

## Original Research Article

### Antibacterial Activity of *Capparis sepiaria* L. (Capparidaceae) Leaves and Fruits

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#### ABSTRACT

The ethanolic extracts of *Capparis sepiaria* L. leaves and fruits were tested for their antibacterial activity against six species of bacteria, *Bacillus subtilis*, *Escherichia coli*, *Enterococcus faecalis*, *Klebsiella* sp., *Pseudomonas aeruginosa* and *Staphylococcus aureus* using disc diffusion technique. The extract concentrations of ethanolic leaf and fruit extracts used were 0 (control), 125, 250, 500 and 1000 ppm in triplicates along with standard antibiotic, chloramphenicol (30 µg). Comparatively, ethanolic fruit extracts showed higher activity than ethanolic leaf extracts of *Capparis sepiaria*. The results showed that In 1000 ppm leaf extract, a maximum of 2.1 cm ZI was observed against *Bacillus subtilis* followed by 2.0 against *Enterococcus faecalis*. The maximum ZI of 2.4 cm was recorded in 1000 ppm ethanolic fruit extract against *Pseudomonas aeruginosa* followed by 2.3 cm and 2.1 cm respectively against *Escherichia coli* and *Enterococcus faecalis*. The activity index found in ethanolic fruit extracts of *Capparis sepiaria* in 1000 ppm was 0.49, 0.79, 0.68, 0.41, 0.75 and 0.57 respectively against *Bacillus subtilis*, *Escherichia coli*, *Enterococcus faecalis*, *Klebsiella* sp., *Pseudomonas aeruginosa* and *Staphylococcus aureus*.

#### Keywords

Antibacterial activity, *Capparis sepiaria*, Fruit extracts, Medicinal plants.

#### Introduction

*Capparis sepiaria* L. belonging to the family, Capparidaceae is a thorny much branched shrub traditionally used for the treatment of jaundice, inflammation and dysentery (Indian Medicinal Plants, 1999; Matthew, 1991). Various parts of *Capparis sepiaria* has been reported to possess anti-inflammatory and analgesic activities (Chaudhari *et al.*, 2004) and, hepatoprotective activity (Satyanarayana

*et al.*, 2009); hepatoprotective activity (Satyanarayana *et al.*, 2009); ethanolic leaf extracts also showed significant *in vitro* free radical scavenging activity (Thirumalaikumaran and Reddy, 2011); stem extracts of *Capparis sepiaria* are reported to possess antimicrobial and anti-inflammatory activity (Satyanarayana *et al.*, 2010). There are very scientific reports available with regard to the biological

activities of *Capparis sepiaria*, the antibacterial activity of leaf and fruit extracts is scanty. Hence, the present study has been aimed to study the antibacterial activity of ethanolic extracts of leaves and fruits of *Capparis sepiaria* against six selective bacteria.

## Materials and Methods

The leaves and fruits of *Capparis sepiaria* L. (Fig. 1) belonging to the Family, Capparidaceae were collected and the identification was confirmed using standard local floras (Gamble and Fischer, 1957; Matthews, 1983). The leaves and fruits collected were transported to the laboratory for further processing. The cold extraction procedure was used for extracting leaves with solvents as per the procedure given below (Prakash and Karmegam, 2012; Vigneshwari *et al.*, 2014). The leaves and fruits of *Capparis sepiaria* collected were individually washed with tap water, blotted with filter paper and spread over news paper for air drying under shade. After complete dryness, the leaves and fruits were powdered using a mixer grinder. A known quantity of the powder (100 g) of each plant was taken in a 250 ml conical flask and added with 100-200 ml of ethanol individually for leaves and fruits. The solvent-powder mixtures were kept at room temperature for 48 hrs and rapidly stirred using glass rod every 8 hrs. After 48 hrs, the extract of each plant part was filtered through Whatmann No.1 filter paper to exclude the powder/particles. Then each filtrate was kept in beaker on a water bath at 45°C until the solvent gets evaporated. A greasy final material (crude extract) obtained for the leaves and fruits was transferred to screw cap tubes and stored under refrigerated condition till use.

By using digital electronic balance, 200 mg of each crude extract was carefully taken in

a standard measuring flask and 5 ml of ethanol was added to dissolve the extract and 1-2 drops of emulsifier (Triton-X100) was added to completely dissolve the extract. Then it was made up to 200 ml by adding distilled water. This forms the stock solution of 1000 ppm (i.e., 1mg/ml), from which different concentrations of test solutions, 125, 250, 500 and 1000 ppm were prepared and used for antibacterial assay. Disc diffusion method of antibacterial assay was used to test the sensitivity of selected test organisms to the ethanolic extracts adopting the method of Bauer *et al.* (1966). Each extract (100 µl) was applied to filter paper discs (Whatman No. 1) measuring 6 mm diameter and allowed to dry before being placed on the agar plate.

