



## Research Article

### ANTIMICROBIAL ACTIVITY OF *MIMOSA PUDICA* THORNS

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Article Received on: 10/06/18 Approved for publication: 29/06/18

DOI: 10.7897/2230-8407.096117

#### ABSTRACT

Traditional medicinal plants play an important role in maintaining the human health. One such traditional plant is *Mimosa Pudica*, also known as humble plant but acts as antimicrobial agent. Ethanolic and aqueous extracts of *Mimosa pudica* thorns were assessed for preliminary phytochemical analysis. Also the extracts were evaluated for the antimicrobial activity against bacteria and fungi by agar well diffusion method. The microorganisms used in the study were *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Bacillus cereus* and *Candida albicans*. The plant extracts were taken in various concentrations (25, 50, 75 and 100 µg/well) and the standards used for antibacterial and antifungal activity were Streptomycin and Clotrimazole respectively. The phytochemical screening revealed flavonoids, saponins, glycosides, alkaloids, terpenoids and coumarins in both ethanolic and aqueous thorn extracts of *Mimosa pudica*. *Escherichia coli* showed maximum zone of clearance in both the ethanolic and aqueous thorns extracts of *Mimosa pudica* at higher concentrations. Amongst the two extracts, the results clearly indicate that the aqueous thorn extracts of *Mimosa pudica* exhibited highest zone of inhibition of 24.2±0.34mm against the bacteria *Escherichia coli* and 18.1±0.17mm against the fungi *Candida albicans* at 100µg/well. Therefore the present study concludes that the phytoconstituents might have contributed the antimicrobial activity and further studies can be made on the plant derived antimicrobials.

**Key words:** *Mimosa pudica*, Antimicrobial activity, Phytoconstituents, Agar well diffusion method

#### INTRODUCTION

Plants are the untapped resources of the secondary metabolites. Plants have played a significant role in maintaining human health and improving the quality of human life<sup>1</sup>. The phytochemical constituents of a medicinal plant could be directly correlated with its pharmacological activity<sup>2</sup>. The secondary metabolites of the plant extracts could be responsible for the antimicrobial activity<sup>3</sup>. Antimicrobials of plant origin possess reduced side effects and are effective against infectious diseases<sup>4</sup>. By searching new bio-molecules of plant origin and evaluating them for antimicrobial property would result in developing eco-friendly management of human infectious diseases<sup>5</sup>. The non nutritive phytochemicals obtained from plants have protective or disease preventive antimicrobial activities, the structural differences in them results in the difference in their mode of action<sup>6</sup>. The mechanism behind antibacterial activity of plant extracts includes disruption of bacterial membrane potential, permeabilization and leakage of the cellular contents<sup>7</sup>.

*Mimosa pudica* is also called touch me not, shy or sensitive plant<sup>8</sup>. It belongs to the family Fabaceae and subfamily Mimosoideae<sup>9</sup>. The presence of terpenoids, flavonoids, alkaloids, quinines, phenols, tannins, saponins and coumarins in methanolic leaf extracts of *Mimosa pudica* makes it more potent to exhibit antimicrobial activity<sup>10</sup>. *Mimosa pudica* possesses sedative, emetic, and tonic properties and also was used in the treatment of various ailments including alopecia, diarrhea, dysentery, insomnia, tumor, and various urogenital infections. And it has shown antidepressant, anticonvulsant, antivenom activities and diuretic effect<sup>11</sup>. In the present study, the phytochemicals were

screened and the antimicrobial activity of the ethanolic and aqueous thorn extracts of *Mimosa pudica* was analyzed.

#### MATERIALS AND METHODS

##### Collection and identification of plant material

The *Mimosa pudica* plant was collected from Thirukalikundram, Kanchipuram District. Authentication of the plant was done by Dr. Sasikala Ethirajulu and Dr. Jega Jothi Pandian, Siddha Central Research Institute, Arignar Anna Government Hospital campus, Arumbakkam, Chennai. *Mimosa pudica* thorns were thoroughly washed with fresh water and then shade dried at room temperature. After which they were powdered using pulverizer and were used for extract preparation.

