RESEARCH ARTICLE OPEN ACCESS

A Study on Phytochemicals, Antioxidant, Antidiabetic and Antimicrobial Activity of the Leaves of Solanum Trilobatum

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Abstract:

S. trilobatum belongs to the family Solanaceae with genus *Solanum* native to India and is found everywhere in Tamil Nadu. It is widely used as an Indian alternative system of medicines like siddha, ayurveda, herbal medicines and natural home remedy for various conditions like stomach pain, asthma, respiratory problems, cough and cold etc., and is also used for various recipes. It is commonly called as Purple fruited pea egg plant or Thai nightshade and in Tamil as '**Thuduvalai**'. This herb is a thorny creeper that grows in bushes. The entire stem and leaves contain thorns all over the plant. The flowers are purple in colour. It can be propagated by seeds. Medicinal plants can serve as a source of novel therapeutic agent due to the presence of diverse bioactive compounds like alkaloids, flavonoids, terpenoids, phenolic compounds, glycosides etc in plants. These phytocompounds are synthesized by primary or rather secondary metabolism of living organisms. They are widely used in the human therapy, veterinary, agriculture, scientific research, etc. The utilization of several medicinal plants as medicine lies in the fact that they contain various phytoconstituents of therapeutic value. The present study is aimed at analyzing the Phytochemical, Antioxidant, Antidiabetic and Antimicrobial activity of the leaves of *Solanum trilobatum.*

INTRODUCTION

S. trilobatum is an herb which has medicinal properties. The plant is said to contain natural steroids. The steroid **SOLASOLINE** is present in the leaves, fruits, seeds and stem which are widely used for steroid drug production (**ANM Mamun *et. al.,* 2014**). Researches of medicinal plants have shown that the purple fruited egg pea plant is containing hepato protective (protection of liver against radiation and UV caused damages) and also anti mitotic properties. International study of research of cancer enumerated that the plant *Solanum trilobatum* can be administered to treat lung cancer (**Dushyant Kumar Sharma *et. al.,* 2015**). It is said to be a rejuvenatory herb. It treats smartly on various respiratory diseases and asthma. The leaves is used to treat dullness in hearing by making ear drops (**Akilan *et. al.,* 2014**). The flower is used to treat with rheumatism, constipation and other gastric problems. In Siddha treatment, the leaves are used to produce blood and prevent thickening of the blood which leads to many serious problems **M. Pratheeba *et. al.,* 2014**). The leaves are said to be used for the treatment of tuberculosis and

long standing cold. It prevents throat infection. **(Doss *et al*., 2009; P. Swapna Latha** and **K. Kannabiran, 2006)** (**Figure 1**).

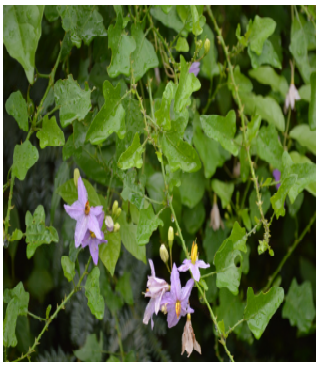

Figure 1: *Solanum trilobatum* plant

The plant is easily available and it is cheap and has many medicinal properties and curing efficiency (**M. Pratheeba *et. al.,* 2014**). The leaves are bitter in taste and are used to cure throat infection, cold, cough, headaches, flu and sneezing. It prevents cancers like oral, uterus and throat cancers because of its anti tumor activity (**Priya G** and **Chellaram C, 2014**). It gives strength to bones as it is rich in calcium. It boosts memory and energy. It improves men's fertility and vitality. It improves blood circulation. It helps to control diabetes. It cures dullness of hearing (**Nirmala Devi Nataraj *et. al.,* 2014**). It is good for gastritis, nerves, asthma, eosinophilia, tuberculosis, difficulty in breathing, constipation, rheumatism, lung disorders and all digestive and respiratory problems. Various dishes like juice, kasayam, soup, chutney, rasam, kuttu, kulambu, etc from the leaves can be made (**G. Priya** and **C. Chellaram, 2014).** The present study is aimed at analyzing the Phytochemical, Antioxidant, Antidiabetic and Antimicrobial activity of the leaves of *Solanum trilobatum.*

MATERIALS AND METHODOLOGY

Collection of sample

The mature and the young leaves of *S. trlobatum* were collected from Guduvanchery, Tamil Nadu. The fresh leaves of these species were collected, washed under running tap water to remove dust and other foreign matter and allowed to shade dry at room temperature for 10 – 15days for long term storage purpose. The shade dried leaves were then powdered using a mechanical grinder.

