PHYTOCHEMICAL EVALUATION OF LEAF AND STEM EXTRACTS OF SIDDHA MEDICINAL PLANT: SIDA CORDATA

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ABSTRACT: Phyto chemicals are the secondary metabolites produce by the plant for its adaptation, which has medicinal value. The plant Sida cordata is a prostrate herb with medicinal value which is found throughout India .The whole plant Sidda cordata is used by the tribal people of Madekeri district to treat various aliment like hepatic disorder, dysentery, cholera etc, it is also one of the component in herbal preparation in Tamilnadu used on cut wounds, to relive pain etc. The traditional medicine involves the use of different plant extracts or bioactive component for the treatment of different health problems at affordable cost. Secondary metabolites are responsible for medicinal activity of plant. Hence, the present study deals with the preliminary phytochemical evaluation of leaf & stem of Sida cordata, the study includes the preparation of different extracts leaf &stem by successive solvent extraction method fluorescence analysis of successive extracts & their powder are noted under visible &UV light, which showed the visibility of varying colors. Further Phytochemical analysis of leaf & stem extracts of Sida cordata was carried out which confirms the presence of primary metabolites like carbohydrates, amino acids, proteins etc and secondary metabolites like the alkaloids, flavonoids, tannin etc. Present study is designed to explore the preliminary phyto-profile and phytochemical analysis of leaf & stem of Sida cordata, which are responsible for its pharmacological properties.

KEYWORDS: sidda medicine, Sida cordata, phytochemical screening, successive solvent extraction.

1. INTRODUCTION: Siddha system Medicine is one of the oldest medical systems known to mankind. This system of Siddha medicine is in usage of herbs, metals, and minerals, has originated in Southern India, in the state of Tamil Nadu, as part of the trio Indian medicines, Ayurveda, Siddha, and Unani & reported to have surfaced more than 2500 years ago. It is assumed that when the normal equilibrium of the three humors (vata, pitha, and kapha) is disturbed, disease is caused. The factors, which assumed to affect this equilibrium, are environment, climatic conditions, diet, physical activities, and stress. Under normal conditions, the ratio between these three humors (vata, pitha and kapha)i.e.:(Vadham, Pittham, Kabam in Tamil) is 4:2:1, respectively. It has both preventive and curative treatments. Preventive include kayakalpa medicines. These Kayakalpa drugs counter the degenerative changes, which leads to aging by means of the Natural antioxidant activity of the herbs. Practicing Siddha medicine is an art rather than a profession. SIDDHA SYSTEM TEACHES us "TO LIVE WITH NATURE".

Sida cordata (Burm.f.)^[1] Waalkes, belongs to the family Malvaceae, is distributed in India, Pakistan and other tropical countries. It is used for the medicinal purposes in the codified Indian systems of medicine namely Ayurveda and Siddha. The whole plant material is believed to rejuvenate and it is also given for chronic liver diseases [2,3,4]. And the roots of these plants along with cow' butter is applied locally to cure piles and relif from the pain .^[5] But it has not been explored properly and remains a silent drug in herbal medicine. It is stated in traditional Siddha literature under the author Bhava Mishra, 'Bhava Prakash Nigandu' ^[6,7,8]. Medicinal herbs have been used in one form or under indigenous form of medicine. Plants have an almost limitless ability to synthesize aromatic substances mainly secondary metabolites, of which at least 12,000 have been isolated, which is less than 10% of the total. In many cases, these substances serve as the molecules of plant defense against predation by microorganisms, insects, and herbivores. Further, some of which may involve in plant odor (terpenoids), pigmentation (tannins and quinines) etc., It is now clear that, the medicinal values of these plants lie in the bioactive phytochemical constituents that produce definite physiological effects on the human body. Though the traditional Indian system of medicine has a long history of use, they lack adequate scientific documentation, particularly in light modern scientific knowledge^{[9].} These natural compounds formed are the base of modern drugs which we are using today. [10,11,12] Phytochemicals may protect human beings from various diseases, these are nonnutritive plant chemicals that have protective or disease preventive properties. Phytochemicals are basically divided into two groups that are primary and secondary metabolites according to their functions in plant metabolism. Primary metabolites includes common sugars, amino acids, proteins and chlorophyll while secondary metabolites consist of alkaloids, flavonoids, tannins and so on^[13, 14]

The present study is carried out on, various leaf & stem extracts of the plant **Sida cordata** to explore its phyto constituents which are responsible for its pharmacological properties.

