

EVALUATION OF ANTIBACTERIAL POTENTIAL OF THE TRADITIONAL MEDICINAL CLIMBER, *SOLENA AMPLEXICAULIS* (LAM.) GANDHI. (CUCURBITACEAE)

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ABSTRACT

Solena amplexicaulis (Cucurbitaceae) is a traditional medicinal climber generally prescribed for wound healing by the local healers in western districts of Tamil Nadu. The aim of the present study was focused on investigating the potential of antibacterial activity via *in vitro* approach. The aqueous and organic solvent extracts (hexane, benzene, chloroform and methanol) of the stem part of *S. amplexicaulis* were tested against 15 human pathogenic bacteria by agar well diffusion method. Results showed promising antibacterial activity against the bacteria tested. Among them, chloroform and benzene extracts were found to have more potent inhibitory effect in comparison to the other extracts. It proves the therapeutic importance of the species in curing infectious diseases and encouraged for its extensive use in health care practices.

Key words: *Solena amplexicaulis*, antibacterial activity, agar well diffusion method.

1. INTRODUCTION

Phytomedicines play a major role in human health care system and a source of great economic value all over the world (Ahmad *et al.*, 1998). These medicinal plants represent a rich source of antimicrobial agents, used medicinally in different countries and as a source of many potent and powerful drugs (Uniyal *et al.*, 2006). These beneficial medicinal effects of plant materials typically result from the combinations of secondary products present in the plant (i.e.) alkaloids, steroids, tannins, phenolics, flavonoids etc which are capable of producing definite physiological action on body (Bishnu *et al.*, 2009). As per the World Health Organization (WHO) reports, 80% of the world populations are presently using herbal medicine for some aspects of primary health care. Many pharmaceutical companies are also showing interest in manufacturing plant derived drug based on their belief of 'Green Medicine' is safe and more dependable than the costly synthetic drugs, which have adverse side effects (Sujatha, 2005).

Solena amplexicaulis (Cucurbitaceae) is a perennial dioecious climber with tuberous root found throughout Asia mainly in hilly dry deciduous forests and scrub jungles. It has been used in traditional Indian medicine for various ailments like spermatorrhoea, thermogenic, appetizer, cardiogenic, diuretic and haemorrhoids (Kritchovsky, 1978) and its leaves have good anti-inflammatory activity and also recommended for skin lesions and other skin diseases (Arun *et al.*, 2011). The leaf juice is taken orally to cure jaundice (Mohammed *et al.*,

2011). The fresh stem is externally used to promote the conception (Ignacimuthu *et al.*, 2008). Unripe fruits are eaten raw to strengthen the body (Jeyaprakash *et al.*, 2011) and used as vegetable also. It helps to increase the secretion of milk during lactation and also used as expectorant, antidepressant and antianxiety and to reduce asthmatic conditions. (Bandyopadhyay and Sobhan Kr Mukherjee, 2009). Root is a stimulant and purgative (<http://hpforest.nic.in/potter.htm>). The decoction of the root is taken orally to cure stomachache (Abdolbaset *et al.*, 2011). The whole plant is determined to be a potential source of natural antioxidant activity (Venkateshwarlu *et al.*, 2011 and Karthika *et al.*, 2012) and also used for the treatment of diabetes (Pullaiah *et al.*, 2003).

Considering the indigenous uses of this species, the present investigation was taken up with an objective to evaluate the antibacterial potential of stem extract against certain human pathogenic bacteria that may provide scientific justification to the traditional uses in treating various ailments.

2. MATERIALS AND METHODS

2.1. Plant material

The stem of *S. amplexicaulis* was collected from Madukkarai, Coimbatore district, Tamil Nadu. Collected plant materials were washed thoroughly in tap water, shade dried and then homogenized to fine powder and stored in air tight bottles.

2.2. Preparation of extracts

About 50g coarsely powdered plant material (50g/250ml) was extracted in the soxhelt extractor for 8 to 10 hours, sequentially with hexane, benzene, chloroform, methanol and water. Then each extract was evaporated to dryness.