##### Preparation of ethanolic & aqueous thorn extracts

**Ethanolic thorn extract:** 20 grams of the powdered thorns of *Mimosa pudica* with 250ml of ethanol were subjected to successive extractions using Soxhlet extractor for 15 refluxes. After condensing the extract using rotary evaporator, the thorn extract was labelled and stored at 5°C for further use.

**Aqueous thorn extract:** 20 grams of the powdered thorns of *Mimosa pudica* were soaked in 250 ml double distilled water and was continuously agitated in an orbital shaker for 24 hours in a closed Erlenmeyer flask. The extract was filtered using Whatmann No.1 filter paper. The solvent from the extract was removed using rotary vacuum evaporator. The extract obtained was labelled and then stored at 5°C for further use.

### Phytochemical screening

Qualitative phytochemical screening of *Mimosa pudica* thorn extracts was done by making use of the standard methods<sup>12, 13</sup>. The phytochemicals screened were flavonoids, steroids, tannins, saponins, glycosides, alkaloids, terpenoids, anthraquinones and coumarins.

### Antimicrobial Activity

#### Test pathogenic microorganisms

Pathogenic Gram negative bacteria such as *Escherichia coli* (MTCC 443), *Pseudomonas aeruginosa* (MTCC 2453), *Klebsiella pneumoniae* (MTCC 530) and Gram positive bacteria such as *Bacillus cereus* (MTCC 430) and fungi, *Candida albicans* (MTCC 227) were used for *invitro* antimicrobial activity. These selected strains were obtained from Microbial Type Culture Collection, IMTECH, Chandigarh.

#### *In vitro* antimicrobial activity

Antimicrobial activity was determined by agar well diffusion method<sup>14</sup>. About 25 ml of molten Mueller Hinton agar was poured into a sterile Petri plate (Himedia, Mumbai, India). The plates were allowed to solidify, after 18 hours (OD adjusted to 0.6) 100  $\mu$ l of above said type strains were transferred onto plate and made culture lawn by using sterile L-rod spreader. After five minute setting of the type strains, a sterile cork borer was used to make 5

mm well on the agar in a uniform manner. The plant extracts were dissolved in sterile saline and loaded into wells with various concentrations (25, 50, 75 and 100  $\mu$ g/well). The solvent saline loaded well served as negative control. Streptomycin (30 $\mu$ g/ml) well served as positive control for bacteria. The plates were incubated for 24 hours at 37°C in a 40W florescent light source (~400 nm). The antibacterial activity was determined by measuring the diameter of the zone of inhibition around the well using antibiotic zone scale (Himedia, Mumbai, India).

#### *In vitro* antifungal activity

The sensitivity of fungi was determined by well diffusion method<sup>14</sup>. About 25 ml of potato dextrose agar was poured into a sterile Petri plate (Himedia, Mumbai, India). The plates were allowed to solidify, after 18hours grown yeast pathogen was swabbed using sterile cotton swab. The plant extracts were dissolved in sterile water and loaded into wells with various concentrations (25, 50, 75 and 100  $\mu$ g/well). The Clotrimazole (30 $\mu$ g/ml) well served as positive control. All the loaded plates were kept for 72 hours. The antifungal activity was determined by measuring the diameter of the zone of inhibition around the well using antibiotic zone scale (Himedia, Mumbai, India).

**Statistical analysis:** The samples were taken in triplicates and analyzed, the results were reported in mean $\pm$ standard deviation (SD).

