

EVALUATION OF *IN VITRO* ANTIBACTERIAL ACTIVITY OF WHOLE PLANT (FRUIT, LEAVES, STEM, AND ROOT) OF *SOLANUM TORVUM* SWARTZ.***Dr. Lincy Sara Varghese, *Ambili P., **Dr. V. T. Antony, *Keerthimol Joy and *Mayamol M. S.**

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ABSTRACT

In this study the antibacterial activity of *Solanum torvum* Swartz. was assayed against *E. coli*, *Salmonella typhi*, *Salmonella paratyphi*, *Staphylococcus aureus*, *Sreptococcus* sp., *Bacillus* sp., *Proteus* sp., *Pseudomonas* sp. and *Klebsiella* sp. The antibacterial activity was determined in ethanolic extracts of fruits, stem, leaves and root of *Solanum torvum*. All the test bacteria were resistant to the stem and root extracts except for *S. aureus* and *Bacillus* sp. The leaf extract maximally inhibited the growth of *Sreptococcus* followed by *Bacillus*; and to a less extent of *S. aureus*, *Klebsiella*, *S. typhi*, *S. paratyphi* as well. However, the fruit extract of *S. torvum* was effective in inhibiting the growth of *Klebsiella*, *S. typhi*, *S. paratyphi*, *S. aureus* and *Bacillus*. The bactericidal activities of *S. torvum* extracts were superior in Gram positive than in gram negative bacteria. The present study observed that the inhibitory effect of *S. torvum* is more against Gram positive bacteria. Also, the root of *S. torvum* was most inhibitory to the growth of bacteria than fruit, leaves or stem.

KEYWORDS: *Solanum torvum*, antibacterial assay, ethanol extract, disc diffusion, ethnomedicine.**INTRODUCTION**

The members of the genus *Solanum*, a member of the family Solanaceae, includes plants of countless medicinal potentialities and is widely used in the therapy of tooth ache, skin complaints, urine troubles, sorethroat, cough, asthma, chest pain, rheumatism, cardiac disorders, tumors, and warts. These plants have also been used as diuretic, diaphoretic, sedative, analgesic, antiplasmodic and narcotic drugs.^[1] In spite of the immense medicinal properties they possess *Solanum* plants have been little exploited in the field of ayurvedic medicine, primarily due to the lack of proper knowledge on its therapeutic potentialities.

Solanum torvum Swartz., commonly known as Sundakai, is a prickly tomentose erect shrub distributed widely in Pakistan, India, Malaya, China, Philippines and Tropical America.^[2] The plant reaches to a height of about 12 feet and is commonly found in hills, waste places and along the road sides.^[3] It is locally known as tit begoon, gota begoon or hat begoon in Bengali and commonly known as turkey berry, susumber, gully-bean, thai eggplant or devil's fig.^[4] Their leaves have no prickles and the flowers are bell shaped. They have lobed fruits which are seated on the calyx. The fruits of this plant are edible, often eaten in their daily diet as vegetables by the common people and tribals of Bangladesh. It is also used as essential ingredient in Thai and Indian cuisine.^[5,6]

Various parts of the plant have been employed as sedative, diuretic and digestive. They have also been used in the treatment of coughs and colds.^[7] The tribals use the leaves, young shoots and berries of this plant as food. It has also been used in ethnomedicine as a tonic and haemopoetic agent and for relief from pain.^[8, 9, 10] The leaf extract of the plant has been reported to possess antibacterial, antiviral and anti-ulcerogenic properties^[11, 12, 13, 14]. Root paste has been used to cure cracks of feet and the fumes of seeds inhaled for toothache.^[15] *S. torvum* has been reported to contain many phytochemicals which include alkaloids, saponins, sapogenins, flavonoids and glycosides.^[11,16,17,18] Sivapriya and Srinivas has reported the presence of antioxidants in the seeds of *S. torvum*.^[19] The present study aimed at investigating the antibacterial activity of fruit, stem, leaves and root extract of *S. torvum* against common Gram positive and Gram negative bacteria of importance to humans.

MATERIALS AND METHODS**Collection of Sample**

Fruit, stem, root and leaves of *S. torvum* were collected from Thiruvalla, Pathanamthitta district, Kerala, India.

Bacterial Stains Used

Bacterial cultures used in this study were obtained from the culture collections of School of Biosciences, Mahatma Gandhi University, Kottayam, Kerala, India.

