a tonic, rejuvenative and Ayurvedic information were the motivating factors behind studying the plant with respect to physico-chemical, powder microscopy and TLC parameters vis-à-vis traditional usage and Ayurvedic indications. Previous studies on *Sesbania grandiflora* demonstrates that apart from improving soil fertility it has broad spectrum antioxidant property that can be attributed to beta carotene, saponins and tannins among many useful compounds. Further, anxiolytic, anticonvulsant and cardio protective potentials hint at a holistic mechanism of action in initiating anti-ageing potentials.

With no such published documentation available on Agasthya, the current study contributes to the field of science with pioneering preliminary findings with regards to physicochemical properties, presence of useful constituents through TLC, Microscopic diagnostic characteristics though powder microscopy and provides enough scientific material to moot research questions and initiate future studies with advanced parameters both in terms of basic science like HPTLC and clinical research so that the benefits of Agasthya reaches out in protection, preservation of health and therapeutics.

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