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Research Article

Screening for phytochemical and antibacterial activity of leaves of *Mimosa Pudica L.* and *Lippia Abysinica* against urinary tract infection causing pathogens

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Abstract

Medicinal plants play a significant role in human health. One such traditional plant is *Mimosa pudica* L. which acts as an antimicrobial agent. The antibacterial activity against bacteria by agar well diffusion method of Ethanolic extracts. The microorganisms used in the study were *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Bacillus cereus*, and *Candida albicans*. Different concentrations of plant extract were used (50, 100 and 150 µg/well). Also, *Mimosa pudica* L. leaves extracts were assessed for preliminary phytochemical analysis. The phytochemical screening of *Mimosa pudica* L. ethanolic leaves extracts revealed the presence of flavonoids, saponins, glycosides, alkaloids, terpenoids, and coumarins. Compared to other Microorganisms *Escherichia coli* showed a maximum zone of inhibition in the ethanolic leaves extracts of *Mimosa pudica* at higher concentrations. Amongst the two extracts, the results clearly indicate that the *Mimosa pudica* L. leaves extract of ethanol exhibited the highest zone of inhibition of 24.2±0.34mm against the bacteria *Escherichia coli*. Therefore the present study concludes that the phytochemical might have contributed to the antimicrobial activity and further studies can be made on the plant-derived antimicrobials

Keywords: Antibacterial, Phytochemical, Lippia Abysinica, Mimosa pudica L. and UTI Pathogen.

Introduction

Plants are the resources of secondary metabolites that have played a significant role in human health and life (Azmi et al., 2011). The pharmacological activity of a medicinal plant is directly correlated with its Phytoconstituents (Das et al., 2013). The secondary metabolites of leaf extract play a major role in antimicrobial activity (Cyril et al., 2013). Antimicrobials of leaf origin possess reduced side effects and are effective against infectious diseases (Jagtap et al., 2012).

Over the past few decades, people are most commonly affected by urinary tract infections (UTI) throughout their lifespan. Urinary Tract Infection (UTI) is complex and influenced by many host biological behavioral factors of complicated and uncomplicated pathogenesis and it has properties of the infecting uro pathogens. The prevalence of urinary tract infection (UTI) by bacteria is problematic

worldwide and among all ages in both genders. Every year, millions of people were diagnosed with UTI with frequent incidence among females. Most UTI infection pathogens include *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella spp.*, *Proteus spp.*, *Pseudomonas spp.*, *Enterobacter spp.*, in immune-compromised patients and pregnant women.

World Health Organization (WHO) report showed that due to the lack of discovery of new antimicrobial drugs, lead to the overuse and misuse of available antibiotics. The pharmaceutical industries make the crisis more severe and life threaten condition UTI infected patients. Urinary tract infection is an important cause of morbidity in the general population and is the second most frequent cause of visits to a doctor's office after respiratory tract infection (Ronald, 2003). UTI is the third most common cause of hospitalization in India.

Medicinal plants are the oldest form of healthcare known to mankind throughout history and are reported for their medicinal potential which is used in the Indian system of medicine, viz Ayurveda, Unani, Siddha, etc. Term “herbal drug” determines the parts of a plant (leaves, flowers, seeds, roots, barks, stems, etc) used for preparing medicine. Medicinal plants are used as a complementary for the treatment of emerging and re-emerging of Because of their availability, accessibility, affordable and acceptance to the local population multidrug-resistant bacteria (MDR) is undergoing revitalization.

Owing to this, the present study was designed to evaluate the antibacterial activity of leaf extract of two medicinal plants such as *Mimosa pudica* L. and *Lippia abyssinica* (Otto & A. Dietr) are renewed medicinal plants with lots of traditional importance.

Mimosa pudica L., also called a sensitive plant or touch-me-not, belongs to the genus *Mimosa* (Family: Mimosaceae). The treatment for some diseases and conditions like diarrhea, insomnia, tumor, headache, fever, skin infections, and blood pressure *M.pudica* played a major role in traditional medicine. The presence of various bioactive compounds in this leaf like tannins, steroids, flavonoids, glycosides, non-protein amino acid leucenine (mimosine), and alkaloids (Tamilarasi et al., 2012). On the basis of these back, it contains active constituents like an alkaloid Mimocine, mucilage, tannins, non-protein amino acids, flavonoid C-glycosides, sterols, and terpenoids. *Mimosa pudica* L. shows certain movements like nyctinastic movements, thigmonastic movements, and seismonastic movements. It exhibits anti-ulcer, anti-inflammatory, anti-microbial, anti-malarial, anti-fungal, analgesic, anti-convulsant, anti-diarrheal, anti-hepatotoxic, anti-helminthic and anti-depressant activities (Ahmad et al., 2012 and Zhang et al., 2011).

