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# PHYTOCHEMICAL ANALYSIS ON THE LEAF EXTRACTS OF *SIDA CORDIFOLIA*

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**Abstract**

Plants and plant extracts have important role in modern medicine as their chemical and medicinal constituents are found in natural form. The secondary metabolites represent a large reservoir of structural moieties which work together exhibiting a wide range of biological activities. Plants and plant based products are bases of many modern pharmaceuticals that are currently in use for various diseases. Plants and plant based products are bases of many modern pharmaceuticals that are currently in use for various diseases. Several species of Sida, commonly known as "Bala", are known to have analgesic, anti – inflammatory, hypoglycemic and hepatoprotective activity. *Sida cordifolia*, L. commonly called "Country mallow", is a perennial or sometimes annual plant in the family Malvaceae. Present study involved phytochemical screening of leaves of *Sida cordifolia*, L. using six different solvents viz; distilled water, acetone, alcohol, chloroform, petroleum ether and benzene. The extracts prepared from powdered plant parts were subjected to qualitative phytochemical screening using standard procedures. Of the 15 phytochemicals tested, 10 were found in various solvent extracts of *Sida cordifolia*, L. By this study, it was confirmed that the selected plant species is a potent source of useful drugs. However, further studies are required in this direction for its comprehensive analysis including qualitative or semi qualitative analysis, characterize its chemical structure and assess its biological activities.
**Keywords:** *Sida cordifolia,* leaves, phytochemicals, phytochemical screening

## Introduction

The use of plants as medicines goes back to early man. Certainly the great civilisations of the ancient Chinese, Indians, and North Africans provided written evidence of man's ingenuity in utilising plants for the treatment of a wide variety of diseases. In ancient Greece, for example, scholars classified plants and gave descriptions of them thus aiding the identification process. Theophrastus, the father of botany, has been recorded many medicinal plants. Phytochemicals are bioactive chemicals of plant origin. They are regarded as secondary metabolites because the plants that manufacture them may have little need for them. They are naturally synthesized in all parts of the plant body, such as bark, leaves, stem, root, flower, fruits, seeds etc. i.e. any part of the plant body may contain these active compounds (Cowan, 1999). In fact, there is lack of information on the distribution of the biological activity in different plant parts essentially related to the difference in distribution of active compound which are frequent in some plant parts (Mojab, 2003; Parekh and Chanda, 2007; Parekh and Chanda, 2008).

Medicinal plants contain some organic compounds which provide definite physiological action on the human body and these bioactive substances include tannins, alkaloids, carbohydrates, terpenoids, steroids and flavonoids (Edoga, 2005; Mann, 1978). These compounds are synthesized by primary or rather secondary metabolism of living organisms. Secondary metabolites are chemically and taxonomically extremely diverse compounds with obscure function. They are widely used in the human therapy, veterinary, agriculture, scientific research and countless other areas (Vasu, 2009). A large number of phytochemicals belonging to several chemical classes have been shown to have inhibitory effects on all types of microorganisms in vitro (Cowan, 1999).

*Sida cordifolia*, L. Commonly known as Country mallow, under the taxonomic family Malvaceae, is widely distributed along with other species are common throughout the tropical and sub-tropical plains all over India and Srilanka up to an altitude of 1050 m, growing wild along the roadside. *Sida cordifolia* grows well through the plains of India, especially in damp climates. The shrub grows up to 0.75 to 1.5 meters in height. The root and stem are stout and strong. The leaves are 2.5 to 7 cm long and 2.5 to 5 cm broad with 7 to 9 veins. They are heart shaped, serrate and trunicate. The flowers are small, yellow to white in color, solitary and axillary in position. The plant flowers from August to December and fruiting occurs from October to January.