The test bacteria, *Bacillus subtilis*, *Escherichia coli*, *Enterococcus faecalis*, *Klebsiella* sp., *Pseudomonas aeruginosa* and *Staphylococcus aureus* maintained in the Laboratory of Department of Microbiology, Kanchi Shri Krishna College of Arts and Science, Kilambi, Kanchipuram which was originally obtained from the Microbial Type Culture Collection (MTTC) of Institute of Microbial Technology (IMTECH), Chandigarh were used for the present study. The Petri plates of 100mm diameter with nutrient agar media were swabbed with broth culture of the test bacteria in separate plates by using sterile swab. Over this, prepared antimicrobial discs were placed under aseptic conditions. Three discs of each extract were placed in triangle. Chloramphenicol (30 µg) was used as standard antibiotic. Also the discs without plant extract were also maintained as control. The plates were then incubated at 37°C for 24 hrs and the zone of inhibition (ZI) was measured in diameter (cm) around the discs and recorded. The assays were performed with three replicates. From the results, activity index was calculated by comparing the ZI of leaf extracts with

standard antibiotic as per the procedure adopted by Prakash and Karmegam (2016).

## Results and Discussion

*Capparis* is a genus distributed in many parts of the world with its many species showing varieties of medicinal properties. The medicinal uses of *Capparis sepiaria* are well known and the supporting scientific data available is very scanty. The present study reports the antibacterial activity of leaf and fruit extracts against six different bacteria. The standard antibiotic, chloramphenicol showed a range of 2.9-3.7 cm zone against the test bacteria. The antibacterial activity of leaf and fruit extracts of *Capparis sepiaria* measured in terms of zone of inhibition (ZI) showed variations among different concentrations of extracts and among different bacterial species tested (Tables 1 and 2). Only minimum activity was found in 125 ppm and 250 ppm concentrations of ethanolic leaf extracts against test bacteria. The leaf extract of 500 ppm showed antibacterial

activity against all bacteria tested excepting *Klebsiella* sp. which ranged from 0.9 to 1.3 cm ZI. In 1000 ppm leaf extract, a maximum of 2.1 cm ZI was observed against *Bacillus subtilis* followed by 2.0 against *Enterococcus faecalis*. The least ZI of 0.9 cm was found in 1000 ppm leaf extract against *Pseudomonas aeruginosa* (Table 1).

In comparison, the fruit extracts showed higher ZI against tested bacteria than ethanolic leaf extracts of *Capparis sepiaria*. The maximum ZI of 2.4 cm was recorded in 1000 ppm ethanolic fruit extract against *Pseudomonas aeruginosa* followed by 2.3 cm and 2.1 cm respectively against *Escherichia coli* and *Enterococcus faecalis* (Table 2). The lowest concentration used in the present study, i.e., 125 ppm of fruit extracts showed minimum activity against *Escherichia coli* and *Pseudomonas aeruginosa* whereas, zone around the disc was observed against *Enterococcus faecalis*, and 500 ppm of fruit extract showed antibacterial activity in all bacteria tested in the present study (Table 2).

**Table.1** Antibacterial Activity of Ethanolic Leaf Extracts of *Capparis Sepiaria* L

| Bacteria tested               | Zone of inhibition (cm) <sup>#</sup> |                    |         |         |         |          |
|-------------------------------|--------------------------------------|--------------------|---------|---------|---------|----------|
|                               | Std.*                                | 0 ppm <sup>§</sup> | 125 ppm | 250 ppm | 500 ppm | 1000 ppm |
| <i>Bacillus subtilis</i>      | 3.5                                  | -                  | AD      | 0.8     | 1.1     | 2.1      |
| <i>Escherichia coli</i>       | 2.9                                  | -                  | -       | -       | 1.0     | 1.5      |
| <i>Enterococcus faecalis</i>  | 3.1                                  | -                  | -       | AD      | 1.3     | 2.0      |
| <i>Klebsiella</i> sp.         | 3.7                                  | -                  | -       | -       | -       | 0.9      |
| <i>Pseudomonas aeruginosa</i> | 3.2                                  | -                  | -       | -       | 0.9     | 1.6      |
| <i>Staphylococcus aureus</i>  | 2.8                                  | -                  | -       | -       | AD      | 1.0      |

<sup>#</sup> - Values are mean of three replicates; <sup>§</sup> - Control (without extract); \*Std. - Standard antibiotic, Chloramphenicol (30 µg); AD – Around the disc.