Lippia abyssinica derives from Latin and means of or from Ethiopia (*Abyssinia*). Traditionally, dried leaves are used as one of the ingredients in the preparation of spiced butter and also for food flavoring agents and preservatives. It has hairy leaves and small flowers that are purple or pink.

Material and method

Materials

Methanol, ethanol, acetone, distilled water, screw cap tubes, and all glassware used from the Department of Microbiology, Annamalai University, Tamil Nadu.

Cleaning of glassware

All the glassware was soaked in a cleaning solution 100 g of potassium dichromate was added to 100 ml of distilled water followed by the addition of 500 ml of concentrated sulphuric acid for about 12 hours and washed in tap water. They were washed with boiled soap water and thoroughly rinsed in tap water

and dried. They were sterilized at 180°C for 3 hours in a Hot air oven.

Collection and Extraction of Plant Materials

Fresh leaves of *Mimosa pudica*. L and *Lippia abyssinica* were collected in March 2022 and identified by Dr. L. Mullainathan, Professor of Botany, Department of Botany, Annamalai University, Tamil Nadu. The voucher specimen was deposited at the Herbarium, Department of Botany, Annamalai University, Tamil Nadu.

The leaves were collected in a polyethylene bag followed by sprinkling water on the leaves and piercing the bag at several points in order to allow air circulation. In the laboratory, the leaves were washed three times under running tap water followed by rinsing once with sterile distilled water, and then air-dried. The leaves were then ground into a fine powder using an electric grinder followed by the preparation of suspensions in methanol, ethanol, and acetone. 10 grams of the fine powder was suspended in 100ml of each of the three solvents separately in sterilized screw-capped 500ml glass beakers. The filtrate was evaporated in a petri dish at room temperature for 2-3 days till the volume was reduced to one-fourth of the original volume of the solvent prepared and stored at 4°C in air-tight bottles.

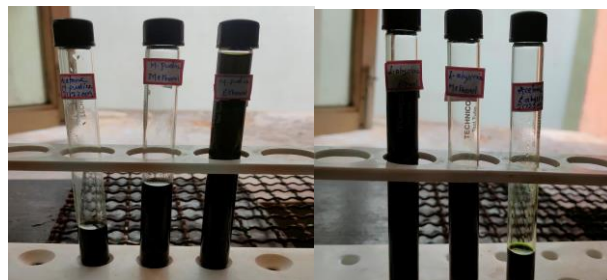


Fig. 1

Fig. 2

Fig. 1 Leaves extract of *Mimosa pudica* L.

Fig. 2 Leaves extract of *Lippia abyssinica*

Preparation of inoculum

Stock bacterial cultures were maintained in nutrient agar slant in test tubes and stored at 4°C. Test cultures were prepared by transferring a loop full of stock culture into test tubes of muller Hinton agar for bacterial strains that were incubated without agitation for 24 hours at 37°C. After 24 hours of incubation, the turbidity of the test bacterial suspension is compared with that of 0.5Mac Farland Standard against a white background with a contrasting black line under adequate light. All the bacterial cultures were checked for viability and purity.

Antibacterial activity

The antibacterial activity of Methanol, ethanol and aqueous extract of *Mimosa pudica* L. and *Lippia abyssinica* was

evaluated separately by agar well diffusion method. The test bacterial strains were inoculated into Mueller-Hinton broth and incubated at 37°C for 24 hours. After incubation, a sterile cotton swab was immersed in the bacterial suspension and swabbed aseptically on the surface of the Muller-Hinton agar medium and allowed to dry for about 3 minutes. A well of 6mm diameter were punched into an agar medium filled with the 50 µl, 100 µl, and 150 µl, concentration of leaves extract of both *Mimosa pudica* L. and *Lippia abyssinica*. The compound was allowed to diffuse for 5 min. The plates were incubated at 37°C for 24 hours. Antibacterial activity was evaluated by measuring the zone of inhibition against the test organisms.

Phytochemical screening of ethanolic extract of *M. pudica* L.

Among the three extract i.e, methanol, ethanol and acetone, it was found that ethanolic extract of *Mimosa pudica* L. shows maximum zone of inhibition against selected UTI pathogens and were subjected to a through phytochemical screening using standard protocols to detect the following secondary metabolites: alkaloids, coumarins, terpenes, tannins, anthraquinone, phenols, leucoanthocyanidin, flavones and saponins. The results are indicated as (+) for the presence and (-) for the absence of phytochemicals.

Result

In the present study an attempt was made to find and compare the antibacterial activity between *Mimosa pudica* L. and *Lippia abyssinica* against selected UTI pathogens.

