**Natural Products** 

Volume 7 Issue 6



Trade Science Inc.

An Indian Journal

Full Paper

NPAIJ, 7(6), 2011 [310-314]

## In vitro antibacterial activity of Cadaba fruticosa and Solanum trilobatum on some common pathogenic organisms

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Received: 16th September, 2011; Accepted: 16th October, 2011

## ABSTRACT

Indian traditional medicinal plants are extensively used for healing the illness and most of the people seeking remedies and health approaches free from side effects caused by synthetic chemicals. Recently, considerable attention has been paid to utilize eco-friendly and biofriendly plantbased products for the prevention and cure of different human diseases. The present study deals with the antibacterial activity of various solvents like acetone, aqueous, benzene, butanol, chloroform and ethanol extract of Cadaba fruticosa L. (Capparaceae family) leaves (10mg/ml) and Solanum trilobatum (Solanaceae family) leaves (10mg/ml) were determined using disc diffusion method against some pathogenic organisms like Streptococcus pyogens, Staphylococcus aureus, Salmonella typhi, Proteus vulgaries, Klebsiella pneumoniae, Pseudomonas aeruginosa, Bacillus subtilis, and Escherichia coli. The plants exhibited significant broad-spectrum activity against Bacillus subtilis, Staphylococcus aureus, and Pseudomonas aeruginosa. These results were compared with results obtained using standard antibiotics, Chloramphenicol (30µg/disc) and streptomycin (30µg/disc), which served as reference for inhibition zone diameter. © 2011 Trade Science Inc. - INDIA

#### **INTRODUCTION**

Medicinal plants are very effective sources in both traditional as well as modern medicine even though persists in research of synthetic compounds<sup>[1]</sup>. The substances derived from medicinal plants are potentially less toxic and are free of side effects on the host<sup>[2]</sup>. This has urged the microbiologists all over the world for formulation of new antimicrobial agents and evaluation of

## KEYWORDS

Cadaba fruticosa L.; Pathogenic organisms; Antibiotics; Disc diffusion method.

the efficacy of natural plant products as the substitute for chemical antimicrobial agents. In India, medicinal plants are extensively used by most of the people either in different indigenous systems of medicine or directly as folklore remedies or indirectly in the preparation of pharmaceuticals of modern medicines<sup>[3]</sup>.

*Cadaba fruticosa* L. (Capparaceae family) is a shrub commonly found in tropical countries especially in lower altitude of India. This plant grows up to 3

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metres in height, bearing cylindrical stems. The leaves are ovate-oblong with glabrous and fully margined<sup>[4]</sup>. The leaves and roots are used as deobstruent, anthelmintic, emmenagogue and uterine obstructions. The leaves and fruits are used to treat worm infestation, swellings, eczema and constipation<sup>[5,6]</sup>. The juice of the leaves are especially used to cure gonorrhoea and vermifuge<sup>[7-9]</sup>. The active principles, Stachydrine and 3hydroxystachydrine isolated from the stem and roots of *C. fruticosa*<sup>[10,11]</sup>. The leaves contain cadabine<sup>[10,12]</sup> and terpenoids, flavones, sugar and proteins<sup>[13]</sup>.

Solanum trilobatum (Solanaceae) is a common shrub, called as 'Tuduvelai', used in various diseases distributed over Gujarat, Deccan, Ceylon, North Circars, Carnatic and Malay Peninsula<sup>[14]</sup>. In Indian Ayurveda and Siddha medicinal system, the roots and leaves are bitter and prescribed in consumptive cases of acute and chronic bronchitis<sup>[14,15]</sup>, asthma<sup>[16,17]</sup>, cough<sup>[18]</sup>, and analgesic action<sup>[19]</sup>. The herbs are useful in treating indigestion, spermatorrhoea, tuberculosis and disease of ear<sup>[20]</sup>. Pharmacological investigations have demonstrated that S. trilobatum possess an antibacterial, antifungal & anticancer activity<sup>[21-28]</sup>; antioxidant activity<sup>[29]</sup>; hepatoprotective activity<sup>[25]</sup>; anti-ulcerogenic activity<sup>[30]</sup> and anti-inflammatory activity<sup>[31,32]</sup>. The leaves of the plant possess calcium, iron, phosphorus, fat, carbohydrates, crude fibre and minerals<sup>[33]</sup>. The whole plant contains alkaloids, phenolics, flavanoides, sterols, saponins and their glycosides<sup>[30]</sup>; solasodine and  $\beta$ -solamarine<sup>[34]</sup>. Several studies have been reported, dealing with antimicrobial screening of extracts of medicinal plants<sup>[35-38]</sup>. Hence, an attempt has been made to study the invitro antibacterial activity of Cadaba fruticosa.

## MATERIALS AND METHODS

## **Collection of plant material**

The plant was collected from Kollihills based on the information provided in the ethanobotanical survey conducted in 1995-97. The plant was authenticated by comparison with reference specimens preserved at the Rapinat Herbarium, St. Joseph's College, Tiruchirapalli. Voucher specimens are kept in the Herbarium for future references.