MATERIALS AND METHODS

2.1 Plant Collection.

Fresh plant material, leaves & stem of Sida cordata was collected from its natural habitat, from the forest region of Somawarpet in Madekeri district Karnataka. The taxonomic identification of this plant was done by the taxonomist from the Dept. of Botany Mysore university Mysore. The collected fresh plant materials (leaves & stem) were washed in water, shade dried at room temperature and then homogenized to fine powder of 40 mesh size and stored in airtight bottles at 4°C.

2.2. Extraction of Plant Material.

About 100gm of each leaf & stem powder were subjected to extraction by a hot percolation method with 150ml of solvents in their increasing polarity (petroleum ether, chloroform, ethyl acetate, ethanol water respectively), in soxhlet apparatus. Each solvent extraction step was carried out for 24 hrs. After extraction, the extracts were concentrated by evaporation and stored at 4°C for further study.

Powder & extracts of leaf and stem were used to determine the physicochemical parameters like extractive values, florescence characteristics, preliminary phyto-profiling and phytochemical analysis,

2.3. Phytochemical screening: The primary metabolites like proteins, carbohydrates and fixed oils ,fats, etc and the secondary metabolites like, alkaloids, flavonoids, saponins, phenolics, tannins volatile oils, terpenoids, glycosides etc were assessed in the leaf and stem extracts of Sida cordata. as per the standard procedures. ^[15,16,17].

All the data generated from the study were subjected to arithmetic mean with standard deviation for statistical

RESULTS: The leaf & stem powder and their extracts were subjected to physiochemical & phyto chemical analysis and the results obtained were found to be very promising .All the results obtained from the present study are represented in the respective tables (1,2,3,&4).

The percentage of yields and the consistency of various extracts were tabulated in table 1.

The consistency was found to be sticky in the non-polar to less polar solvent extracts, whereas the polar solvent extracts were found to be non-sticky. Highest % of yield (w/w) was recorded in aqueous extract of both leaf &stem (2gms & 1.8 gms respectively). The fluorescence characteristics of leaf & stem powder followed by leaf & stem extracts was studied under ordinary and UV light (366nm). Which showed the visibility of varying colors which are tabulated in the Table no.2 and 3. The preliminary phytochemical screening revealed the presence of most of these compound which are tabulated in table. 4

Primary metabolites like, Protein, Carbohydrate, Fixed oils, fats and secondary metabolites like, alkaloids, flavonoids, saponins, volatile oils, phenols and tannins, glycosides, terpenoids etc were tested .Our investigation result showed that the plant possesses the primary metabolites like, Carbohydrates in all extract of leaf & stem, however Proteins were found in the aqueous extract only, free amino acids were detected in all the leaf & stem extracts but, sulphur containing amino acids were absent. Tannins were found in all the leaf & stem extracts except in the stem extract of ethyl acetate & ethanol. Flavonoids were detected in all the leaf extracts & in stem it is present in petroleum ether and ethanol extracts only. Saponins were found only in the ethanol & aqueous extract of both leaf & stem. Steroids were found in all the leaf extracts & in stem it was found only in the aqueous extract. Phenols were found in the ethyl acetate & aqueous extract of both stem & leaf. Alkaloids were found in all the leaf extract and completely absent in stem extracts. Anthrocyanins were found only in the ethanol & aqueous extracts of leaf, it is absent in the all the stem extracts. Coumarins were present in the ethyl acetate extract of leaf only. Emodin were found only in the ethanol extracts of leaf and it is absent in the all the stem extracts. Anthraquinones were found only in the ethanol & aqueous extracts of leaf, and it is absent in the all the stem extracts. Catechins were found in all the leaf extracts & in stem it was found only in the aqueous extract.

DISCUSSION: The preliminary phytochemical analysis results revealed the presence of Carbohydrates in all extracts, this may be because these are the essential biomolecules synthesized by the plants abundantly for its energy requirement and other physiological processes. Proteins are present in very negligible amount in the aqueous extracts, this may be due to ,their activity might have been suppressed by the presence of other secondary metabolites, and in addition, probably the solvent might have denatured the proteins because free amino acid were found in all most all the extracts and proteins in the aqueous extract only. Fats & Volatile oils were completely absent.