**Table.2.** Antibacterial Activity of Ethanolic Fruit Extracts of *Capparis Sepiaria* L

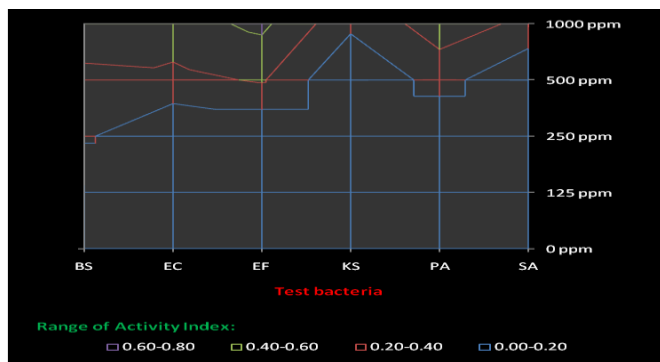
| Bacteria tested               | Zone of inhibition (cm) <sup>#</sup> |                    |         |         |         |          |
|-------------------------------|--------------------------------------|--------------------|---------|---------|---------|----------|
|                               | Std.*                                | 0 ppm <sup>§</sup> | 125 ppm | 250 ppm | 500 ppm | 1000 ppm |
| <i>Bacillus subtilis</i>      | 3.5                                  | -                  | -       | AD      | 1.1     | 1.7      |
| <i>Escherichia coli</i>       | 2.9                                  | -                  | 0.9     | 1.2     | 1.8     | 2.3      |
| <i>Enterococcus faecalis</i>  | 3.1                                  | -                  | AD      | 1.0     | 1.5     | 2.1      |
| <i>Klebsiella</i> sp.         | 3.7                                  | -                  | -       | -       | 1.0     | 1.5      |
| <i>Pseudomonas aeruginosa</i> | 3.2                                  | -                  | 0.9     | 1.3     | 1.8     | 2.4      |
| <i>Staphylococcus aureus</i>  | 2.8                                  | -                  | -       | AD      | 1.2     | 1.6      |

<sup>#</sup> - Values are mean of three replicates; <sup>§</sup> - Control (without extract); \*Std. - Standard antibiotic, Chloramphenicol (30 µg); AD – Around the disc.

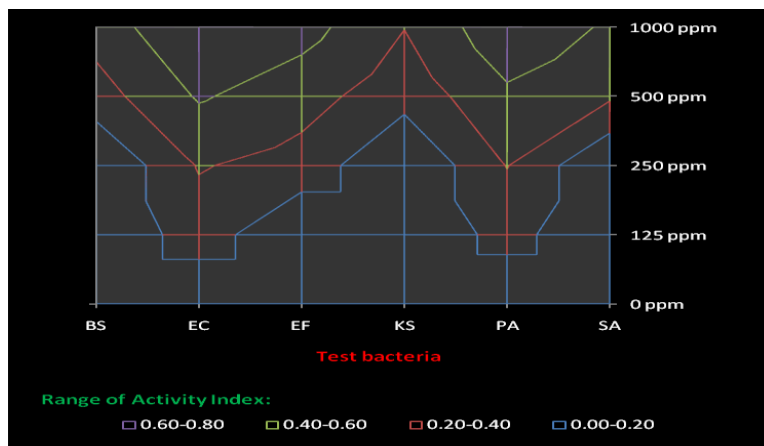
**Fig.1** *Capparis sepiaria* L. with Fruits



**Fig.2** The Range of Activity Index of Ethanolic Leaf Extracts of *Capparis sepiaria* L. Against Bacteria. Standard Antibiotic used: Chloramphenicol (30 µg); BS - *Bacillus Subtilis*, EC- *Escherichia Coli*, EF-*Enterococcus Faecalis*, KS-*Klebsiella* sp., PA-*Pseudomonas Aeruginosa* and SA-*Staphylococcus Aureus*



**Fig.3** The Range of Activity Index of Ethanolic Fruit extracts of *Capparis sepiaria* L. Against Bacteria. Standard Antibiotic used: Chloramphenicol (30 µg); BS - *Bacillus subtilis*, EC-*Escherichia coli*, EF-*Enterococcus faecalis*, KS-*Klebsiella sp.*, PA-*Pseudomonas aeruginosa* and SA-*Staphylococcus aureus*



The activity index exerted by ethanolic extracts of leaf and fruit of *Capparis sepiaria* showed a range of 0.23-0.79 (Fig. 2 and Fig. 3). A minimum activity index of 0.23 was observed in 250 ppm ethanolic leaf extracts of *Capparis sepiaria* against *Bacillus subtilis* and the maximum activity index of 0.79 was recorded in 1000 ppm ethanolic fruit extract against *Escherichia coli*. The activity index found in ethanolic leaf extracts of *Capparis sepiaria* in 1000 ppm was 0.60, 0.52, 0.65, 0.24, 0.50 and 0.36 respectively against *Bacillus subtilis*, *Escherichia coli*, *Enterococcus faecalis*, *Klebsiella sp.*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Least activity was recorded for lower concentrations and no activity was found in 125 ppm leaf extract. The activity index found in ethanolic fruit extracts of *Capparis sepiaria* in 1000 ppm was 0.49, 0.79, 0.68, 0.41, 0.75 and 0.57 respectively against *Bacillus subtilis*, *Escherichia coli*, *Enterococcus faecalis*, *Klebsiella sp.*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*.

