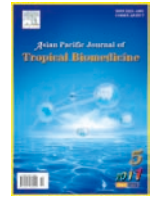




Contents lists available at ScienceDirect

## Asian Pacific Journal of Tropical Biomedicine

journal homepage: www.elsevier.com/locate/apjtb



Document heading doi:10.1016/S2221-1691(11)60089-0 © 2011 by the Asian Pacific Journal of Tropical Biomedicine. All rights reserved.

Antibacterial activity of various leaf extracts of *Merremia emarginata*EK Elumalai<sup>1</sup>, M Ramachandran<sup>2</sup>, T Thirumalai<sup>1</sup>, P Vinothkumar<sup>3\*</sup><sup>1</sup>P.G. and Research Department of Zoology, Physiology Wing, Voorhees College, Vellore-632001(T.N.), India<sup>2</sup>Bioinformatics Center, Central Agricultural Research Institute Port Blair, Andaman & Nicobar Islands-744 105, India<sup>3</sup>P.G. and Department of Microbiology, Mohamed Sathak College of Arts and Science, Sholinganallur, Chennai-600 119 (T.N.), India

## ARTICLE INFO

## Article history:

Received 15 March 2011  
 Received in revised form 2 April 2011  
 Accepted 15 April 2011  
 Available online 10 May 2011

## Keywords:

*Merremia emarginata*  
 Methanol extract  
 Aqueous extract

## ABSTRACT

**Objective:** To investigate the antibacterial activity and phytochemical screening of the aqueous, methanol and petroleum ether leaf extracts of *Merremia emarginata* (*M. emarginata*). **Methods:** The antibacterial activity of leaf extracts of *M. emarginata* were evaluated by agar well diffusion method against four selected bacterial species. **Results:** The presence of tannins, flavonoids, amino acids, starch, glycosides and carbohydrates in the different leaf extracts was established. The methanol extract was more effective against *Bacillus cereus* and *Escherichia coli*, whereas aqueous extract was more effective against *Staphylococcus aureus* and *Pseudomonas aeruginosa*. **Conclusions:** The results in the present study suggest that *M. emarginata* leaf can be used in treating diseases caused by the tested organisms.

## 1. Introduction

In recent years, human pathogenic microorganisms have developed resistance in response to the indiscriminate use of commercial antimicrobial drugs commonly employed in the treatment of infectious diseases. This situation, the undesirable side effect of certain antibiotics, and the emergence of previously uncommon infections, has forced scientists to look for new antimicrobial substitutions from various sources such as medicinal plants<sup>[1]</sup>. The screening of plant extracts and plant products for antimicrobial activity has shown that plants represent a potential source of new anti-infective agents<sup>[2–7]</sup>.

The plant *Merremia emarginata* (*M. emarginata*) Hallier f., belongs to family of convolvulaceae. In India it is mainly found in Chennai, and some place of Andhra Pradesh<sup>[8]</sup>. And it is known by various names in different regions viz., Mooshakarnee in Sanskrit, Indurkani in Bengal, Tolnnuatali in Telugu, Paerttaekirae in Tamil<sup>[9]</sup>. The plant was therapeutically used as deobstruent, diuretic, and for

cough, headache, neuralgia and rheumatism<sup>[10]</sup>. The present study was carried out to test the antibacterial efficacy of the leaves extract of *M. emarginata* Linn against bacterial spp.

## 2. Materials and methods

## 2.1. Plant material

The *M. emarginata* leaves collected during June–July of 2010 in and around Vellore, Tamilnadu were authenticated by Department of Botany. The voucher specimens were kept in the Department of Botany in C. Abdul Hakeem College, Melvisharam, Vellore, Tamilnadu, India.

