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PHYTOCHEMICAL AND PHARMACOGNOSTICAL STUDIES ON *STYLOSANTHES FRUTICOSA LINN*

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ABSTRACT

Stylosanthes Fruticosa Linn (*Fabaceae*) is a copiously branching woody herb or ascending shrub, and traditionally it has been used for diabetes, antihelmenthiasis and various other disorders. Since there is no other data regarding this plant our efforts were devoted to study the morphological, microscopical (transverse section and powder microscopy), fluorescence analysis, proximate analysis, Measurement of length and width of fibre and preliminary phytochemical profile of *Stylosanthes Fruticosa Linn.*were studied and documented as per the standard procedures available in the World Health Organization Geneva.

Keywords: *Stylosanthes Fruticosa,* Morphology, Microscopy, Phytochemical, Proximate Analysis.

INTRODUCTION

***Morphological Description*:**

Copiously branching woody herb, ascending shrub or under shrub, reaching 50 cm in height. Branches densely clothed with short yellowish pubescence. Leaflets oblanceolate narrowed to both ends, long mucronate at the apex, 9 to 18 mm long, prominently nerved, and both surfaces nearly glabrous, Flowers in dense oblong terminal heads. Pod with two articulations, about 6 mm long, both faces and remains of style densely silky (Andrews, 1952). Beaks 1.5 to 3 mm long and the plant have evenly pubescent stems. It is a perennial which may behave as an annual in the subtropics. *Distributions:* Native to the South Sahelian and North Sudanian ecozones from Senegal to Rep. of Sudan (Kordofan) and to East and South Africa. Found in the Sudan, Nigeria, Kenya, Uganda, Tanzania, Zambia, Mozambique, Zimbabwe, South Africa and south India
[1-3].

MATERIALS AND METHOD

The plant was identified and Authenticated by Dr. B.Ravi Prasad Rao, Professor, Dept of Botany, S.K.University, Anantapur, Andhra Pradesh. Prepared herbarium was submitted and the plant was certified as *Stylosanthes Fruticosa Linn* under the family: *Fabaceae*.

Transverse section of leaf

A thin T. S was taken by free hand using a sharp razor blade in the laboratory. Phloroglucinol and hydrochloric acid in the ratio of 1:1 was used as a stain and mounted by using glycerin with the help of cover slip on a glass slide and focused under microscope.

Powder analysis of leaf

Shade dried whole plant was powdered with the help of an electric mixer grinder till the moderately fine powder was obtained. Powder was subjected to analyse powder microscopy using equal quantities of chloral hydrate and hydrochloric acid as stain. Mounted on a glass slide and focused under microscope.

Fluorescence analysis

The leaf powder was mixed with different types of solvents like 1 N hydrochloric acid, 50 % sulphuric acid, 40 % sodium hydroxide, 40 % sodium hydroxide-ethanol.

Determination of leaf constants

The different parameters like stomatal number, stomatal index, vein islet number, vein termination number and palisade ratio was determined as per the standard procedure available in WHO guidelines.

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Proximate analysis

The various physicochemical parameters like ash values, extractive values, and crude fibre content was determined as per the procedure available in the WHO guidelines.

Measurement of length and width of fibre

The experiment was performed as per the standard procedures [4-6].

RESULTS AND DISCUSSIONS

Transverse section

A thin T. S of leaf showed dorsiventral nature. The following tissues were observed under lamina and midrib region (showed in Figure 1). Few anomocytic type of stomata were present on the upper epidermis. Single layer of elongated palisade cells were seen below the upper epidermis. Calcium oxalate crystals were present in the spongy parenchymatous cells. Trichomes and stomata were present in more in lower epidermis than the upper epidermis. Below the upper epidermis & above the lower epidermis collenchymas cells were present. Arc shaped vascular bundles were present with which were surrounded by pericyclic fibers. Xylem towards the ventral surface and phloem towards the dorsal surface were observed. Collateral type of vascular bundles was observed.

Powder microscopy

When observed under microscope this revealed the presence of calcium oxalate crystals, lignified fibres and trichomes (showed in Figure 2).

Fluorescence analysis

Different colour ranges were obtained for the whole plant powder of *stylosanthes fruticosa linn* in different reagents was expressed in Table 1.