Preparation of extract

10g of the powder was extracted with different organic solvents viz, Chloroform, ethanol, and water and placed on a mechanical shaker for overnight. The powdered samples were initially air dried and then extracted with solvents. The extracts were filtered through Whatman No.1 filter paper to remove all unextractable matter, including cellular materials and other constitutions that are insoluble in the solvent extraction. The entire extract was concentrated to crude extract and was collected in amber colored sample bottles and was stored.

Biochemical assay for Phytochemical Screening – Qualitative Analysis

Preliminary Qualitative Phytochemical screening of various alcoholic and aqueous extracts of *S. trlobatum* was carried out by following standard procedures to identify the secondary metabolites present in them.

Biochemical assay for Phytochemical screening – Quantitative Analysis

Quantitative analyzes of alkaloids, flavanoids, tannins, saponins, terpenoids, proteins, amino acids and cardiac glycosides were carried out for the plant leaves of *S. trlobatum.*

ANTIMICROBIAL ASSAY OF PLANT EXTRACTS

The ethanolic extracts of *S. trlobatum* was individually tested against a panel of microorganisms, including four bacteria, *Escherichia coli, Bacillus subtilis, Staphylococcus aureus* and *Pseudomonas aeruginosa* and three pathogenic fungi, *Aspergillus niger, Candida albicans* and *Penicillium spp*. The pure bacterial and fungal strains were obtained commercially. Bacterial strains were cultured overnight at 37 °C in Muller Hinton Agar while fungal strains were cultured for three days at 28 °C in Sabouraud Dextrose Agar.

Besides this Antibacterial and Antifungal Activity, Minimum Inhibitory Concentration, Antioxidant Activity by DPPH Method and Antidiabetic Activity were carried out.

RESULTS AND DISCUSSION

QUALITATIVE PHYTOCHEMICAL SCREENING

Out of three extracts (ethanol, chloroform and aqueous) ethanolic extract demonstrated the maximum occurrence of phytoconstituents (10/15) such as flavonoids, tannins, saponins, glycosides, terpenoids, alkaloids, fixed oil, protein, amino acids and carotenoids while carbohydrates, sterols, phenols, quinones and anthroquinones were observed absent in *S. trilobatum.* In case of chloroform extract of *S. trilobatum,* alkaloid, phenol, tannins, carotenoids and fixed oils were present and carbohydrates, saponin, proteins, terpenoids, sterols, glycosids, amino acids, flavanoids quinones and anthroquinones were absent. In aqueous extract of *S. trilobatum,* presence of alkaloids, flavonoids, saponins, sterols, glycosides, protein, amino acid, terpenoids, quinones and absence of tannins, anthraquinone, fixed oils, carotenoid, phenols and carbohydrates were observed **(Table 1).** The presence and absence of the phytoconstituents depends on the solvent medium used for extraction and the physiological property of individual taxa.

S. No	Qualitative Phytochemical Test	Ethanol extract	Chloroform extract	Aqueous extract
01.	Alkaloids	+	+	+
02.	Carbohydrates	-	-	-
03.	Saponin	+	-	+
04.	Fixed Oils	+	+	-
05.	Protein	+	-	+
06.	Amino Acid	+	-	+
07.	Terpenoids	+	-	+
08.	Carotenoids	+	+	-
09.	Sterols	-	-	+
10.	Glycosides	+	-	+
11.	Flavanoids	+	-	+
12.	Phenols	-	+	-
13.	Tannins	+	+	-
14.	Quinones	-	-	+
15.	Anthroquinones	-	-	-