Antibacterial activity

The antibacterial activity of ethanol, methanol, and acetone extract of leaves of *Mimosa pudica* L. and *Lippia abyssinica* were evaluated against the bacteria causing UTI infections such as *Escherichia coli*, *Enterobacter spp.*, *Staphylococcus*

aureus, *Proteus spp.*, *Pseudomonas spp.*, and *Klebsiella spp.*, by agar well diffusion method. It was found that the leaves extract of *Mimosa pudica* L. exhibits greater antibacterial activity as compared to the leaf extract of *Lippia abyssinica* against selected UTI pathogens. The ethanol extract of *Mimosa pudica* L. was found to show maximum antibacterial activity for selected pathogens followed by acetone extract and least antibacterial activity exhibited by methanol extract. The ethanol extract of *Mimosa pudica* L. exhibit maximum zone of inhibition against *Escherichia coli* followed by *Klebsiella spp.*, *Pseudomonas spp.*, *Proteus spp.*, *Enterobacter spp.*, and *Staphylococcus aureus* was found resistant to ethanolic extract. The bacteria *Escherichia coli* (22±1.52) and *Klebsiella spp* (22±1.15) show a maximum inhibition zone at 150 µl and the least shown by *Staphylococcus aureus* at 50µl is (12±0.56). [Table 1](#) represents the leaves extract of *Mimosa pudica* L. against selected pathogens at different concentrations (50µl,100µl and 150µl).

The leaves extract of *Lippia abyssinica* exhibit the least antibacterial activity against all pathogens. The bacteria *Staphylococcus aureus* shows the maximum zone of inhibition against methanolic extract at 150µl (17±0.88) and all the pathogens were highly resistant to its acetone extract. [Table 2](#) represents the leaves extract of *Lippia abyssinica* against selected pathogens at different concentrations (50µl,100µl and 150µl).

Phytochemical screening of plant extract

Since the ethanol extract of *Mimosa pudica* L. showed significant activity by the presence of, Resins, Carboxylic acid, Steroids, Flavonoid, Carbohydrates, Saponification and Protein, The absence of Tannins, Glycosides, Phenol, Biuret, Saponin, Gum, Flavonol glycosides and Alkaloids. [Table 3](#) shows the phytochemical screening of ethanol leaves extract of *Mimosa pudica* L was performed qualitatively ([Mohamed Sham Shihabudeen et al., 2013](#)).

Table 1. Antibacterial activity of *Mimosa pudica* L. (mm)

S.No	Test Organisms	Ethanol (mm)			Methanol (mm)			Acetone (mm)		
		50 µl	100 µl	150 µl	50 µl	100 µl	150 µl	50 µl	100 µl	150 µl
1.	<i>Escherichia coli</i>	13±1.15	20±0.23	22±1.52	10±1.12	12±1.32	14±1.04	11±0.45	12±1.05	15±1.12
2.	<i>Enterobacter</i>	13±0.13	14±1.23	18±1.22	11±1.11	13±0.12	14±1.11	15±1.11	18±1.23	20±0.16
3.	<i>Proteus</i>	13±1.23	15±1.63	18±1.43	13±0.52	16±0.43	18±1.54	13±1.07	16±1.19	18±1.45
4.	<i>Pseudomonas</i>	10±1.34	16±0.72	20±0.24	11±1.00	13±1.17	16±1.32	12±1.43	14±0.54	18±1.00
5.	<i>Klebsiella</i>	13±1.12	17±0.53	22±1.15	12±1.32	14±1.15	16±1.54	15±0.45	16±0.23	18±1.32
6.	<i>Staphylococcus Aureus</i>	12±0.56	14±1.45	16±1.55	11±0.43	14±0.39	16±1.23	11±0.22	13±0.11	15±1.11

Table 2. Antibacterial activity of *Lippia abyssinica* (mm-millimetre)

S.No	Test Organisms	Ethanol (mm)			Methanol (mm)			Acetone (mm)		
		50 µl	100 µl	150 µl	50 µl	100 µl	150 µl	50 µl	100 µl	150 µl
1.	<i>Escherichia coli</i>	11±0.07	11±1.09	14±0.04	10±0.06	12±0.09	13±1.01	10±0.01	11±1.32	12±1.23
2.	<i>Enterobacter</i>	10±1.00	12±0.56	14±1.87	12±0.45	13±0.76	14±1.12	10±0.45	11±1.21	13±1.34
3.	<i>Proteus</i>	10±0.26	10±0.73	12±0.45	10±1.65	12±0.54	13±1.19	11±1.02	12±1.43	13±0.23
4.	<i>Pseudomonas</i>	12±0.34	13±1.54	15±1.43	11±1.23	12±1.43	14±1.43	12±1.24	14±0.65	15±0.54
5.	<i>Klebsiella</i>	11±1.12	10±1.76	15±1.23	13±1.09	14±1.12	15±1.23	11±1.18	13±0.23	14±1.09
6.	<i>Staphylococcus Aureus</i>	10±1.02	13±0.45	17±0.87	11±1.10	12±1.32	17±0.88	10±1.45	12±0.13	15±1.11