The presence of, tannins^[18] as a phyto-chemical in Sida cordata leaf & stem extract reveals that it is useful in the treatment of inflammatory conditions like ulcers and they have

remarkable activity in cancer prevention .^[19] The Flavonoids ^[20] present in extracts are of great importance ,as these are involved in cell protection via their action on membrane permeability, and by inhibiting membrane-bound enzymes such as the ATPase and phospholipase A₂ ^[21]. These observations support the usefulness of this plant in folklore remedies in the treatment of stress related ailments and as a dressings for wounds normally encountered in circumcision rites, bruises, cuts and sores ^[22]. Saponins present in the extracts suggests that this plant may become one of the possible source in the treatment of cancer, because Saponins, are anti carcinogenic agents as they possess surface-active characteristics due their amphiphilic nature of their chemical structure, the proposed mechanisms of anticarcinogenic properties of saponins include direct cytotoxicity, immune-modulatory effects, bile acid binding and normalization of carcinogen-induced cell proliferation. However, the anticarcinogenic effects of saponins from commonly consumed plant foods have not been studied. Soybeans are one of the most important sources of dietary saponins. They are the main protein supplier in many vegetarian diets.^[23]

The leaf & stem extract also contains steroids which are very important compounds especially in the synthesis of sex hormone.^[24] The presence of phenolic compounds in this plant contributed to their anti-oxidant properties and thus its usefulness in herbal medicament. Alkaloids present in the extracts have been associated with medicinal uses for centuries and one of their common biological properties is their cytotoxicity ^[25]. Anthrocyanins present in the extract helps the immune system to work more efficiently to protect against viral infection It is little bit more complex, specific types of Anthrocyanins may have a direct effect in decreasing influenza viruses infectivity by decreasing the ability of the virus itself to get into the human cell or to be related [26]. The presence of coumarin in the extracts may serve as a potent natural anti oxidant because various studies have been demonstrated that coumarin is a potential antioxidant and its antioxidant activity is due to its ability to scavenge free radicals and to chelate metal ions ^[27]. Presence of Emodin in the extracts shows that this plant can be used as anticancerous, antimicrobial and anti-inflammatory agent because many pharmaceutical studies have demonstrated that emodin has many biological effects, such as anticancer, antimicrobial and anti- inflammatory effects ^[28]. Prencese of Anthraguinones in the extracts are of great importance as Anthraquinones are considered to be one of the most active agents in metastatic breast cancer. [29]

Presences of Catechins in the extracts are also has greater importance because Catechins transcriptomic studies shows that catechin reduces atherosclerotic lesion development in apo E- deficient mice ^[30]. And catechin seem to have stereospecific opposite effects on glycogen metabolism in isolated rat hepatocytes ^[31]. Catechin inhibits intestinal tumor formation in mice.^[32] Catechin inhibits the oxidation of low density lipoprotein.^[33]. The presence of Emodin, Anthraquinones, and Catechin in the leaf & stem extracts of Sida cordata may be used as a potent drug in the treatment of cancer.

Although, absence of certain phytochemicals in one extract and its presence in the other can be safely attributed to the various physiological and biosynthetic reactions taking place inside the plant, the effect of the environment should not be neglected, as the environment always modify the things. Anti microbial & preliminary phytochemical analysis of various extracts from different plants have been of great interest in both research & also in food industries because of their possible use as natural additives emerges from the growing tendency to replace synthetic antioxidants. By considering the above facts we can conclude that the leaf & stem of the plant **Sida cordata are of great medicinal value**, which is routinely used by many tribal people in the Madikeri district for the treatment of various diseases, leaf & stem containing these compounds may also serve as potent medicament for the treatment of inflammatory disorders and in cancer.

Further studies are in progress on Sida cordata as anti microbial, anti oxidant, anti inflammatory, anti-carcinogenic source and, other biochemical properties, isolation of the bioactive compounds responsible for the therapeutic value for the benefit of human welfare.

s. no.	Solvent used	color		consistenc	y	% of yield w/w			
		Leaves	stem	Leaves	Leaves stem		Stem		
01	Petroleum ether	Green	Light Green	Sticky	Sticky	1.3	0.9		
02	Chloroform	Green	Green	Sticky	Sticky	1.9	1.3		
03	Ethyl acetate	Dark Green	Green	Sticky	Sticky	1.4	1.1		
04	Ethanol	Green	Green	Non Sticky	Non Sticky	1.8	1.2		
05	Water	Brownish	Brownish	Non Sticky	Non Sticky	2.0	1.8		

