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Research Article

PRELIMINARY PHYTOCHEMICAL STUDIES AND IN-VITRO ANTHELMINTIC ACTIVITY OF VARIOUS EXTRACTS OF STEM OF *Sarcostemma acidum* (Roth.) Voigt

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Abstract:

Plants are used as medicines by humans and animals, since time immemorial ancient man used many plants or a part of the plant as very good remedies for various ailments it is well known that man depend on plants as a source of food and some of the constituents derived from plants are used as a household remedy by common people and our grandmother's herbal remedies are still effective. The indigenous medicinal system like Ayurveda, Siddha, Unani practiced in India and the folklore of various countries depends mainly on natural plant products, as a source of drugs. The aim of present research is to investigate the phytochemical studies and In-Vitro Anthelmintic activity of stem of *Sarcostemma acidum* (Roth.) Voigt. The phytochemical screening of the various extract of powdered stem of *Sarcostemma acidum* (Roth.) Voigt shows the presence of Secondary metabolite like Alkaloids, sterols, Carbohydrate, proteins, Amino acid, Flavanoids, Phenolic compounds, Tannins, Oils, Saponins and other. The various extract of *Sarcostemma acidum* (Roth.) Voigt were use to evaluate the In-vitro Anthelmintic activity in dose dependent manner the dose of 5mg/ml possess the less time taken for paralysis and more time to cause death, when compared to standard drug. *Sarcostemma acidum* may be consider as Valuable plant in both Ayurvedic and Modern drug development areas of its Versatile Medicinal uses.

Key words: *Sarcostemma acidum*. In Vitro anthelmintic activity.

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INTRODUCTION:

As the half of the world suffering from bacterial and helminths infection. The source of infection being very common due to sanitation, poor family hygiene, malnutrition and crowded living conditions. In recent years, the importance of herbal drugs in medicine has tremendously increased because of their fewer side effects. Consequently, the demand for the herbal formulation is increasing day by day. The phytochemical constituents and their standardization are accelerated with the development of instrumental analysis and this field becomes important and new for investigation. So, there is a need to develop anthelmintic drugs from herbal source. With the advancement of scientific knowledge in chemistry and botany, medicinal plants came under chemical investigation leading to isolation of their active principles. Beginning with 1800 AD, there was continuous activity in these areas and many of the medicinally useful plants were chemically investigated their active principles characterized and used in modern medicines, either in pure state or in the form of standardized extracts.

The chemical investigations of plants, related isolating new compounds and elucidating their structure in order to discover their therapeutic value has been a major concern of scientist. Yet only about 5 % of a total of 6 lakhs plant species growing on earth has been subjected to such chemical and pharmacological investigation. Pharmacognostical research on indigenous drugs and medicinal plants has made rapid stride. Pharmacognosy has steadily grown during this year into a dynamic, wide planked and multi-dimensional study, embracing in its fold biology, chemistry and biochemistry, pharmacology and other disciplines.

COLLECTION AND AUTHENTICATION OF THE PLANT SPECIES

The *Sarcostemma acidum* (Roth.) Voigt. Shrub was collected from the hills of Seranmagadevi in Tirunelveli district, during the month of August-2019 and was Authenticated by V.Chelladurai, Research Officer-Botany, Central Council for Research in Ayurveda & Siddha, Government of india.(Retired) The stem was collected in the month of august using the sharp knife. The stem was cleaned to remove the debris and allowed to shade drying for a week. Selected pieces were coarsely powder by hand grinding mill. The coarsely powdered material was again dried at room temperature in our laboratory. The material is used separately for successive extraction respectively.