Satyanarayana et al. (2010) reported that

ethanol soluble fractions of *Capparis sepiaria* stem showed potent activity against certain Gram positive (*Enterococcus faecalis*, *Staphylococcus aureus*) and Gram negative (*Pseudomonas aeruginosa* and *Escherichia coli*) pathogenic bacteria of Gastrointestinal tract. Mahboubi and Mahboubi (2014) revealed the scientific support to the traditional uses of roots and fruit extracts as antimicrobial agents by studying the methanol, ethanol and ethyl acetate extracts from fruit and root of *Capparis spinosa*, another important medicinal plant species of *Capparis*, which exhibited good activity against microorganisms, especially fungi. In the present study, the leaves and fruits were subjected to antimicrobial activity against selective bacteria in which the results revealed that the ethanolic fruit extracts had good antibacterial activity.

In conclusion, the ethanolic leaf and fruit extracts of *Capparis sepiaria* L. showed concentration dependent antibacterial activity. The extracts showed maximum activity in the highest concentration of 1000 ppm used in the present study. Ethanolic

fruit extracts of *Capparis sepiaria* showed higher activity than leaf extracts. The maximum ZI of 2.4 cm was recorded in 1000 ppm ethanolic fruit extract against *Pseudomonas aeruginosa* followed by 2.3 cm and 2.1 cm respectively against *Escherichia coli* and *Enterococcus faecalis*.

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### References

- Bauer, A.W., Kirby, W.M.N., Sherris J.C., Turck, M., 1966. Antibiotic susceptibility testing by a standardized single disc method. *Amer. J. Clin. Pathol.* 45, 493-496.
- Chaudhari, S.R., Chavan, M.J., Gaud, R.S., 2004. Phytochemical and pharmacological research on the roots of *Capparis sepiaria*. *Ind. J. Pharmaceut. Sci.* 66, 454-457.
- Gamble, J.S., Fischer, C.E.C., 1957. *Flora of the Presidency of Madras (Vols. I-III)*. Authority of Secretary of State for India in Council, London. 1389p.
- Indian Medicinal Plants, 1999. Vol. 1. Orient Longman Publishers, CSIR, New Delhi. 348p.
- Mahboubi, M., Mahboubi, A., 2014. Antimicrobial activity of *Capparis spinosa* as its usages in traditional medicine. *Kerba Polonica.* 60(1), 1-10.
- Matthews, K. M., 1991. Ovary borne on gynophore-ovules parietal armed or unarmed of Capparaceae family, in *Excursion flora of central Tamilnadu, India*. Oxford & IBH Publishers, New Delhi.
- Matthews, K.M., 1983. *The Flora of Tamil Nadu Carnatic*, St. Joseph's College, Tiruchirappalli, Tamil Nadu, India. 1034 p.
- Prakash, M., Karmegam, N., 2012. *In-vitro* antibacterial activity of certain plant extracts against plant disease causing bacteria isolated from citrus plant. *Int. J. Curr. Microbiol. App. Sci.* 1(1), 1-11.
- Satyanarayana, T., Kshama Devi, Mathews, A. A., 2009. Hepatoprotective activity of *Capparis sepiaria* stem against carbon tetrachloride-induced hepatotoxicity in rats. *J. Pharmaceut. Res. Health Care.* 1(1), 34-35.
- Satyanarayana, T., Mathews, A. A., Anjana Male, Ch. K. V. L. S. N., Surendra, G., 2010. Screening of anti-inflammatory and antimicrobial activities of stem extract of *Capparis sepiaria* Linn. *Res. J. Pharmaceut. Biol. Chem. Sci.* 1(3), 330-336.
- Thirumalaikumar, R., Reddy, C.U., 2011. Evaluation of hepatoprotective activity of *Capparis sepiaria* leaves. *Int. J. Biomed. Res.* 2(3), 204-212.
- Vigneshwari, C., Nagaraj, R., Karmegam, N., 2014. Synergistic anti-*Staphylococcus aureus* (methicillin resistant) activity of ethnomedicinal plants from Shevaroy hills (Eastern Ghats), south India. *Int. J. Curr. Res. Biosci. Plant Biol.* 1(2), 51-59.