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## Materials and Methods

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## Materials and Methods

***Collection of plant materia***l

The leaves and root of *Sida cordifolia*, L. were collected from five different locations of Thiruvananthapuram. Collected plant parts were shade dried for ten days. Constant monitoring was carried out to avoid microbial contamination. The dried plant materials was taken and ground using motor and pestle to obtain a fine powder. The powder was further passed through a 2 mm sieve to obtain finer particles. The powdered samples were stored in a clean glassware container and stored in low temperature until needed for analysis (Das et al., 2010).

***Preparation of plant extracts***

5 gram of dried and powdered sample was taken. It was put separately in acetone, petroleum ether, chloroform, ethyl alcohol, benzene and distilled water. Mixed and extracted for 24 hours on a stirrer with continuous stirring. After extraction, the extracts were filtered through Whatman No.1 filter paper, centrifuged the filtrate for clarification and stored for further phytochemical investigations (Das et al., 2010).

***Preliminary phytochemical investigations***

Analysis on the presence of different phytochemical constituents such as proteins, amino acids, carbohydrates, alkaloids, saponins, phytosterols, glycosides, phenols, tannins, flavonoids, steroids, terpenoids, and vitamin C were analysed according to the standard procedures, as described below, were used (Harborne, 1998).

***A. Test for Proteins***

Biuret Test:- Test solution was treated with equal volume of 10% sodium hydroxide solution and two drops of 1% copper sulphate solution, mixed well and observed for the formation of violet/pink colour. If it is so, presence of proteins was detected.

Millon's Test:- Two ml of crude extract when mixed with 2ml of Millon's reagent, if a white precipitate appeared which turned red upon gentle heating and disappeared on cooling confirmed the presence of protein.

Xanthoproteic Test:- Two ml of extracts were treated with few drops of conc. Nitric acid. Mixed well. Formation of light to dark yellow colour was noted which indicates the presence of proteins.

***B. Test for Free Amino Acids***

Ninhydrin Test :- Test solution when boiled with 0.2% solution of Ninhydrin. Formation of purple color suggests the presence of free amino acids.

***C. Test for Carbohydrates***

Benedict's test :- Test solution was mixed with few drops of Benedict's reagent (alkaline solution containing cupric citrate complex) and boiled in water bath, observed for the formation of reddish brown precipitate to show a positive result for the presence of carbohydrate.

Fehling's Test :- Filtrates were hydrolysed with dil. HCl, neutralized with alkali and heated with Fehling's A & B solutions. Formation of red precipitate indicates the presence of reducing sugars.

Molisch's Test :- Filtrates were treated with 2 drops of alcoholic α-naphthol solution in a test tube. Formation of the violet ring at the junction indicates the presence of Carbohydrates.

***D. Test for Alkaloids***

Wagner's Test :- A fraction of extract was treated with 3-5drops of Wagner's reagent [1.27g of iodine and 2g of potassium iodide in 100ml of water] and observed for the formation of reddish brown precipitate (or colouration) which indicates the presence of alkaloids.

Mayer's Test:-Filtrates were treated with Mayer's reagent (Potassium Mercuric Iodide). Formation of a yellow coloured precipitate indicates the presence of alkaloids.

***E. Test for Saponins***

Foam Test :- Test solution was mixed with water and shaken and observed for the formation of froth, which should be stable for 15 minutes. This result indicates the presence of Saponins.

Froth Test:- Extracts were diluted with distilled water to 20ml and this was shaken in a graduated cylinder for 15

minutes. Formation of 1 cm layer of foam indicates the presence of saponins.

***F. Test for Phytosterols***

Salkowski's Test :- Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of Conc. Sulphuric acid, shaken and allowed to stand. Appearance of golden yellow colour indicates the presence of triterpenes.

***G. Test for Glycosides***

Liebermann's test :- Crude extract was mixed with each of 2ml of chloroform and 2ml of acetic acid. The mixture was cooled in ice. Carefully concentrated H2SO4 was added. A colour change from violet to blue to green indicated the presence of steroidal nucleus, i.e., glycine portion of glycoside.