## RESULTS AND DISCUSSION

**Table 1: Antimicrobial activity of *Mimosa Pudica* aqueous thorn extracts**

Name of the organism	Zone of inhibition (mm)				ZOI(mm) Positive control
	25 $\mu$ g/well	50 $\mu$ g/well	75 $\mu$ g/well	100 $\mu$ g/well	
Concentration	25 $\mu$ g/well	50 $\mu$ g/well	75 $\mu$ g/well	100 $\mu$ g/well	30 $\mu$ g/well
<i>Escherichia coli</i>	19.33 $\pm$ 0.57	20.16 $\pm$ 0.28	22.13 $\pm$ 0.23	24.2 $\pm$ 0.34	20.06 $\pm$ 0.11*
<i>Pseudomonas aeruginosa</i>	14.13 $\pm$ 0.23	16.23 $\pm$ 0.40	18.33 $\pm$ 0.57	20.1 $\pm$ 0.17	19.33 $\pm$ 0.57*
<i>Klebsiella pneumoniae</i>	---	---	---	8.1 $\pm$ 0.17	13.16 $\pm$ 0.28*
<i>Bacillus cereus</i>	12.06 $\pm$ 0.11	14.1 $\pm$ 0.17	15.13 $\pm$ 0.23	17.16 $\pm$ 0.28	15.2 $\pm$ 0.34*
<i>Candida albicans</i>	10.06 $\pm$ 0.11	12.13 $\pm$ 0.23	13.06 $\pm$ 0.11	18.1 $\pm$ 0.17	16.33 $\pm$ 0.57 <sup>§</sup>