PLANT INRODUCTION



Sarcostemma acidum(Roth.)Voigt commonly known as “Somlata,” is an important member of family Asclepiadaceae. According to mythological belief the plant was a source of “Soma” that was used to prepare a rejuvenating drink “Somras,” although this belief is still debated by scholars. *Sarcostemma acidum* (Roth.) Voigt a perennial leafless, jointed trailing shrub with green, cylindrical, fleshy glabrous, twining branches having milk latex, leaves reduced to scales, opposite, flowers are white or pale greenish white ,fragrant ,in umbels on branch extremities, fruits follicles, tapering at both ends, seeds are flat, ovate. Stem is cylindrical and 3.0 – 6.0 mm in diameter. It is green pubescent in young stage and gradually turns glabrous. Internodes are 1.0 – 3.0 mm long. It is propagated by stem cutting with nodes. The plant is a part of traditional medicine among aboriginal communities in India since time immemorial. The plant is known to exist mainly in warmer part of India and generally prefers dry or semi-dried rocky places with low rainfall, shade, and moisture conditions. The plant is a perennial jointed shrub, with fleshy glabrous twig having milky white latex . Leaves are opposite mainly reduced to scales. Roots are brownish, and flowers actinomorphic, with pale green or white in colour. The plant is bitter, cooling, narcotic, acrid, emetic, and rejuvenating containing carbohydrate, terpenes, phytosterols, saponins, and lignin. The plant possesses antimicrobial, anti-inflammatory, antifertility, anti-viral, anti-asthmatic, anti-rabies, anti-ulcer, anxiolytic, antipsychotic, and central nervous system inhibitory property.

AIM AND OBJECTIVE

The aim of present research is to investigate the phytochemical studies and In-Vitro Anthelmintic activity of stem of *Sarcostemma acidum* (Roth.) Voigt.

The present work has been planned to carry out the by Pharmacognostic Studies Authentication and collection of plant followed by Physicochemical parameter: Extractive value with various reagent (Petroleum ether, Chloroform, Ethyl acetate, Ethanol 95%). Phytochemical Studies Preparation of Extracts (Petroleum ether, Chloroform, Ethyl acetate, Ethanol 95%) Qualitative Analysis, Preliminary phytochemical screening of various extracts (Petroleum ether, Chloroform, Ethyl acetate, Ethanol 95%), Pharmacological Studies- In-Vitro Anthelmintic activity by Ajaiyeoba et al., method.

PHYTOCHEMICAL STUDIES:

Phyto Chemical Tests The various extracts of *Sarcostemma acidium* were subjected to following chemical tests

1. Test for Alkaloids The extracts were made alkaline and was extracted with Chloroform and Alcohol separately and evaporated to dryness. Few drops of dilute hydrochloric acid were added and stirred well, filtered. The filtrate was tested with various alkaloidal reagents such as Mayer's reagent (cream precipitate) Dragendroff's reagent (orange brown precipitate) Hager's reagent (yellow precipitate) and Wagner's reagent (reddish brown precipitate).

2. Test for Sterols The residue was dissolved in chloroform and tested for sterols. A. Salkowski Test A few drops of concentrated sulphuric acid was added to the above solution, shake well and set aside. The lower layer of solution turned red in colour indicating the presence of Sterols. B. Libermann-Buchard Test To the chloroform solution a few drops of acetic anhydride and 1ml of concentrated sulphuric acid were added through the sides of the test tube and set aside. At the junction of the two layers a brown ring was formed. The upper layers turned green indicates the presence of Sterols.

3. Test for Carbohydrates A. Fehling's Test A little of extract dissolved in water was treated with Fehling's solution A and Fehling's solution B and heated. A red precipitate was formed indicating the presence of reducing sugars B. Molish's Test A purple colour when treated with alcoholic solution of naphthol in the presence of sulphuric acid was obtained, indicating the presence of carbohydrate.

4. Test for Fixed Oil A few ml of sample is saponified by using alkalies indicates the presence of fixed oil.

5. Test for Phenolic compounds and tannins Small quantities of alcoholic and aqueous extract were taken separately in water and tested with dilute ferric

chloride 5%, 1% solution of gelatine containing 10% sodium chloride, 10% lead acetate and aqueous bromine solution.

6. Test for proteins and Aminoacids A. Biuret Test To one portion of aqueous ethanolic extract, 1ml of 10% sodium hydroxide solution was added followed by this one drop of dilute copper sulphate solution was added. Violet colour was produced. It showed the presence of proteins. B. Ninhydrin Test Same of the extract was dissolved in water, the solution was tested with ninhydrin solution and warmed. A blue colour showed the presence of Amino acid.

7. Test for terpenoids A little chloroform solution was warmed gently with tin and thionyl chloride, the formation of colour indicates the presence of terpenoids.