2.3. Bacterial strains

In vitro antibacterial activity was examined for the crude extracts of stem of the study plant, against 15 bacterial species which include the Gram positive strains viz., *Streptococcus faecalis*, *S. pyogenes*, *Bacillus subtilis*, *B. thuringiensis*, *Staphylococcus aureus* and *Enterococcus faecalis* and Gram negative strains viz., *Klebsiella pneumoniae*, *Salmonella paratyphi*, *S. paratyphi* A, *S. paratyphi* B, *Escherichia coli*, *Proteus vulgaris*, *P. mirabilis*, *Serratia marcescens* and *Pseudomonas aeruginosa*.

All these bacterial strains were obtained from the Department of Microbiology, Tamil Nadu Agricultural University, Coimbatore. All the bacteria were maintained at 4°C on nutrient agar slants for further use.

2.4. Bacterial susceptibility testing

An inoculum of each of the pathogenic bacterial strains was suspended in 5ml of nutrient broth and incubated at 37°C for 18 hrs (10^6 - 10^8 bacteria CFUml⁻¹). Bioassay was carried out by using agar well diffusion method (Perez *et al.*, 1990; Murray *et al.*, 1995; Olurinola, 1996). Inoculum was spread over Muller – Hinton agar medium with sterile glass spreader. A well of 6 mm diameter was made using a sterile cork borer and filled with 50µl of different extracts by using micropipette in each well in aseptic condition. The plates were kept at room temperature for absorption of extract in the medium and further incubated in an incubator at 37°C for 24 hrs. The antibacterial activity was evaluated by measuring the diameter of inhibition zone (mm). Ampicillin was used as positive control (50µl/ml) and DMSO (Dimethyl sulphoxide) as negative control (50µl).

2.5. Statistical analysis

All the analyses were done in triplicate and results were expressed as mean±SD. The data were subjected to one way analysis of variance (ANOVA) and the significance of the difference between mean was determined by Duncan's Multiple Range Test with significance level, P<0.05. ANOVA was performed using the statistical software SPSS (SPSS Inc. Chicago, USA).

3. RESULTS AND DISCUSSION

Agar well diffusion method is widely used in *in vitro* method of evaluation of antimicrobial activity of various chemicals. The size of the inhibition zone depends on the solubility of the test material, time and temperature of incubation (Weiss *et al.*, 1996). In the present study the antibacterial activity of aqueous and solvent extracts (hexane, benzene, chloroform and methanol) of stem part of *S. amplexicaulis* against certain human pathogenic bacterial which include both gram positive and gram negative in terms of inhibition ability were presented in Table – 1. Among the five extracts, benzene and chloroform extracts showed broad spectrum of antibacterial activity in comparison to hexane and methanol extracts which showed mild activity with the zone of inhibition diameter ranged from 8 - 24mm and 6 - 12mm, respectively. The water extract showed almost no activity. Different solvents have been reported to have different capacity to extract phytoconstituents according to their solubility or polarity, and most of the compounds are dissolved well in alcoholic solvents than in water (Marjorie, 1999). The antibacterial activity was compared with the inhibitory activity of antibiotic, Ampicillin which showed varied inhibitory zones of 13 - 24 mm. The negative control DMSO, which showed no zone of inhibition. Among the 15 bacteria, *Staphylococcus aureus* of Gram positive type showed higher susceptibility to stem extract than the antibiotic, ampicillin. *S. aureus* is a well known wound pathogen (Lullmann *et al.*, 2000) which can be found as the part of the normal skin flora and in the nasal passages (Kluytmans *et al.*, 1997 and Cole *et al.*, 2001). In the study, Gram positive bacteria are little bit sensitive than that of Gram negative bacteria because the latter are frequently reported to have developed multidrug resistance due to their outer membrane which act as a barrier to many environmental substances, including antibiotics (Tortora *et al.*, 2001; Johansson *et al.*, 2011; Johnson *et al.*, 2011; Ramakant *et al.*, 2011). The zone of inhibition \geq 9-15mm is an indication of strong antimicrobial activity (Rani and Khullar, 2004). So, the stem extract of *S. amplexicaulis* having the potential for killing the bacteria. High antibacterial effects of alcoholic extracts of certain Cucurbitaceae members were already reported well *Trichosanthes cucumerine* (Arawwawala *et al.*, 2011), *Citrullus colocynthis* (Gurudeeban *et al.*, 2010) and *Coccinia grandis* (Farrukh *et al.*, 2008)).