Table 1: Preliminary phyto-profile for leaves & stem of Sida cordata

S .	Particulars of the	Under ordinary	/ light	Under UV li	ght (366		
no.	treatment			nm)			
					1		
		Leaves	stem	Leaves	stem		
01	Powder as such	Green	Green	Dark green	Black		
02	Powder + 1N NaOH	Greenish	Grown	Black	Black		
	(aqueous)	brown					
03	Powder +1N NaOH	Light green	Light green	orange	Black		
	(alcoholic)						
04	Powder + 1N HCl			yellowish	Yellow		
		colorless	colorless				
05	Powder + $H_2SO_4(1:1)$	Pale yellow	Pale yellow	orange	orange		
06	Powder + HNO ₃ (1:1)	Reddish	Reddish	Black	orange		
07	Powder + Ammonia	Florescent	Florescent	orange	orange		
		green	green				
08	Powder + Iodine	brown	brown	Black	Black		
09	Powder + 5% FeCl ₃	Blackish green	Pale yellow	Black	Black		
10	Powder + Acetic acid	Grey	Greenish	Black	Black		

Sl. No.	Extract	Under ordinary light		Under UV light (366 nm)				
		Leaves	stem	Leaves	stem			
01	Petroleum ether	Green	Green	Black	Black			
02	Chloroform	Florescent Light Screen	Green	Brown	Black			
03	Ethyl acetate	Dark Green	Green	Black	Black			
04	Ethanol	Green	Green	Black	Black			
05	Water	Brown	Brown	Black	Black			

Table4: Phytochemical analysis of different extracts of leaves &stem of Sida cordata

S.	Phyto	Name of the Test		*	Ccl _{4*}		Et.a*		Et.oh*		H ₂ o*	
No.	compounds		L*	S*	L*	S*	L*	S*	L*	S*	L*	S *
01	Alkaloids	a.Mayer's test	+	-	+	-	+	-	+	-	-	-
		b.Wager'test	+	-	+	-	+	-	+	-	-	-
		c.Dragondoff' test	+	-	+	-	+	-	+	-	-	-
		d.Hager's test	+	-	+	-	+	-	+	-	-	-
02	Phytosterols	a. Liebermann Test	+	-	+	-	+	-	+	-	+	+
	/triterpenoids	b. Salkowski Test	+	-	+	-	+	-	+	-	+	+
03	Saponins	a. Froth test	-	-	-	-	-	-	-	+	+	+
		b. Foam test	-	-	-	-	-	-	-	+	+	+
04	carbohydrate	a.Molisch's test	+	+	+	+	+	+	+	+	+	+
		b. Benedict's test	+	+	+	+	+	+	+	+	+	+
05	Tannins	Gelatin test	+	+	+	+	+	-	+	-	+	+
06	Flavonoids	a. Alkaline reagent test		+	+	-	+	-	-	+	+	-
		b. Lead acetate test	+	+	+	-	+	-	-	+	+	-
07	Glycosides	Borntrager's test	+	+	+	+	+	+	-	-	+	+
08	Fixed oil/fat	Spot test	-	-	-	-	-	-	-	-	-	-
09	Phenol	Ferric chloride test	-	-	-	-	+	+	+	-	+	+
10	Gum		-	-	-	-	-	-	-	-	-	-
11	Proteins	a. Biurete test	-	-	-	-	-	-	-	-	+	+
		b. Lead acetate test for	-	-	-	-	-	-	-	-	-	-
		sulphur containing amino										
		acids										
		c. Xanthoproteic test	+	+	+	+	+	+	+	+	+	+

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		d. Ninhydrin test	+	+	+	+	+	+	+	+	+	+
12	Volatile oil		-	-	-	-	-	-	-	-	-	-
13	Coumarin		-	-	-	-	+	-	-	-	-	-
14	Emodin		-	-	-	-	-	-	+	-	-	-
15	Anthrocyanin		-	-	-	-	-	-	+	+	+	+
16	Anthraquinones	Sulphuric acid test	-	-	-	-	-	-	+	+	+	+
17	Catechins	Erhlish test	+	-	+	-	+	-	+	-	+	+

L -leaf, S-stem, PE- petroleum ether, Ccl4, chloroform, Et.a-ethyl acetate, Et.oh-ethanol

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