## **Extraction of plant material**

The plant material was shade dried at 30°C for 15

days; 100g (1:6 w/v) sample preparation was powdered by using a waring blender. Then it was loaded into an empty glass column. A total of 600ml of various polar and non-polar solvents namely acetone, aqueous, benzene, butanol, chloroform and ethanol were passed through the column. The compounds were leached out based on their polarity and collected. The left over residue solvents were completely evaporated by using vacuum rotary evaporator. The final weight of the various crude extracts were weighed and prepared the concentration.

#### Microorganisms

Salmonella typhi, Proteus vulgaries, Pseudomonas aeruginosa, Escherichia coli, Klebsiella pneumoniae (Gram negative bacteria), Bacillus subtilis, Streptococcus pyogenes, Staphylococcus aureus (Gram positive bacteria), four different laboratory bacterial strains were used as test organisms. The bacteria were incubated on a nutrient agar slant (Stationary culture) for 48h at 37°C followed by inoculation in Muller Hinton agar medium. The bacteria were supplied by the Department of Microbiology, Selvamm Arts and Science College, Namakkal, Tamilnadu.

## Antibacterial assay

Antibacterial activity was demonstrated using a modification of the method originally described by Bauer et al.<sup>[39]</sup> which is used for the antibacterial susceptibility testing<sup>[40]</sup>. A loopful bacteria was taken from the sock culture and dissolved in 0.1ml of saline. All the tests were done by placing the disc (5mm diameter) impregnated with various crude solvent extract on the Muller Hinton Agar (MHA) surface previously inoculated with 10ml of (MHA) liquid medium with gram positive and gram negative bacteria. Respective solvents without plant extract served as negative control. Standard antibiotics of chloramphenicol (30µg/disc) and streptomycin (30µg/disc) were used as reference. Plates were incubated at 37°C for 24 hours to observe formation of clearing zone around the disc.

## RESULTS

TABLE 1 and 2 illustrates that the data pertaining to the evaluation of antimicrobial activity of *Cadaba* 

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*fruticosa* and *Solanum trilobatum* on different tested microorganisms. The maximum inhibition zone diameters were observed from ethanolic leaves extract of both medicinal plants obtained from *Escherichia coli* (23mm, +++), *Bacillus subtilis* (23mm, +++) and *Klebsiella pneumoniae* (23mm, +++). These results were compared with antibiotic drugs chloramphenicol and streptomycin in TABLE 3 as a standard reference.

All the solvents of *Cadaba fruticosa* exhibited the maximum zone of inhibition for *Streptococcus pyogens* (20, ++). The acetone extract of *Cadaba fruticosa* showed the maximum zone of inhibition for all tested microorganisms (20, ++). The aqueous extract of the

 TABLE 1 : Antibacterial activity of various solvents crude

 extracts of Cadaba fruticosa L

| Extraction | Disc diffusion method; size of inhibition<br>zones (9mm); bacteria <sup>a</sup> |    |     |    |     |    |     |      |  |
|------------|---|----|-----|----|-----|----|-----|------|--|
|            | Ι   | II | III | IV | V   | VI | VII | VIII |  |
| Acetone    | ++  | ++ | ++  | ++ | ++  | ++ | ++  | ++   |  |
| Aqueous    | ++  | ++ | +   | ++ | +   | ++ | +   | +    |  |
| Benzene    | ++  | +  | -   | -  | -   | -  | +   | +    |  |
| Butanol    | ++  | +  | +   | -  | -   | ++ | +   | ++   |  |
| Chloroform | ++  | +  | +   | -  | ++  | +  | -   | ++   |  |
| Ethanol    | ++  | ++ | ++  | ++ | +++ | ++ | +++ | +++  |  |

Grading of results: +, Zone of inhibition (10-15mm); ++, Zone of inhibition (16-20mm); +++, Zone of inhibition (21-23mm) in diameter.

<sup>a</sup>Microorganisms: I- Streptococcus pyogens, II- Staphylococcus aureus, III- Salmonella typhi, IV- Proteus vulgaries, V- Klebsiella pneumoniae, VI- Pseudomonas aeruginosa, VII- Bacillus subtilis, VIII- Escherichia coli.

 TABLE 2 : Antibacterial activity of various solvents crude

 extracts of Solanum trilobatum L

| Extraction | Disc diffusion method; size of inhibition<br>zones (9mm); bacteria <sup>a</sup> |    |     |    |     |    |     |      |  |
|------------|---|----|-----|----|-----|----|-----|------|--|
|            | Ι   | Π  | III | IV | V   | VI | VII | VIII |  |
| Acetone    | +   | +  | +   | +  | +++ | ++ | ++  | ++   |  |
| Aqueous    | ++  | ++ | +   | ++ | ++  | ++ | ++  | +    |  |
| Benzene    | ++  | ++ | +   | +  | +   | +  | ++  | +    |  |
| Butanol    | ++  | ++ | ++  | +  | +   | ++ | +   | ++   |  |
| Chloroform | ++  | ++ | ++  | +  | ++  | ++ | +   | ++   |  |
| Ethanol    | ++  | ++ | ++  | ++ | +++ | ++ | +++ | +++  |  |

Grading of results: +, Zone of inhibition (10-15mm); ++, Zone of inhibition (16-20mm); +++, Zone of inhibition (21-23mm) in diameter.