\*: Streptomycin; <sup>§</sup>: Clotrimazole

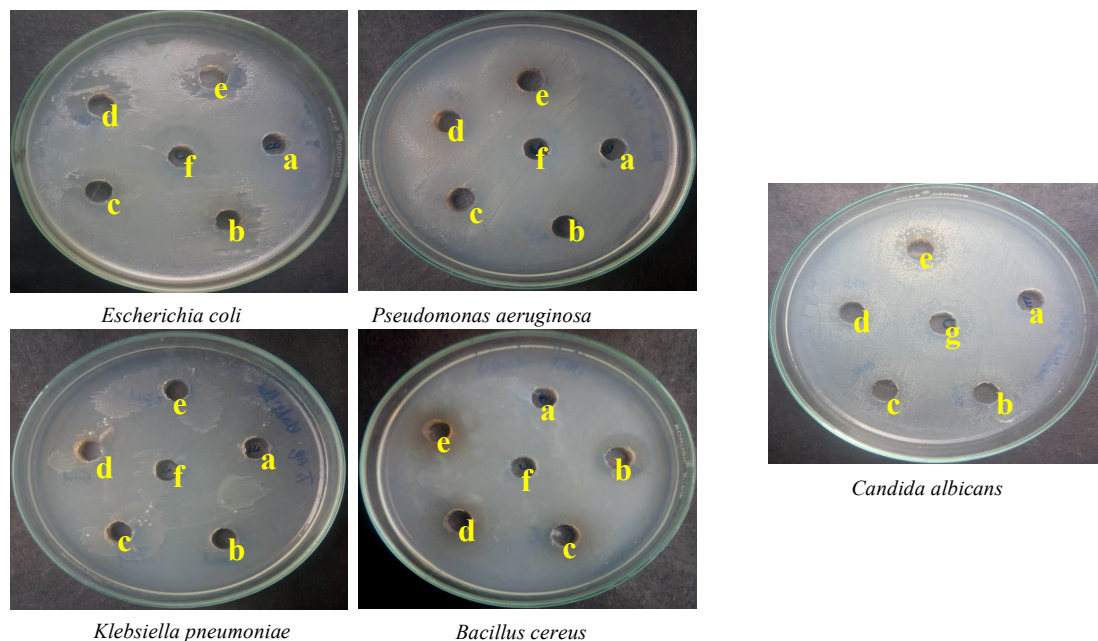
Data are mean  $\pm$  SD values; n=3

**Table 2: Antimicrobial activity of *Mimosa Pudica* ethanolic thorn extracts**

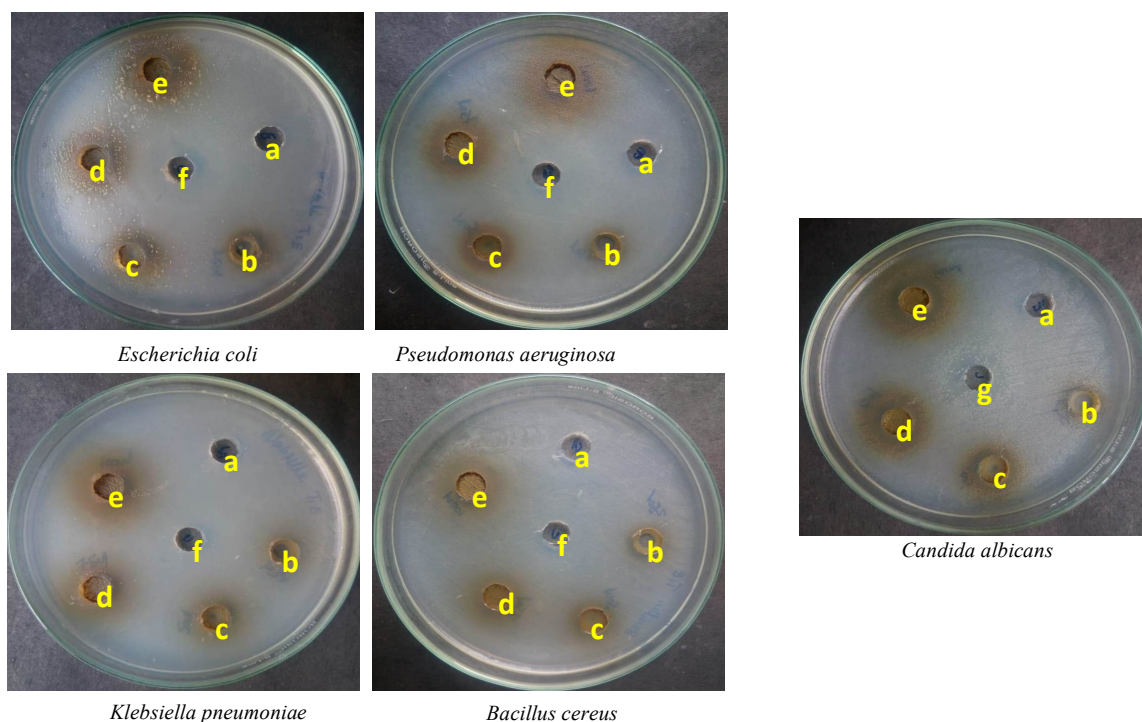
Name of the organism	Zone of inhibition (mm)				ZOI(mm) Positive control
	25 $\mu$ g/well	50 $\mu$ g/well	75 $\mu$ g/well	100 $\mu$ g/well	
Concentration	25 $\mu$ g/well	50 $\mu$ g/well	75 $\mu$ g/well	100 $\mu$ g/well	30 $\mu$ g/well
<i>Escherichia coli</i>	9.1 $\pm$ 0.17	11.13 $\pm$ 0.23	14.06 $\pm$ 0.11	16.23 $\pm$ 0.4	22.13 $\pm$ 0.23*
<i>Pseudomonas aeruginosa</i>	8.13 $\pm$ 0.23	11.06 $\pm$ 0.11	13.03 $\pm$ 0.05	13.23 $\pm$ 0.25	20.1 $\pm$ 0.17*
<i>Klebsiella pneumoniae</i>	7.06 $\pm$ 0.11	8.13 $\pm$ 0.23	11.1 $\pm$ 0.17	13.2 $\pm$ 0.34	14.16 $\pm$ 0.28*
<i>Bacillus cereus</i>	7.1 $\pm$ 0.17	8.66 $\pm$ 0.57	10.06 $\pm$ 0.11	13.16 $\pm$ 0.28	16.2 $\pm$ 0.34*
<i>Candida albicans</i>	6.06 $\pm$ 0.11	8.13 $\pm$ 0.23	11.16 $\pm$ 0.28	13.3 $\pm$ 0.3	15.1 $\pm$ 0.17 <sup>§</sup>