Table 1: Phytochemical analysis conducted on different extracts of *S. trilobatum*

QUANTITATIVE PHYTOCHEMICAL SCREENING

The quantitative estimation of the phytochemicals which showed better results in all the three extracts were quantitatively analysed and their concentration is estimated using standard methods. The quantitative estimation of some major phytochemicals were carried out in the leaf powder of *S. trilobatum* **(Table. 2)**. The quantitative estimation was carried out for phytochemicals like alkaloid, flavanoid, saponin, tannin, terpenoid, cardiac glycosides, proteins

and amino acids. The estimation revealed that *S. trilobatum* had 7.0% of alkaloid, 10.9% of flavanoid, 25% of glycosides, 3% of saponin, 2% of terpenoid, 40% of protein, 10% of amino acid and 1.14mg GAE/g extract.

S. No	Phytochemicals	*S. trilobatum* % in 10g
01.	Alkaloid	7.0
02.	Flavanoid	18.5
03.	Cardiac Glycosides	25
04.	Terpenoids	2
05.	Saponins	3
06.	Protein	40
07.	Amino Acid	10
08.	Tannin	1.14*

Table 2: Quantitative Estimation of Phytochemicals in *S. trilobatum*

*Unit - mg GAE/g extract

ANTIMICROBIAL ACTIVITY

The Antimicrobial activity of the ethanolic extracts and different fractions from the leaves of *S. trilobatum* against a panel of food-borne and pathogenic microorganisms were assessed. The results are presented in **Table 3**. The results from the zone inhibition method, followed by measurement of minimum inhibitory concentration (MIC), indicated that ethanolic extracts of concentrations 50, 100, 150 and 200 µL showed good activity against the test organisms. In case of Antibacterial activity, the extracts was checked for its efficiency against four bacterial species among which to were gram positive - *Bacillus subtilis , Staphylococcus aureus* and two were gram negative - *Escherichia coli,* and *Pseudomonas aeruginosa.* The extracts showed least action towards *Pseudomonas aeruginosa* (10 – 16mm) and for other species of bacteria, the zones were very prominent (10 – 26mm) inhibiting the bacterial growth **(Figures 2 - 5)**.

S. No	Test Organisms	Zone of Inhibition (mm)			
		50µL	100µL	150µL	200µL
01.	*Bacillus subtilis*	12	14	18	22
02.	*Escherichia coli*	20	22	24	26
03.	*Pseudomonas aeruginosa*	12	14	15	18

04.	*Staphylococcus aureus*	14	15	17	18

Table 3 : Antibacterial Activity by Zone Inhibition Method

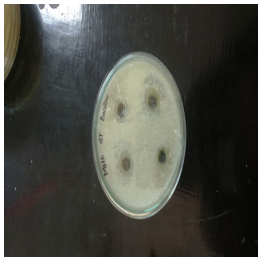

Figure 2: Antibacterial Activity against *B. subtilis*

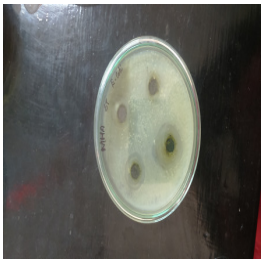

Figure 3: Antibacterial Activity against *E. coli*

Figure 4: Antibacterial Activity against *S. aureus*

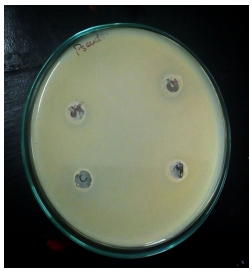

Figure 5: Antibacterial Activity against *Ps. aeruginosa*

The Antifungal activity of the ethanolic extracts and different fractions from the leaves of *S. trilobatum* was tested against three species of fungi **-** *Aspergillus niger*, *Candida albicans* and

Penicillium spp. Both the extracts showed good results by visible zones inhibiting fungal growth **(Table 4 and Figures 6 - 8).**

S. No	Test Organisms	Zone of Inhibition (mm)			
		50µL	100µL	150µL	200µL
01.	*Aspergillus niger*	20	22	24	26
02.	*Candida albicans*	20	22	23	24
03.	*Penicillium spp*	10	11	13	15