8. Test for Saponins A few ml of extract was taken and it was haemolysed by means of heating

9. Test for Gums and Mucilages 10ml of aqueous extract of leaves was added slowly to 25ml of absolute alcohol with constant stirring. The precipitate obtained was filtered and dried in air. Then it was examined for the presence of carbohydrates after hydrolysis.

10. Test for Flavones A. Shinoda Test To the ethanolic extract, a few magnesium turnings and few drops of concentrated hydrochloric acid were added. Boiled for five minutes. A red colour produced indicates the presence of flavones. B. Aqueous Sodium hydroxide Test To the few ml of extract add sodium hydroxide solution 10% or Ammonia was added. A yellow colour indicates the presence of flavones.

In-Vitro Anthelmintic activity

Helminthiasis or infection with parasitic worm is pathogenic for human beings. Immature forms of the parasites invade human beings via the skin or Gastro intestinal tract (GIT) and evolve into well differentiated adult worms that have characteristic tissue distribution. Anthelmintic are drugs that act locally to expel worms from the GIT or systemically to eradicate adult helminths or development forms that invade organs and tissues. Most of the existing anthelmintic produces side effect such as abdominal pain, nausea, vomiting, headache and diarrhoea. Chemotherapy is the only treatment and effective tool to cure and control helminths infection as effective vaccine against helminths have not been developed so far. Indiscriminate use of synthetic anthelmintic can lead to resistance of parasite. Most disease caused by helminths are of a chronic and debilitating in nature

and could be of value in preventing the development of resistance.

Worms' collection: *Pheretima posthuma* (Adult Indian earthworm) were collected from the local agricultural land of about 5-7cm long were used for the present study. Standard drug: Albendazole suspension in the concentration of 10mg/ml was used as standard to compare the test result.

In-Vitro Anthelmintic Assay by Ajai yeoba et al., method: The worms were placed Petri dish in solution of crude extracts (Ethanol 95%, Petroleum ether, Chloroform, Ethyl acetate) in three different concentration [5mg/ml, 10mg/ml, 20mg/ml in distilled water] respectively. This was done in duplicates for all the worms' types. Mean time for paralysis (p, in minutes) were taken when no movement of any sort could be observed, expect when the worms were shaken vigorously. Time of death of worms (D, minutes) were recorded after that worms neither moved when shaken vigorously nor when dipped in warm water 50°C. Albendazole (10 mg/ml) was

included as reference compound while distilled water was included as control.

RESULTS AND DISCUSSION:

The percentage of Extractive value of *Sarcostemma acidum* (Roth.) Voigt of various extracts shows in the Table No: 1.1 Phytochemical screening of various extracts shows the presence of Secondary metabolites like Alkaloids, Steroids, Carbohydrates, Proteins, Amino acids, Flavanoids, Tannins, Phenolic compounds, Saponins The various extract of *Sarcostemma acidum* were used to evaluate the In-Vitro Anthelmintic activity showed the effect of in Dose dependent manner. The mean \pm SEM value were calculated. The data shown in the Table No: 1.2 The dose 5mg/ml of various extract possess Less time to taken for Paralysis indicates More time to cause Death of Earthworms. When compared with Standard.

Data shows the Percentage of Extractive value of *Sarcostemma acidum* (Roth.) Voigt of various Extracts

TABLE NO: 1.1

Data shows the Percentage of Extractive value of *Sarcostemma acidum* (Roth.) Voigt of various Extracts