\*: Streptomycin; <sup>§</sup>: Clotrimazole

Data are mean  $\pm$  SD values; n=3



**Figure 1: Antimicrobial activity of *Mimosa pudica* aqueous thorn extracts**  
a: 0 µg/well; b: 25 µg/well; c: 50 µg/well; d: 75 µg/well; e: 100 µg/well; f: 30 µg/well streptomycin / g: 30 µg/well clotrimazole



**Figure 2: Antimicrobial activity of *Mimosa pudica* ethanolic thorn extracts**  
a: 0 µg/well; b: 25 µg/well; c: 50 µg/well; d: 75 µg/well; e: 100 µg/well; f: 30 µg/well streptomycin / g: 30 µg/well clotrimazole

**Phytochemical analysis**

Preliminary phytochemical analysis of ethanolic and aqueous thorn extracts of *Mimosa pudica* revealed the presence of flavonoids, saponins, glycosides, alkaloids, terpenoids and coumarins. Steroids were present in the ethanolic thorn extracts of *Mimosa pudica* and in the aqueous thorn extracts, tannins were present. The phytochemical results of earlier studies suggest that

the chloroform extract of *Mimosa pudica* Lin., leaves revealed the presence of steroids, flavonoids, glycosides, alkaloids, phenolic compounds<sup>15</sup>. And in another study, it was reported that the methanolic extract of *Mimosa pudica* consists of the phytoconstituents such as flavonoids, glycosides, alkaloids and that might be the reason for its hepatoprotective activity<sup>16</sup>. The crude ethanolic extract of *Mimosa pudica* leaves and roots has showed the presence of tannins, proteins and steroids<sup>17</sup>. It is

supported by a study that presence of alkaloids and tannins in the *Mimosa pudica* ethanolic leaf extracts would be the reason for its antimicrobial activity<sup>18</sup>.

Also it is suggested in a study that antimicrobial activity of plant extracts are attributed by either individual or combined effects of the phytochemicals like flavonoids, saponin, alkaloids and glycosides<sup>19</sup>. It is reported in a previous finding that the flavonoids, alkaloids and tannins are antimicrobial active plant principles<sup>20</sup>. Flavonoids are synthesized in response to microbial infection and tannins act as antimicrobial agents by precipitating the microbial protein<sup>21,22</sup>. It is reported in a study that the saponins exhibits antibacterial activity due to its potential in increasing the permeability of bacterial cell wall<sup>23</sup>.

#### Antimicrobial activity

The aqueous thorn extracts of *Mimosa pudica* exhibited highest zone of inhibition of 24.2±0.34mm for *Escherichia coli*. Similarly, the ethanolic thorn extracts of *Mimosa pudica* exhibited highest zone of inhibition (16.23±0.4mm) against *Escherichia coli* at 100µg/well. In a previous antibacterial study, it was reported that aqueous extracts of *Mimosa Pudica* leaf extracts showed a maximum zone of inhibition against *Escherichia coli*<sup>24</sup>. The efficacy of *Mimosa Pudica* thorn extracts against *Escherichia coli* might be due to the presence of the phytoconstituents. It was reported in a study that the antibacterial activity of the plant extract can be due to the phytochemical content<sup>25</sup>. *Escherichia coli* is reported to be a causative of intestinal infection like diarrhea. Alkaloids possess antidiarrheal effect due to their effects on transit time in the small intestine<sup>26</sup>. Therefore in the present study, the presence of alkaloids in the thorn extracts of *Mimosa pudica* might have contributed the antibacterial activity against *Escherichia coli*.