Table 4 : Antifungal Activity by Zone Inhibition Method

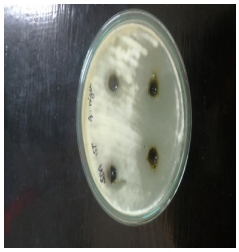

Figure 6: Antifungal activity against *A. niger*

Figure 7: Antifungal activity against *C. albicans*

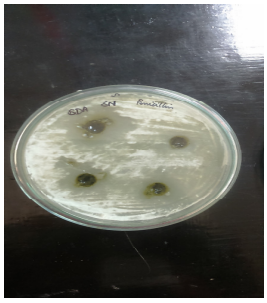

Figure 8: Antifungal activity against *Penicillium spp*

MINIMUM INHIBITORY CONCENTRATION

The MIC in this study against test organisms ranged between 0.4 and 1.5mg/ml for bacteria and while for fungi MIC ranged between 0.8 and 2mg/ml. Antimicrobial agents with low activity against an organism had a high MIC while a highly active antimicrobial agent gave a low MIC. The results of the present study support the traditional use of *S. trilobatum* as a green medicine.

ANTIOXIDANT ACTIVITY

The redox properties of antioxidants play an important role in absorbing and neutralizing free radicals, quenching singlet and triplet oxygen or decomposing peroxides. In doing so, the antioxidants themselves become oxidised. This urges the constant need of antioxidants to replenish them. The antioxidant properties of *S. trilobatum* was evaluated by DPPH assay. The aqueous extract was taken in different concentrations varying between 10 and 60µgmL and results showed that the antioxidant activity, the percentage of inhibition was 40% when analysed (Fig. 11). The results are tabulated below **(Table 5 and Figures 9 - 10).**

S. No.	Concentration(µg/ml)	% of Inhibition of *Solanum trilobatum*
1	10	4.95
2	20	9.63
3	30	15.63
4	40	19.23
5	50	24.75
6	60	40.09

Table 5 : Antioxidant Activity using DPPH Assay

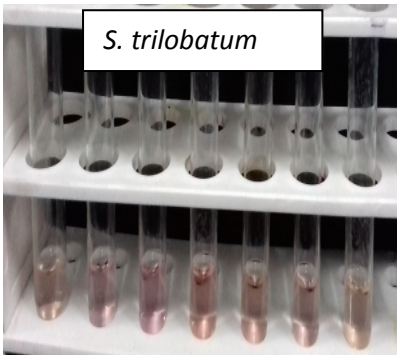

Figure 9: Antioxidant activity by DPPH assay

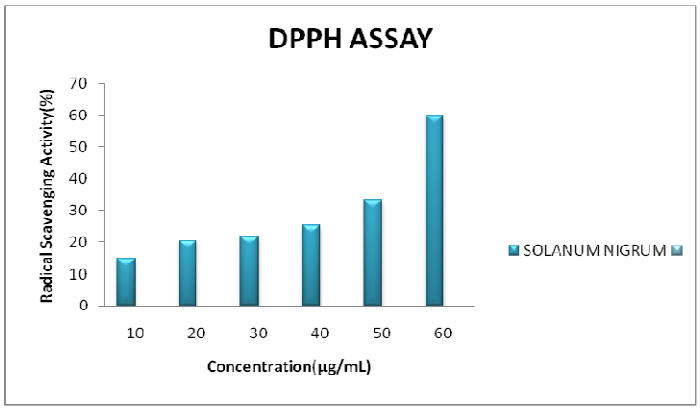

Figure 10: Graph showing Antioxidant Activity

ANTI-DIABETIC ACTIVITY

Diabetes being a metabolic disorder occurs when the blood glucose is too high which is the main source of energy. Insulin, a hormone plays a vital role in aiding glucose from the food to reach the cells to be used as energy. When the body doesnot make insulin or in inadequate amount then glucose stays in the blood causing hyperglycemia. Hence, to avoid or prevent this condition, these plant products may be used. When the extracts of these plants were analysed for anti-diabetic activity of *S. trilobatum* showed 24 – 57% activity **(Table 6 & Figures 11 - 12).**