For the study nine bacterial cultures namely *Staphylococcus aureus*, *E. coli*, *Klebsiella*, *Salmonella typhi*, *Salmonella paratyphi*, *Streptococcus*, *Bacillus*, *Pseudomonas* and *Proteus* were used. The bacterial strains were maintained on Nutrient Agar (HiMedia, India) plates or slants and were stored at 4 °C before use.

Surface Cleaning and Sterilization of the Samples

In this study the various samples were surface sterilized following the modified procedure of Aneja.^[20] The samples namely fruit, stem, root, and leaves of *S. torvum* were washed in running tap water for 10 minutes followed by detergent wash in 10 % Extran (Merck, India) for 10 minutes. The samples were then rinsed with distilled water and cut into small pieces in aseptic condition. The cut pieces were rinsed in 70 % ethanol for 30 seconds and washed again in distilled water till the ethanol smell completely diminished. These were spread out in clean trays for oven drying.

Preparation of Extracts

A comparative assay of extracts of fruit, stem, root and leaves of *S. torvum* was carried out in this study. The cleaned and cut samples of *S. torvum* were oven dried at 60 °C, continuously, for 7 days. The dried samples were powdered using a clean grinder. The powder was stored in air sealed containers at room temperature before extraction. A fixed weight of 30 gm of each powdered material was weighed out, respectively, in aseptic condition and was extracted with ethanol using the Soxhlet Apparatus at a temperature of 60 °C. The Soxhlet extraction was carried out continuously for 8 hrs. Each extract was concentrated by evaporation and made up to a final volume of 10 ml. The extracts were stored at room temperature, in sterile screw capped containers, till use.

Determination of Antimicrobial Activity

Preparation of Bacterial Suspension

Pure isolated colonies of the test bacteria were inoculated into 1 % peptone water and incubated at 37 °C for 48 h and were used as inoculum for lawn culture on Mueller Hinton Agar (HiMedia, India).

Sensitivity Discs

Sensitivity discs of 6 mm diameter were prepared from Whatman No. 1 filter paper. The discs were sterilized by autoclaving and stored at room temperature till use. The discs were soaked in the extracts for 10 minutes and were used for disc diffusion assay.

Disc Diffusion Method

Mueller-Hinton Agar (MHA) was used as the base medium for screening of antibacterial activity. Pure cultures of the test bacteria were used as inoculum on MHA. Using sterile cotton swab, 0.2 ml of 24 hr old culture was inoculated evenly on to the surface of MHA to make a lawn culture. For analyzing the antibacterial activity the discs carrying the extracts were impregnated on the seeded agar plate (2 discs per plate). Discs

carrying ethanol were used as controls. The experiment was performed in duplicates. The plates were incubated at 37 °C for 24 hrs and observed for zone of inhibition of growth around the discs.

Zone Analysis

The plates were checked for zone of clearance around the discs. The antibacterial activity of the extracts was assayed by measuring the diameter of zone of inhibition around the discs to the nearest mm.

RESULTS AND DISCUSSION

S. torvum Swartz. is a plant of great medicinal importance and is also valued high in the field of ethnomedicine. The plant is an important source of many pharmacologically active chemicals such as steroids, glycosides, sitosterols, stigmaterols and capesterol. It has variety of effects like antihypertensive, antioxidant, cardiovascular, antibacterial, antifungal and antiviral activities.^[21] It has been used in traditional medicine in the treatment of diseases like coughs, lung diseases, tooth problems and cracked foot. Chopra et al have reported the use of *S. torvum* based drugs in the treatment of cough and as sedative, diuretic and digestive tonic.^[22] In the present study, the antibacterial activity of ethanol extract of *S. torvum* was assayed against six Gram negative and three Gram positive bacteria. All the test bacteria exhibited resistance to the stem and root extract except for *S. aureus* and *Bacillus* (Table 1).

The root extract was greatly inhibiting the growth of *S. aureus* and *Bacillus* with zone of inhibition of 20.5 and 14 mm, respectively. Bari et al has also obtained concordant results for *Bacillus* strains such as *B. cereus* and *B. subtilis*.^[4] Unlike in our study Bari et al has observed clear zone of inhibition of *S. typhi*, *Shigella dysenteriae* and *Streptococcus β-haemolyticus*.^[4] However, the root extract in the present study was not inhibitory to the growth of *S. typhi* and *Streptococcus* sp.