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#### TABLE 3 : Antibacterial reference standards

| Extraction       | Disc diffusion method; size of inhibition<br>zones (9mm); bacteria <sup>a</sup> |     |     |     |     |     |     |      |  |
|------------------|---|-----|-----|-----|-----|-----|-----|------|--|
|                  | Ι   | Π   | ш   | IV  | V   | VI  | VII | VIII |  |
| Chloram phenicol | +++   | +++ | +++ | +++ | +++ | +++ | +++ | +++  |  |
| Streptomycin     | +++   | +++ | +++ | +++ | +++ | +++ | +++ | +++  |  |

Grading of results: +, Zone of inhibition (10-15mm); ++, Zone of inhibition (16-20mm); +++, Zone of inhibition (21-23mm) in diameter.

<sup>a</sup>Microorganisms: I- Streptococcus pyogens, II- Staphylococcus aureus, III- Salmonella typhi, IV- Proteus vulgaries, V- Klebsiella pneumoniae, VI- Pseudomonas aeruginosa, VII- Bacillus subtilis, VIII- Escherichia coli.

plant observed the maximum zone of inhibition for *Strep*tococcus pyogens, *Staphylococcus aureus*, *Proteus vulgaries* and *Pseudomonas aeruginosa* (20, ++). The butanol extract of *Cadaba fruticosa* showed the maximum zone of inhibition for *Streptococcus pyogens*, *Pseudomonas aeruginosa* and *Escherichia coli* (20, ++). The chloroform extract of *Cadaba fruticosa* exhibited the maximum zone of inhibition for *Streptococcus pyogens*, *Klebsiella pneumoniae* and *Escherichia coli*.

All the solvents of Solanum trilobatum exhibited the maximum zone of inhibition for Salmonella typi (20, ++). The acetone and aqueous extract of Solanum trilobatum showed the maximum zone of inhibition for all tested microorganisms (20, ++). The benzene extract of the plant observed the maximum zone of inhibition for Streptococcus pyogens, Staphylococcus aureus, and Bacillus subtilis (20, ++). The butanol extract of Solanum trilobatum showed the maximum zone of inhibition for Streptococcus pyogens, Staphylococcus aureus, Klebsiella pneumoniae and Pseudomonas aeruginosa (20, ++). The chloroform extract of Solanum trilobatum exhibited the maximum zone of inhibition for *Streptococcus pyogens* (20, ++), Pseudomonas aeruginosa (20, ++) and Escherichia *coli* (23mm, +++).

#### DISCUSSION

Recently most of the researchers have been isolated active principles from popular medicinal plants and these are having a significant role in covering essential health needs in developing countries and these plants may produce a new source of antimicrobial agents with

<sup>&</sup>lt;sup>a</sup>Microorganisms: I- Streptococcus pyogens, II- Staphylococcus aureus, III- Salmonella typhi, IV- Proteus vulgaries, V- Klebsiella pneumoniae, VI- Pseudomonas aeruginosa, VII- Bacillus subtilis, VIII- Escherichia coli.

significant activity against infectious microorganisms<sup>[41]</sup>.

Nowadays, it has been lot of research interest in natural products which are very active against antibacterial activities<sup>[42]</sup>. The medicinal plants are not only used to heal all kinds of diseases, but also provide natural food for human beings<sup>[43,44]</sup>.

Medicinal plants are proving as an important source of potentially therapeutic drugs which are curing all kinds of infectious diseases throughout the world since the tradition of mankind, are still broadly used and have considerable importance in international trade<sup>[45]</sup>. Also it is a vital importance to identify novel substances active towards highly resistant pathogens<sup>[46]</sup>.

In the present study shows that organic solvents particularly ethanol exhibited better antibacterial activity than aqueous extracts and may be due to the antibacterial principles which are either polar or non polar and effectively extracted only through organic solvent medium<sup>[47-49]</sup>. The plant extracts are significantly active against one or more microorganisms used in the bioassay were also compared with standard antibiotics of chloramphenicol (30µg/disc) and streptomycin (30µg/ disc), which served as reference. These plants used for the treatment of several infections some have invitro activity against Gram-positive pathogenic bacteria. Thus, there is now preliminary scientific validation for the use of this medicinal plant for antibacterial activity. Further laboratory and clinical studies of this plant are required in order to understand better antibacterial principles, which will allow the scientific communities to recommend their use as an accessible alternative to synthetic antibiotics.

#### ACKNOWLEDGEMENTS

Authors are thankful to Correspondent, Principal and Head, Microbiology Department of Selvamm Arts and Science College, Namakkal, Tamilnadu, India for providing all necessary laboratory facilities during this research work

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