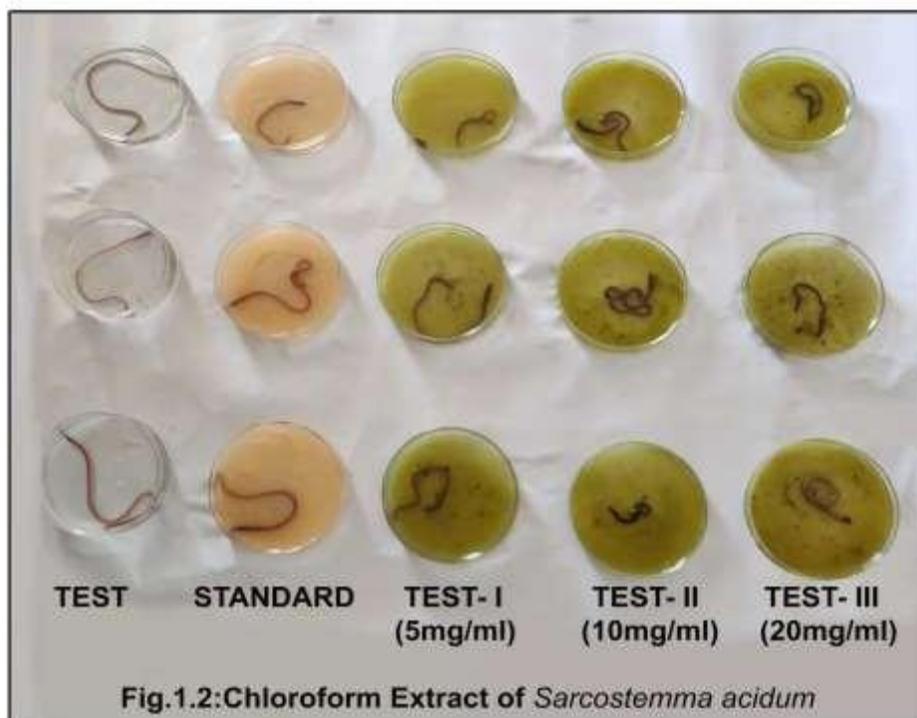
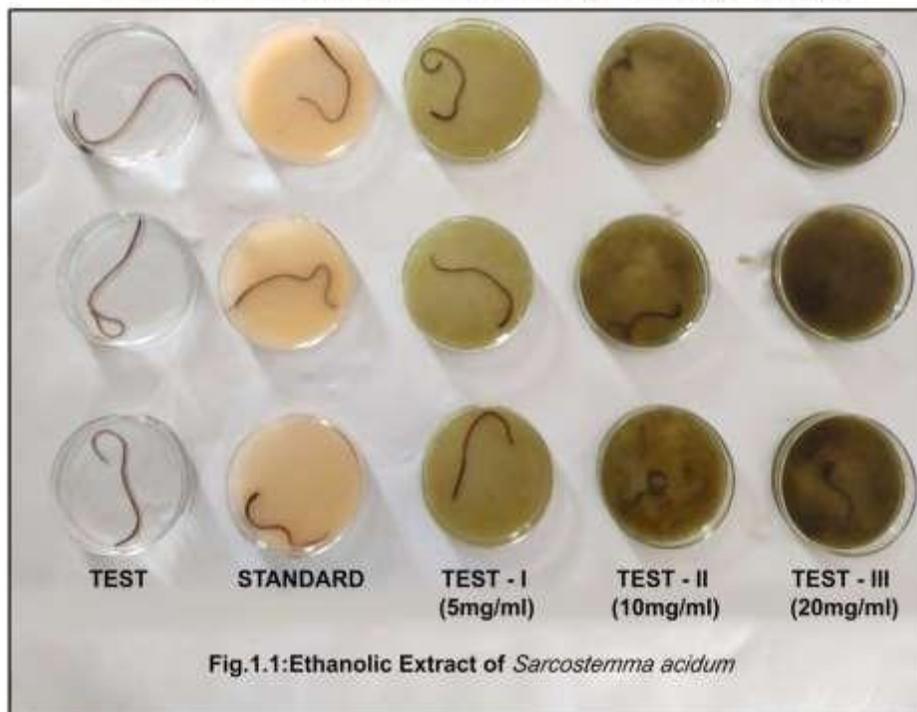
SL.NO	SOLVENT	COLOUR OF EXTRACTS	NATURE OF EXTRACTS	% OF EXTRACTIVE VALUE
1.	Petroleum ether (60-80°C)	Dark Brown	Greasy	3.15
2.	Chloroform	Dark Green	Semisolid Mass	3.13
3.	Ethyl acetate	Dark Greenish Black	Semisolid mass	4.47