Salkowski's test :- Crude extract was mixed with 2ml of chloroform. Then 2ml of concentrated H2SO4 was added carefully and shaken gently. A reddish brown colour indicated the presence of steroidal ring, i.e., glycone portion of the glycoside.

Keller Killiani Test :- Test solution was treated with few drops of glacial acetic acid and Ferric chloride solution and mixed. Concentrated sulphuric acid was added, and observed for the formation of two layers. Lower reddish brown layer and upper acetic acid layer which turns bluish green would indicate a positive test for glycosides.

***H. Test for Phenols***

Ferric Chloride Test:- Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenols.

***I. Test for Tannins***

Gelatin Test :- To the extract, 1% gelatin solution containing sodium chloride was added. Formation of white precipitate indicates the presence of tannins.

Braymer's test :- 2ml of extract was treated with 10% alcoholic ferric chloride solution. Formation of blue or greenish colour solution shows the presence of Tannins.

***J. Test for Flavonoids***

Shinoda test :- Crude extract was mixed with few fragments of magnesium ribbon. Con. HCl was added drop wise. Pink scarlet colour appeared after few minutes which indicated the presence of flavonoids.

Alkaline reagent test :- Crude extract was mixed with 2ml of 2% solution of NaOH. An intense yellow colour was formed which turned colourless on addition of few drops of diluted acid which indicated the presence of flavonoids.

***K. Test for Steroids***

Liebermann Burchard test :- Crude extract was mixed with few drops of acetic anhydride, boiled and cooled. Concentrated sulphuric acid was then added from the sides of the test tube and observed for the formation of a brown ring at the junction of two layers. Green coloration of the upper layer indicate a positive test for steroids.

***L. Test for Vitamin C***

DNPH Test :- Crude extract was treated with Dinitrophenyl hydrazine dissolved in Con. H2SO4. The formation of yellow precipitate would suggest the presence of Vitamin C.

***M. Test for Phlobatannins***

Precipitate test :- Deposition of a red precipitate when 2mls of extract was boiled with 1ml of 1% aqueous hydrochloric acid was taken as evidence for the presence of phlobatannins.

***N. Test for Quinones***

HCl Test :- A small amount of extract was treated with concentrated HCL and observed for the formation of yellow precipitate (or coloration).

***O. Test for Oxalate***

Acid Test :- To 3ml portion of extracts were added a few drops of ethanoic acid glacial. A greenish black coloration indicates presence of oxalates.

## Results and Discussion

Results obtained for qualitative screening of phytochemicals in different parts of *Sida cordifolia*, L. is presented in Table 1. Of the 15 phytochemicals, 10 were found in various solvent extracts. Phytochemical screening showed presence of proteins, amino acids and carbohydrates in distilled water, alcohol, chloroform, petroleum ether and benzene extract and all these are absent only in acetone extract. Alkaloids present only in petroleum ether extract and was absent in other extracts. Phenols present in all the five extracts except chloroform extract. Tannins present in distilled water, acetone and alcohol extracts. Flavonoids are present in distilled water, alcohol and chloroform extracts and is absent in all other extracts. Steroids shows presence only in alcohol and chloroform extracts. Vitamin C is absent in five out of the six extracts of *Sida cordifolia*, L. and present in extract prepared using distilled water. Phytochemicals such as phytosterols, glycosides, phlobatannins, quinones and oxalate were absent in all extracts. Among the extracts tested maximum result was observed in water and alcohol extract of *Sida cordifolia*, L. followed by chloroform, petroleum ether and benzene. Acetone extract of *Sida cordifolia*, L. shows least result. It shows only the presence of phenols and tannins.