5. CONCLUSION

The present research is a right step to the direction of searching novel and more effective antibacterial compounds in plants. In conclusion the species, *S. amplexicaulis* extracts exhibited antibacterial activity against both Gram positive and Gram negative bacterial strains mediating the presence of a broad spectrum of antibacterial compounds. This study suggests that further research will be needed for pharmacological aspects and elucidate the specific phytoactive compounds in the stem extract of *S. amplexicaulis* and hence to go for commercial application through pharmaceutical industries.

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Table 1: *In vitro* antibacterial activity of stem extract of *Solena amplexicaulis* by agar well diff method.

S.No	Name of the bacteria	Diameter of inhibition zone (mm)					
		Control*	Hexane	Benzene	Chloroform	Methanol	Water
Gram positive							
1.	<i>Streptococcus faecalis</i>	24.33±0.15 ^a	-	11.33±0.93 ^b	12.33±0.52 ^b	9.00±0.10 ^c	-
2.	<i>S. pyogenes</i>	20.00±0.81 ^a	6.67±0.53 ^b	11.33±0.16 ^c	13.67±0.23 ^{ac}	8.33±0.51 ^{bc}	-
3.	<i>Bacillus subtilis</i>	23.33±0.21 ^a	6.67±0.58 ^b	14.33±0.63 ^c	16.33±0.72 ^c	8.33±0.89 ^b	6.82±0.29 ^b
4.	<i>B. thuringiensis</i>	18.33±0.58 ^a	8.00±0.65 ^b	8.00±0.20 ^b	12.67±0.44 ^c	7.67±0.53 ^b	-
5.	<i>Staphylococcus aureus</i>	19.67±0.53 ^a	11.33±0.21 ^b	24.33±0.59 ^c	23.67±0.15 ^c	11.33±0.08 ^b	-
6.	<i>Enterococcus faecalis</i>	20.00±0.65 ^a	6.33±0.31 ^b	13.33±0.16 ^c	18.00±0.36 ^{ac}	10.00±0.30 ^{bc}	-
Gram negative							
5.	<i>Klebsiella pneumonia</i>	15.00±0.10 ^a	-	13.00±0.81 ^a	15.61±0.81 ^a	7.67±0.79 ^b	-
6.	<i>Salmonella paratyphi</i>	23.00±0.73 ^a	-	9.67±0.53 ^b	11.33±0.57 ^c	8.00±0.26 ^b	7.67±0.62 ^b
7.	<i>Salmonella paratyphi A</i>	16.00±0.20 ^a	6.67±0.28 ^b	11.00±0.10 ^c	13.67±0.58 ^c	7.33±0.11 ^b	-
8.	<i>S. paratyphi B</i>	13.33±0.58 ^a	8.33±0.15 ^b	13.33±0.15 ^a	17.00±0.20 ^c	10.00±0.20 ^{ab}	-
9.	<i>Escherichia coli</i>	16.33±0.23 ^a	8.00±0.65 ^b	13.67±0.36 ^c	16.00±0.61 ^a	9.00±0.10 ^b	-
10.	<i>Proteus vulgaris</i>	19.00±0.73 ^a	8.33±0.28 ^b	16.67±0.51 ^c	17.67±0.64 ^{ac}	12.33±0.41 ^{bc}	-
11.	<i>P. mirabilis</i>	19.00±0.36 ^a	6.50±0.35 ^b	12.33±0.44 ^c	13.67±0.53 ^c	7.67±0.53 ^b	-
12.	<i>Serratia marcescens</i>	18.00±0.20 ^a	7.67±0.31 ^b	18.00±0.20 ^a	17.00±0.30 ^a	9.33±0.58 ^c	-
13.	<i>Pseudomonas aeruginosa</i>	16.67±0.89 ^a	7.67±0.52 ^b	10.00±0.65 ^c	14.67±0.31 ^{ac}	9.67±0.15 ^{bc}	-

*Amphicillin, '-' indicates no activity.

Values were performed in triplicates and represented as mean±SD.

Mean values followed by different superscript in a column are significantly different (P<0.05).