TABLE NO: 1.2**In-Vitro Anthelmintic activity of *Sarcostemma acidum***

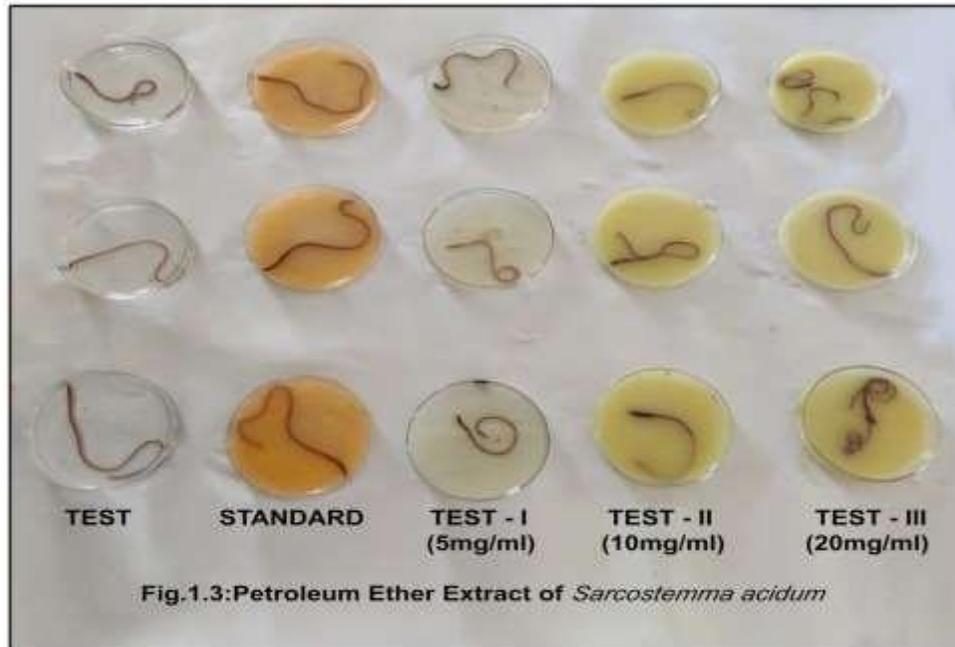
S.NO	Treatment	Concentration (mg/ml)	Time taken for Paralysis (Min) X±S.D	Time taken for Death (Min) X±S.D
01.	Control	-	-	-
02.	Standard (Albendazole)	10mg/ml	25.32±0.123	35.52±0.23
03.	Ethanol extract of <i>Sarcostemma acidum</i>	5mg/ml	32.50±1.44	48.60±1.72
		10mg/ml	29.86±1.05	48.40±1.83
		20mg/ml	22.84±1.98	48.80±1.32
04.	Chloroform extract of <i>Sarcostemma acidum</i>	5mg/ml	31.40±1.52	40.13±1.82
		10mg/ml	32.40±0.12	41.65±1.45
		20mg/ml	36.50±1.32	47.80±1.32
05.	Petroleum ether extract of <i>Sarcostemma acidum</i>	5mg/ml	41.60±1.01	57.00±0.12
		10mg/ml	42.40±0.25	59.80±0.63
		20mg/ml	45.41±0.43	61.00±0.83
06.	Ethyl acetate extract of <i>Sarcostemma acidum</i>	5mg/ml	34.60±0.12	57.40±1.03
		10mg/ml	38.60±1.32	53.00±0.32
		20mg/ml	42.00±1.98	51.10±0.41

All values are mean ± SEM analysis by one way ANOVA

ANTHELMINTIC ACTIVITY OF *Sarcostemma acidum* (Roth.) Voigt



ANTHELMINTIC ACTIVITY OF *Sarcostemma acidum* (Roth.) Voigt



CONCLUSION:

The present study revealed the Various extract of *Sarcostemma acidum* shows In-vitro Anthelmintic activity. The study posses the activity due to the presence of Secondary metabolities like Flavonoids, Alkaloids, Sterols and Glycosides.

Sarcostemma acidum may be consider as Valuable plant in both Ayurvedic and Modern drug development areas of its Versatile Medicinal uses.

As per the study of *Sarcostemma acidum* shows medicinal uses like Thrombolytic activity, CNS Studies, Cell line Studies, Diabetic Studies, Gastro-intestinal Studies and Anti-microbial studies. Hence further studies required for the Active principle of Secondary metabolite for this In-vitro Anthelmintic activity for its effectiveness and pharmacological properties.

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