Determination of leaf constants

The leaf constants like palisade ratio, vein islet and termination number, stomatal number and index were obtained and tabulated in Table 2.

Proximate analysis:

The values of ash values and extractive values were given in Table 3.

Measurement of length and width of fibre: was expressed in Table 3.

Preliminary Phytochemical Analysis:

Qualitative phytochemical studies of different extracts of whole plant of stylosanthes fruticosa linn were performed on its alcoholic, hydroalcoholic (70% alcohol) and water extracts to identify its Alkaloid, Carbohydrate and Glycoside, Saponin, Protein & Amino acid, Phenolic compounds & Flavonoids and Phytosterols by using suitable chemicals and reagents (Table 4).Alkaloid test results of leaf, stem and root showed slightly positive in all four tested reagents. However 70% alcoholic extract of leaf, stem and root showed negative in Mayer's test. Qualitative phytochemical studies of Carbohydrate & Glycoside showed a good characteristic colour and precipitate in all five tested reagent. Slight presence of Saponin was confirmed by foam test in leaf, stem and root in all extracted solvents. Protein and amino acid was found absent in all tests. However in Millon's test alcoholic extract showed slight presence of protein. Phenolic compounds and Flavonoids were abundantly present in all the extracts. However alkaline test showed the moderate result in comparison to other two tests [7-11].

Libermann-Burchards test showed slight presence of phytosterol in all the extracts. The above qualitative phytochemical screening showed that the whole plant is a rich source of Glycosides, Phenols & Flavonoids. However, presence of protein and alkaloids is limited in whole plants [12,13].