S. No	Test Samples	Concentration (µg/mL)			
		100	200	300	400
1.	*S. trilobatum*	24%	31.6%	46.8%	57.6%

Table 6 : Anti-diabetic activity - % of Inhibition

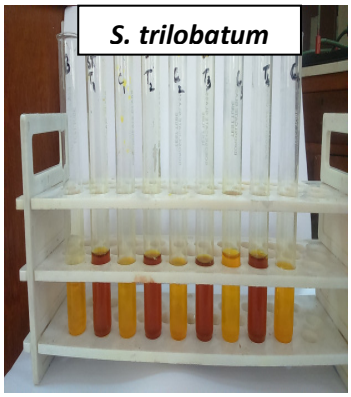

Figure 11: Anti-diabetic Activity

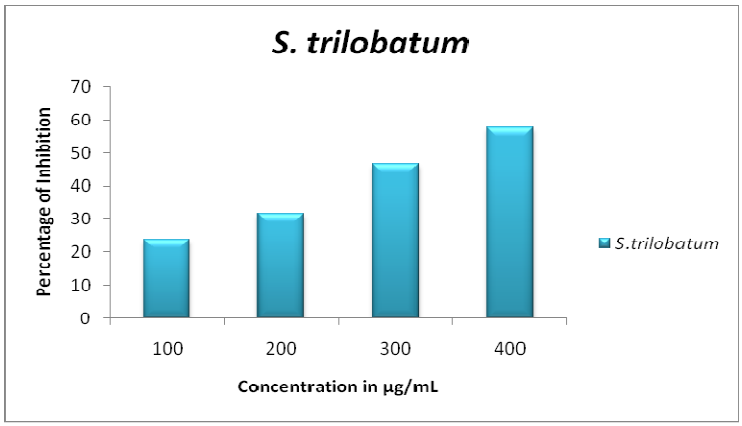

Figure 12: Graph showing Anti – Diabetic Activity

CONCLUSION

This study was completely worked on the plant *S. trilobatum* to check the activity in terms of Antimicrobial, Antioxidant and Antidiabetic. The physio-chemical evaluation of drugs is an important parameter in detecting adulteration or improper handling of drugs. It is important to assess the quality of the plant material to suggest it as drug for application. So, plant extracts containing alkaloids, flavanoids, phenolic, proteins, amino acid, sterols, fixed oil, carotenoid, carbohydrate, terpenoids, tannin, cardiac glycosides, saponin quinones and anthroquinones were all analysed qualitatively and the quantitative tests includes alkaloids, flavanoids, tannins, terpenoids, saponins, proteins, cardiac glycosides and amino acid. These compounds are considered as potent bioactive compounds that could be used for therapeutic purpose or which are precursors for the synthesis of useful drugs as these compounds possess antimicrobial, antiviral, antidiarrhoeal, anticancer, antihelmintic property. The Antimicrobial activity showed very good results in *S. trilobatum* against four bacterial species and three fungal species when the ethanolic extracts (50, 100, 150 and 200µL concentrations) were used. The Antimicrobial activity was analysed using Zone Inhibition method. The bacterial species which were used for analyzing the antibacterial activity was *Bacillus subtilis, Staphylococcus aureus* and *Escherichia coli,* and *Pseudomonas aeruginosa.* The antifungal activity was tested against *Aspergillus niger*, *Candida albicans* and *Penicillium spp.* The Minimum Inhibitory Concentration was carried for the same species of bacteria and fungi as mentioned earlier. The Antioxidant activity was analysed using DPPH method using aqueous extracts in concentrations 10 - 60µg/mL which showed 40% activity in *S. trilobatum.* Finally, the Antidiabetic activity was evaluated using the extracts in concentrations 100, 200, 300 and 400mg/mL which showed 61% inhibition *S.*

trilobatum. Thus from the present study carried it is very clear that all the above mentioned work were found to be dose dependent. Hence, to develop a therapeutic drug it is important to find out its exact concentration or dose in which a particular disease can be treated.

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