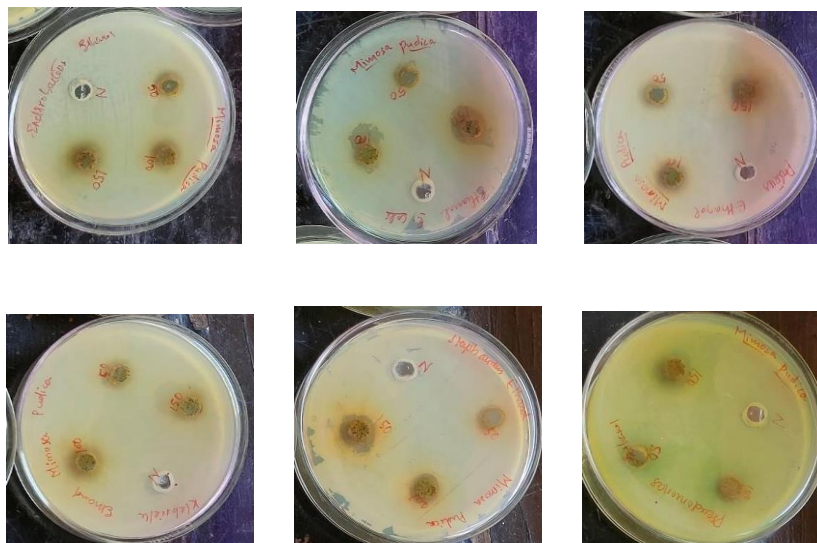


Fig. 1 Antibacterial activity of *Mimosa pudica* ethanol leaves extract

Table 3. Qualitative phytochemical screening methods of ethanolic extract of *Mimosapudica* L.

Sl. No.	Name of the Sample	Phytochemical compound	Result
1	Ethanolic Extract of <i>Mimosapudica</i> L.	Resins	+
2		Carboxylic acid	+
3		Tannins	-
4		Steroids	+
5		Flavonoid	+
6		Carbohydrates	+
7		Glycosides	-
8		Saponification	+
9		Protein	0.63
10		Phenol	-
11		Biuret	-
12		Saponin	-
13		Gum	-
14		Flavonol glycosides	-
15		Alkaloids	-



Fig. 2 Qualitative phytochemical screening methods

Discussion

In the present study, plants and leaves are abundant resources, but these resources are dwindling fast due to the onward march of civilization (Vogel, 1991). Although a significant number of studies have been used to obtain purified plant chemicals, very few screening programs have been initiated on crude plant materials. In the present study, the ethanol leaves extract exhibited higher antibacterial properties than the aqueous extract. In fact, the ethanol leaves extract resisted four tested pathogens while the aqueous extract revealed no inhibition of growth of *Escherichia coli* and *Klebsiella sp.* Moreover, the inhibition zone produced by the ethanol solvent against selected bacteria was larger than Methanol and the Aqueous solvent *Mimosa pudica L.* It has also been widely observed and accepted that the medicinal value of plants lies in the bioactive photo components present in the plants (Veeramuthu et al., 2008).

Conclusion

The two different herbal plants *Mimosa pudica L.* and *Lippia abyssinica* were collected from Sivapuri village and TS Pettai, Cuddalore district, Tamil Nadu. Plants of *Mimosa pudica L.* and *Lippia abyssinica* were tested for antibacterial activity against UTI pathogens i.e., *Escherichia coli*, *Enterobacter spp.*, *Staphylococcus aureus*, *Klebsiella spp.*, *Pseudomonas and proteus spp.* Among two herbal plants, *Mimosa pudica L.* exhibited the maximum zone of inhibition against bacteria, followed by *Lippia abyssinica*.

Mimosa pudica L. exhibited maximum zone of inhibition against *Escherichia coli* and *Klebsiella* and *Lippia abyssinica* exhibited maximum zone of inhibition against *Staphylococcus aureus*. This study of screening antibacterial activity of leaves extract of *Mimosa pudica L.* and *Lippia abyssinica* derived the conclusion that *Mimosa pudica L.* possess strong antibacterial activity when compared to *Lippia abyssinica* Though both the plants possess some sort of activity, *Mimosa pudica L.* can be considered as a more potent agent in drug research against UTI causing bacteria. These findings support the traditional knowledge of local users and it is preliminary, scientific validation for these plants to create awareness among the local community.

Conflict of Interest

The author hereby declares no conflict of interest.

Consent for publication

The author declares that the work has consent for publication.

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