Fig. 1: T.S of the leaf of *stylosanthes fruticosa linn*

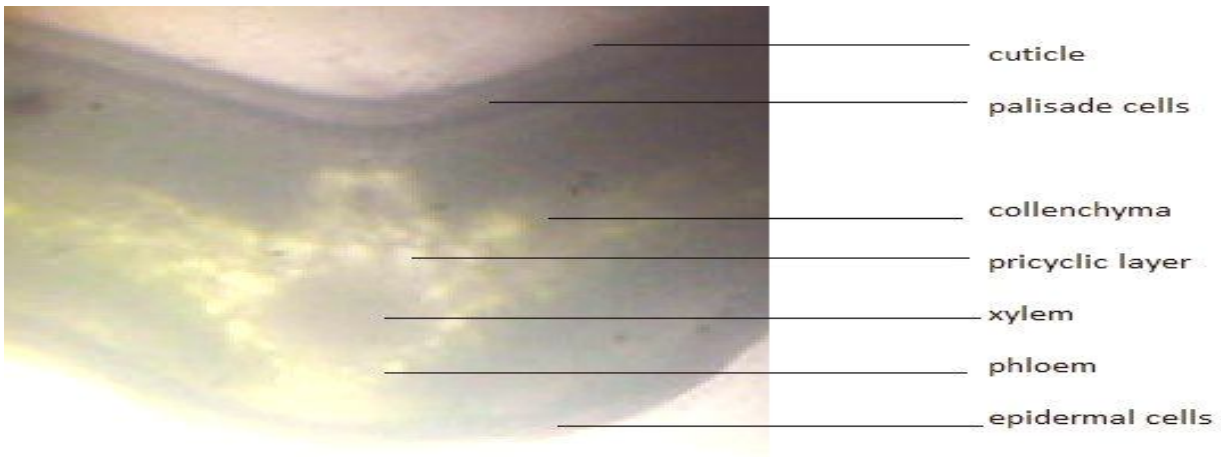

Fig 2: Powder microscopy of whole plant of *stylosanthes fruticosa linn*

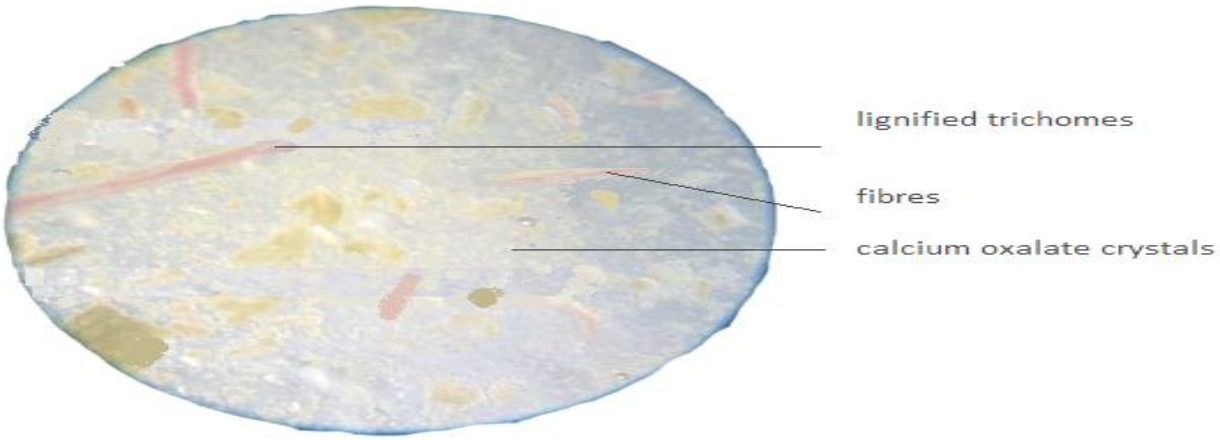

Table 1: Fluorescence analysis

	Ordinary Light	**UV Long**	**UV Short**
Powder as such	Green	Green	Dark green
Powder + 1 N HCl	Yellowish green	Blackish green	Dark green
Powder + 50 % sulfuric acid	Brownish black	Black	Dark green
Powder + 40 % NaOH	Yellowish green	Dark green	Greenish yellow
Powder + 40 % NaOH-ethanolic	Dark green	Blackish yellow	Green

Table 2: Proximate analysis

Moisture content	Not more than 7
Solubility	Completely soluble in alcohol, partially soluble in water
Ash values	Total ash: 20 % acid insoluble ash: 7.55 % water soluble ash: 25 %
Extractives	Water soluble extractive: not less than 32 % w/w, Alcohol soluble extractives not less than 6.2 % w/w
Crude fibre content	15.35 residue (Dutch method)

Table 3: Determination of leaf constants

Palisade ratio:	Upper surface 5.5.i.e; 1:5 Lower surface: 3.5 i.e:1:3
Vein islet Number:	3
Vein Termination Number:	6
Stomatal Number	Upper Surface: 3 Lower surface: 5
Stomatal Index	Upper surface: 42.8-50.0 Lower surface: 55.5-60.0
Measurement of fibre	
Length of fibre	42.9-104 µm
Width of fibre	4.8-17.6 µm

Table 4: Qualitative Phytochemical Screening of whole plant of *Stylosanthes Fruticosa Linn*

Phytochemical test	Cold Maceration			Sohxalation
	Alcoholic Extract	Hydro Alcoholic Extract	Aqueous Extract	Ethanolic Extract by Sohxalation
1. Alkaloids				
Mayer's test	+	+	-	+
Wagner's test	+	+	+	+
Hager's test	+	+	+	-
Dragendorff's test	+	+	+	+
2. Carbohydrates & Glycosides				
Molish's test	+++	+++	+++	+++
Fehling's test	+++	+++	+++	+++
Barfoed's test	+++	+++	+++	+++
Benedict's test	+++	+++	+++	+++
Borntrager's test	+++	+++	+++	+++
3.Saponins				
Foam test	+	+	+	+
4. Proteins & amino acid				
Millon's test	+	+	-	-
Biuret's test	-	-	-	-
Ninhydrin test	-	-	-	-
5. Phenolic compounds & flavonoids				
Ferric chloride test	+++	+++	+++	+++
Lead acetate test	+++	+++	+++	+++
Alkaline test	++	+	++	++
6. Phytosterol :				
Libermann-Burchard's test	+	+	+	+
-, Negative; +, Slight; ++, Moderate; +++, Frequent;				

CONCLUSION

Physicochemical studies finding posses total and water soluble ash content has been higher in stem and acid insoluble ash higher in root, it may be due to the earth components. Extractive value has been found higher in stem water extract, however alcoholic extract has found higher in leaf. Total carbohydrate content has found higher in leaf however, protein, phenol and tannin content found higher in stem portion of *Stylosanthes Fruticosa Linn.*

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