## 2.2. Extraction procedure

Shade dried leaves (200 g) were coarsely powdered and subjected to successive solvent extraction by continuous Soxhlet extraction. The extraction was done with different solvents in their increasing order of polarity such as petroleum ether [(60–80) °C], methanol and water. Each time the marc was air dried and later extracted with other solvents. All the extracts were concentrated by distilling the solvent in a rotary flash evaporator. The yield was found

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to be 2.04%, 1.06%, and 3.37% w/w with reference to the air dried plant. The dried extracts were dissolved in dimethyl sulphoxide (DMSO) and subjected to antibacterial activity.

### 2.3. Test organisms

The bacterial spp. used for the test were *Staphylococcus aureus* (*S. aureus*), *Bacillus cereus* (*B. cereus*), *Escherichia coli* (*E. coli*) and *Pseudomonas aeruginosa* (*P. aeruginosa*). All the stock cultures were obtained from Microlab, Institute of Research and Technology, Vellore, Tamilnadu, India.

### 2.4. Culture media and inoculum preparation

Nutrient agar /broth (Himedia, India.) were used as the media for the culturing of bacterial strains. Loops full of all the bacterial cultures were inoculated in the nutrient broth (NA) at 37 °C for 72 hrs.

### 2.5 Preliminary phytochemical screening

All the extracts were subjected to preliminary phytochemical qualitative screening for the presence or absence of various primary or secondary metabolites<sup>[11]</sup>.

### 2.6 Antibacterial activity

The extracts obtained above were screened for their antibacterial activity in comparison with standard antibiotic penicillin (10 µg/mL) *in-vitro* by disc diffusion method<sup>[12]</sup>

using *S. aureus*, *B. cereus*, *E. coli* and *P. aeruginosa* as test organisms. Each extract was individually loaded on the 3 mm sterile disc at the concentration of 10 µg/mL, 25 µg/mL, 50 µg/mL and 100 µg/mL and subjected to antibacterial activity. The results were recorded by measuring the zone of growth inhibition surrounding the disc. The experiments were done in triplicate.

### 2.7. Statistical analysis

The results were expressed as mean ± SEM. Statistical analysis of the data were carried out using Student's *t*-test and the results were considered significant when *P*<0.05.

## 3. Results

The results of antibacterial activity are given in the Table 1 and 2, which clearly show that all the extracts at various concentrations have shown antibacterial activity equivalent to that of standard against the entire tested organisms. Aqueous, methanol and petroleum ether extracts have shown better activity than the standard against all the four microorganisms. Aqueous extract was more effective against *B. cereus* and *S. aureus*. Petroleum ether extract was more effective against *P. aeruginosa*, *B. cereus* and *E. coli*. Similar studies elsewhere recorded antibacterial activity of seed extracts against *S. aureus*, *Staphylococcus epidermidis*, *E. coli*, and *P. aeruginosa*<sup>[13]</sup>. The presence of various phytochemicals was shown in Table 3.

**Table 1**

Antibacterial activity of different extracts of leaves of *M. emarginata* against Gram positive organisms (Mean±SEM) (mm).

| Conc. of extract (µg/mL) | Zone of inhibition |            |            |                  |            |            |
|--------------------------|--------------------|------------|------------|------------------|------------|------------|
|                          | <i>S. aureus</i>   |            |            | <i>B. cereus</i> |            |            |
|                          | AE                 | ME         | PEE        | AE               | ME         | PEE        |
| 10                       | 13.01±0.01         | 12.06±0.03 | 11.05±0.25 | –                | 13.00±0.00 | 12.03±0.09 |
| 25                       | 13.01±0.23         | 12.04±0.04 | 11.02±0.01 | 07.00±0.00       | 13.05±0.03 | 12.02±0.10 |
| 50                       | 13.06±0.12         | 12.5±0.03  | 11.04±0.16 | 09.01±0.23       | 13.05±0.03 | 12.04±0.17 |
| 100                      | 13.7±0.23          | 12.05±0.03 | 12.00±0.01 | 09.20±0.12       | 13.5±0.02  | 12.04±0.15 |
| Penicillin (10)          |                    | 16.01±0.12 |            |                  | 17.03±0.16 |            |

AE: Aqueous extract; ME: Methanol extract; PEE: Petroleum ether extract.