According to Tiwari et al., the factors affecting the choice of solvent are; quantity of phytochemicals to be extracted, rate of extraction, diversity of different compounds extracted, diversity of inhibitory compounds extracted, ease of subsequent handling of the extracts, toxicity of the solvent in the bioassay process, potential health hazard of the extractant. The logic in using different solvents when

**Table 1. Result of phytochemical screening of *Sida cordifolia*, L.**

| Phytochemicals | Extract | | | | | |
|---|---|---|---|---|---|---|
| | Distilled Water | Acetone | Alcohol | Chloroform | Petroleum ether | Benzene |
| Proteins | | | | | | |
| 1. Biuret Test | + | – | + | + | + | + |
| 2. Xanthoproteic Test | + | – | + | + | + | + |
| 3. Millon's Test | + | – | + | + | + | + |
| Amino Acids | | | | | | |
| 1. Ninhydrin Test | + | – | + | + | + | + |
| Carbohydrates | | | | | | |
| 1. Benedict's test | + | – | + | + | + | + |
| 2. Molisch's Test | + | – | + | + | + | + |
| 3. Fehling's Test | + | – | + | + | + | + |
| Alkaloids | | | | | | |
| 1. Wagner's Test | – | – | – | – | + | – |
| 2. Mayer's Test | – | – | – | – | + | – |
| Saponins | | | | | | |
| 1. Foam Test | + | – | – | – | – | + |
| 2. Froth Test | + | – | – | – | – | + |
| Phytosterols | | | | | | |
| 1. Salkowski's Test | – | – | – | – | – | – |
| Glycosides | | | | | | |
| 1. Liebermann's test | – | – | – | – | – | – |
| 2. Salkowski's test | – | – | – | – | – | – |
| 3. Keller Killiani Test | – | – | – | – | – | – |
| Phenols | | | | | | |
| 1. Ferric Chloride Test | + | + | + | – | + | + |
| Tannins | | | | | | |
| 1. Gelatin Test | + | + | + | – | – | – |
| 2. Braymer's test | + | + | + | – | – | – |
| Flavonoids | | | | | | |
| 1. Shinoda test | + | – | + | + | – | – |
| 2. Alkaline reagent test | + | – | + | + | – | – |
| Steroids | | | | | | |
| 1. Liebermann Burchard test | – | – | + | + | – | – |
| Vitamin C | | | | | | |
| 1. DNPH Test | + | – | – | – | – | – |
| Phlobatannins | | | | | | |
| 1. Precipitate test | – | – | – | – | – | – |
| Qui nones | | | | | | |
| 1. HCl Test | – | – | – | – | – | – |
| Oxalate | | | | | | |
| 1. Acid Test | – | – | – | – | – | – |

**+ indicates presence and - indicates absence**

screening for phytochemicals in plant materials was clearly validated in present study. For instance, the results shows that alkaloids were exceptionally present in petroleum ether and benzene extracts but absent in all other extracts. Steroids showed their presence in alcohol extract. This corroborates the reports of Misra et al. Proteins and carbohydrates showed their presence in all extracts irrespective to the solvents and plant parts.

Phytochemical screening of the extracts of *Sida cordifolia*, L. revealed the presence of alkaloids, steroids, flavonoids, aminoacids and phenols (table1). These compounds have significant application against human pathogens, including those that cause enteric infections (El-Mahmood et al.). *Sida cordifolia*, L. hold promises as source of pharmaceutically important phytochemicals. Alkaloids which play some metabolic role and control development in living system is also present in petroleum ether and benzene extracts. They are also involved in protective function in animals and are used as medicine especially the steroidal alkaloids. Tannins are known to inhibit pathogenic fungi, is present in distilled water, acetone and chloroform extracts. The flavonoids and phenolic compounds in plant have been reported to exert multiple biological effects including antioxidant, free radical scavenging abilities, anti-inflammatory, anti-carcinogenic etc.

## Conclusion

It is very necessary to introduce new and biologically safe and active drugs for an eco-friendly life style. Phytochemicals found present in the *Sida cordifolia*, L. indicates their potential as a source of principles that may supply novel medicines. However, further studies are required in this direction for its comprehensive analysis including qualitative or semi qualitative analysis, characterize its chemical structure and assess its biological activities.

Exploration of maximum potential of this valuable plant species is necessary, in medicinal field and pharmaceutical sciences, for their appropriate application.

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