Ethanolic thorn extracts of *Mimosa pudica* thorns showed moderate activity against *Klebsiella pneumoniae*. But it was resistant against the aqueous extracts of *Mimosa pudica* thorns and revealed lowest zone of inhibition of 8.1±0.17mm at 100µg/well. The results are supported by a study, in which it was stated that *Klebsiella pneumoniae* was least susceptible organism to *Mimosa pudica* extracts<sup>27</sup>.

The ethanolic extracts of *Mimosa pudica* displayed effective antibacterial activity against *Escherichia coli*, but showed comparatively less zone of inhibition against *Pseudomonas aeruginosa* and *Bacillus cereus* at 100µg/well. And the positive control (Streptomycin) produced zone of inhibition against all selected microorganisms. Investigations done earlier states that the *Mimosa pudica* leaf extracts showed maximum zone of inhibition against *Escherichia coli* and *Pseudomonas aeruginosa* and exhibited good sensitivity towards the same<sup>28</sup>. The results of the current study is supported by previous research in which the ethanolic extracts of *Mimosa pudica* exhibits antimicrobial activity at higher concentrations and a clear zone of inhibition was not shown at lower concentrations<sup>29</sup>. In an earlier study, it was observed that ethanolic extracts of *Mimosa pudica* twigs showed antibacterial activities<sup>30</sup>. Previous studies revealed that higher concentrations of phenolic compounds in plant extracts might be the reason for the higher antibacterial activity<sup>31</sup>. Flavonoids were found to exhibit antibacterial activity by the mechanisms like inhibiting nucleic acid synthesis, cytoplasmic membrane functions and energy metabolism<sup>32</sup>.

Both the aqueous and ethanolic thorn extracts of *Mimosa pudica* were evaluated by antifungal activity using *Candida albicans*. Amongst the two extracts, the results clearly show that the aqueous thorn extracts of *Mimosa pudica* exhibited highest zone

of inhibition of 18.1±0.17mm against the fungi, *Candida albicans* at 100µg/well. Ethanolic thorn extracts of *Mimosa pudica* showed lowest zone of inhibition of 13.3±0.3mm against *Candida albicans* at 100µg/well. The positive control used in antifungal assay was Clotrimazole which produced zone of inhibition against *Candida albicans*. In a previous invitro studies, it was reported that coumarin was found to inhibit *Candida albicans*<sup>26</sup>. It was suggested in a study that antifungal activity of extracts of *Mimosa pudica* might be due to their complex forming ability with extracellular and soluble proteins and with cellwall<sup>33</sup>. The *Mimosa pudica* extract exhibited modest activity against *Candida albicans*<sup>21, 27</sup>.

#### CONCLUSION

In the current study, the results evaluated that the aqueous thorn extracts of *Mimosa pudica* exhibited higher antibacterial and antifungal activity compared with the ethanolic thorn extracts of *Mimosa pudica* except with *Klebsiella pneumoniae*. The presence of flavonoids, saponins, glycosides, alkaloids, terpenoids and coumarins in the ethanolic and aqueous thorn extracts of *Mimosa pudica* were revealed in the phytochemical analysis. The rich source of wide variety of plant derived biomolecules would be the reason for the antimicrobial activity. In this era of change where antibiotic resistant organisms rise in alarming rate, the novel bioactive principles could be used as leads to develop medicines to treat infectious diseases. So further studies could be done to find the nature and efficacy of the phytochemicals in the plant.

#### ACKNOWLEDGEMENT

The authors are thankful to members who helped during plant collection and analysis.

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**Cite this article as:**

R.Lakshmibai and D.Amirtham. Antimicrobial activity of *Mimosa pudica* thorns. Int. Res. J. Pharm. 2018;9(6):202-206 <http://dx.doi.org/10.7897/2230-8407.096117>

Source of support: Nil, Conflict of interest: None Declared

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