**Table 2**

Antibacterial activity of different extracts of leaves of *M. emarginata* against Gram negative organisms (Mean±SEM) (mm).

| Conc. of extract (µg/mL) | Zone of inhibition |            |            |                      |            |            |
|--------------------------|--------------------|------------|------------|----------------------|------------|------------|
|                          | <i>E. coli</i>     |            |            | <i>P. aeruginosa</i> |            |            |
|                          | AE                 | ME         | PEE        | AE                   | ME         | PEE        |
| 10                       | 08.01±0.03         | 11.00±0.01 | 07.02±0.10 | 10.02±0.01           | 07.00±0.00 | 11.00±0.05 |
| 25                       | 08.09±0.01         | 11.20±0.03 | 07.06±0.05 | 10.40±0.11           | 07.09±0.23 | 11.08±0.12 |
| 50                       | 09.02±0.14         | 11.07±0.08 | 08.00±0.11 | 11.01±0.03           | 09.02±0.01 | 11.09±0.15 |
| 100                      | 09.08±0.13         | 12.01±0.09 | 08.80±0.06 | 11.09±0.02           | 09.08±0.06 | 12.01±0.05 |
| Penicillin(10)           |                    | 16.09±0.15 |            |                      | 16.02±0.13 |            |

AE: Aqueous extract; ME: Methanol extract; PEE: Petroleum ether extract.

**Table 3**

Preliminary phytochemical analysis of *M. emarginata* leaves extracted with different solvents

| Phytoconstituents | AE | ME | PEE |
|-------------------|----|----|-----|
| Alkaloids         | –  | –  | –   |
| Tannins           | –  | +  | +   |
| Flavonoids        | +  | +  | +   |
| Terpenoids        | +  | –  | –   |
| Amino acids       | +  | +  | +   |
| Cardic glycosides | +  | –  | –   |
| Carbohydrates     | +  | +  | +   |
| Sponins           | –  | –  | –   |
| Starch            | +  | –  | +   |

AE: Aqueous extract; ME: Methanol extract; PEE: Petroleum ether extract.

#### 4. Discussion

The therapeutic value of medicinal plants lies in the various chemical constituent's presents in it. The bioactivity of plant extracts is attributed to phytochemical constituents. For instance, plant rich in tannins have antibacterial potential due to their basic character that allows them to react with proteins to form stable water soluble compounds thereby killing the bacteria by directly damaging its cell membrane<sup>[14]</sup>. Flavonoids are a major group of phenolic compounds reported for their antiviral<sup>[15]</sup>, antimicrobial<sup>[16]</sup> and spasmolytic<sup>[17]</sup> properties. Alkaloids isolated from plant are commonly found to have antimicrobial properties<sup>[18]</sup>. Extract of the seeds of *Vitex agnus-castus* was reported to possess antimicrobial activity which was associated with its alkaloids, saponins, tannins, flavonoids, and glycosides contents<sup>[19]</sup>. The antibacterial activity of the leaf extracts of *M. emarginata* as recorded in present study may therefore be attributed to the presence of above phytochemicals *i.e.* flavonoids, terpenoids, amino acids, glycosides and starch in aqueous extract, tannins, flavonoids, amino acids and carbohydrates in methanol extract and tannins, flavonoids, amino acids, glycosides and carbohydrates in petroleum ether extract.

It is concluded that the plant extract possess antibacterial activity against tested organisms. The zone of inhibition varied suggesting the varying degree of efficacy and different phyto constituents of herb on the target organism. The antibacterial activity of the plants may be due to the presence of various active principles in their leaves. Further studies are needed to isolate and characterize the bioactive principles to develop new antibacterial drugs.

#### Conflict of interest statement

We declare that we have no conflict of interest.

#### Acknowledgements

The authors are thankful to Dr. B Senthilkumar and Dr.

Ernest David, Vellore, Tamilnadu for his valuable suggestion and constant encouragement.

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