In our study the leaf extract potentially inhibited the growth of *Streptococcus* and *Bacillus* with a zone of growth inhibition of 12 and 10.5 mm respectively, but also inhibited the growth of *Klebsiella*, *S. typhi*, *S. paratyphi* and *S. aureus* though to a limited extent (Table 1). Bari et al has also reported the antibacterial activity of chloroform and methanol extract of leaf extract of *S. torvum* against *S. typhi*, but failed to observe antibacterial action against *S. aureus*, *Bacillus* sp., *Streptococcus*, *Shigella dysenteriae*, *Klebsiella*, *Sarcina*, *Pseudomonas* and *Proteus*.^[4] The aqueous, methanol, ethanol and chloroform leaf extracts of *S. torvum* have been reported to possess antibacterial and antifungal activity against plant pathogens such as *Xanthomonas campestris*, *Pyricularia oryzae*, *Alternaria alternate*, *Bipolaris oryzae*, *tricoconis padwickii*, *Dresclera teramera*, *D. halodes*, *Curvularia lunata*, *Fusarium oxysporum*, *F. Moniliforme* and *F. solani*.^[23] The stem extract of this study was more effective against *Bacillus* than *S. aureus* as has been reported by Bari et al also.^[4]

Table 1. Antibacterial assay of ethanol extract of *Solanum torvum* against pathogenic bacteria by disc diffusion method

Bacteria	Average diameter of Zone in mm			
	Fruit	Leaf	Stem	Root
<i>E. coli</i>	R	R	R	R
<i>Klebsiella</i> sp.	9	7	R	R
<i>S. typhi</i>	8	8.5	R	R
<i>S. paratyphi</i>	7	7	R	R
<i>Proteus</i> sp.	R	R	R	R
<i>Pseudomonas</i> sp.	R	R	R	R
<i>S. aureus</i>	7	7	7.5	20.5
<i>Streptococcus</i> sp.	R	12	R	R
<i>Bacillus</i> sp.	9	10.5	10.5	14

Balachandran et al has reported the antimycobacterial activity of methyl caffeate, a compound present in the methanolic extract of fruit of *S. torvum*.^[24] In this study the fruit extract inhibited the growth of *Klebsiella* and *Bacillus* equally, with a zone of growth inhibition of 9 mm each. The fruit extract also inhibited the growth of *S. typhi*, *S. paratyphi* and *S. aureus* considerably. Sivapriya et al also observed effective antibacterial activity of fruit coat of *S. torvum* against *S. aureus*, *Bacillus subtilis*, *Salmonella typhimurium* and *Vibrio cholerae*.^[25] The leaf and fruit extracts of *S. torvum* could not inhibit the growth of *E. coli*, *Proteus*, and *Pseudomonas*. On the contrary, Sivapriya et al has reported effective antibacterial activity of ethanol extract of *S. torvum* fruit coat against *E. coli* and *Pseudomonas*.^[25] In the current study the fruit extract inhibited the growth of *Streptococcus* as well. Chah et al has also reported previously about the wide spectrum antibacterial activity of methanolic extracts of fruits of *S. torvum* against human and animal clinical isolates.^[26] Bari et al has reported the antifungal activity of root and stem extract of *S. torvum* against *Aspergillus fumigatus*, *Candida albicans* and *Vasin factum*.^[4]

CONCLUSIONS

The present study is a preliminary investigation on the antibacterial activity of *S. torvum*. The results of this study show that the bacterial strains viz. *E. coli*, *Proteus* and *Pseudomonas* were resistant to all the four extracts. *S. aureus* and *Bacillus*, on the contrary, were inhibited by all the four extracts, though in varying levels. Both *S. aureus* and *Bacillus* were inhibited to the maximum by the root extract of *S. torvum*. The present study not only demonstrates the potential antibacterial activity of various parts of *S. torvum*, but also proves that the ethanolic extracts of the plant parts are greater inhibitory to Gram positive rather than to Gram negative bacteria. The studies on the antimicrobial profile of *S. torvum* are limited. Further studies of this kind on this plant are hence very essential in identifying the bioactive compounds and also to identify dose wise responses for each of the harmful or pathogenic bacteria. Attempts should also be made to document ethnic uses of the plant so that it could be exploited more effectively by the pharmacologists for the preparation of herbal and modern medicines.

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