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# Absence of anti fungicidal activity of few Indian medicinal plants methanolic extracts

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## ABSTRACT

**Objective:** The present study was aimed at evaluating the In vitro antimicrobial activity of few Indian medicinal plants *H. tiliaceus* L., *N. arbortristis* Linn., *S. rhombifolia* Linn., *T. procumbens* Linn methanolic extracts against *A. alternata*, *A. flavus*, *A. niger*, *M. phaseolina*, *R. solani* using agar well diffusion technique. **Methods:** Soxhlet extraction method was used to get the extracts of methanol. The length of zone of inhibition was measured in millimeters from the edge of the well to the edge of the inhibition zone. **Results:** It showed zero results with *H. tiliaceus*, *N. arbortristis* with all three 100, 300, 500 mg/ml DMSO concentrations. **Conclusions:** Various medical plants have been used for years in daily life to treat disease all over the world throughout the history of mankind. Plant Species *H. tiliaceus* and *N. arbortristis* may not be useful in controlling diseases against *A. alternata*, *A. flavus*, *A. niger*, *M. phaseolina*, *R. solani*. This study give an idea to avoid work of activity of these medicinal plants extracts on mentioned fungal for screening.

## 1. Introduction

Plants are the largest biochemical and pharmaceutical sources ever known on our planet. These living factories are able to generate endless biochemical compounds [1]. Medicinal plants represent a rich source of antimicrobial agents [2–6]. *Hibiscus tiliaceus* L. (Family–Malvaceae), is a mangrove plant growing in tropical Asia and abundant in littoral forests and mangrove forest margins of atolls and high islands [7]. In folk medicine, the leaves of this plant used to treat fevers, soothe coughs, ulcer, wounds and various skin diseases [8]. *Nyctanthes arbortristis* Linn. (Family– Oleaceae) commonly known as Night Jasmine is one of the well known medicinal plants. Juice of the leaves is used as digestives, antidote to reptile venoms, mild bitter tonic, laxative, diaphoretic and diuretic [9,10,11]. Traditionally the powdered stem bark is given in rheumatic joint pain, in treatment of malaria and also used as an expectorant [12]. *Sida rhombifolia* Linn. (Malvaceae) is a perennial or sometimes annual plant native to the tropic and subtropic areas. In India, infusion of leaves of *S. rhombifolia*

has been shown to possess diuretic and aphrodisiac effects *Tridax procumbens* Linn. (Family–Asteraceae) the whole plant was reported to treat various ailments, such as bronchial catarrh, dysentery, diarrhea, preventing hair loss, and to check hemorrhage from cuts [13].

## 2 Materials and methods

### 2.1 Solvents and chemicals used

All chemicals were purchased from Merck, Qualigens fine Chemicals and SD fine chemicals, Mumbai.

### 2.2 Plant material and extraction

Healthy, disease free, plant materials were collected from different places of Andhra Pradesh including Coringa Mangrove Wetland, Kakinada India. They were taxonomically identified and the Voucher specimens were deposited in the herbarium of the Department of Botany, Andhra University, Visakhapatnam. The plant material were dried under shade with occasional shifting and then powdered with a mechanical grinder and stored in an airtight container. The powder 100 gms obtained were

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subjected to Soxhlet extraction with Methanol.

2.3 Test microorganisms

The microbial strains *Alternaria alternata* (MTCC 1362), *Aspergillus flavus* (MTCC 4633), *Aspergillus niger* (MTCC 2723), *Macrophomina phaseolina* (MTCC 2165), *Rhizoctonia solani* (MTCC 4633) fungi were procured from Microbial Type Culture Collection (MTCC), Chandigarh. The strains are maintained at Potato Dextrose Agar (PDA) for fungi. Active cultures were generated by inoculating a loopful of culture in separate 100 mL potato dextrose broths and incubating on a shaker at 37°C overnight. The cells were harvested by centrifuging at 4000 rpm for 5 min, washed with normal saline, spun at 4000 rpm for 5 min again and diluted in normal saline to obtain  $5 \times 10^5$  cfu/mL.

2.4 Determination of antimicrobial activity

The crude extracts of the different plant parts of different

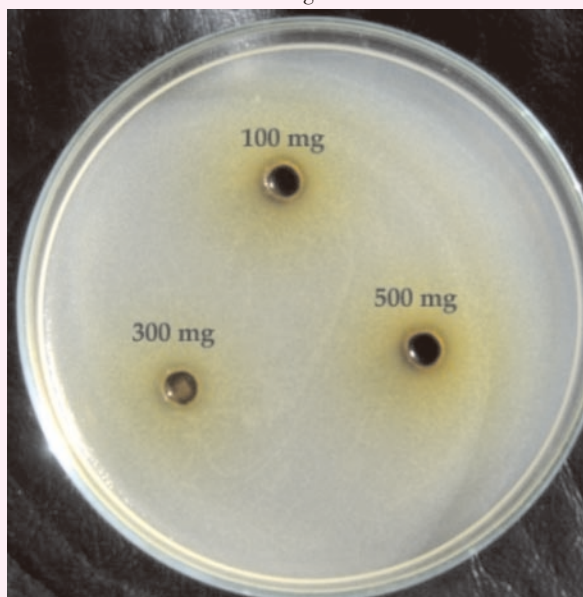
species were subjected to antimicrobial assay using the agar well diffusion method of [14] modified by [15]. 20 ml of nutrient agar was dispensed into sterile universal bottles these were then inoculated with 0.2 ml of cultures mixed gently and poured into sterile petri dishes. After setting a number 3–cup borer (6mm) diameter was properly sterilized by flaming and used to make three to five uniform cups/wells in each petri dish. A drop of molten nutrient agar was used to seal the base of each cup. The cups/wells were filled with 50 µl of the extract concentration of 100mg/ml, 300mg/ml and 500 mg/ml and allow diffusing for 45 minutes. The solvents used for reconstituting the extracts were similarly analyzed. The plates were incubated at 37 °C for 24 hours for bacteria. The above procedure is allowed for fungal assays but except the media potato dextrose agar instead of nutrient agar and incubates at 25 °C for 48 hours. The zones of inhibition were measured with antibiotic zone scale in mm and the experiment was carried out in triplicates.

Table 1

Fungal strains	Name of Plant Species											
	<i>H. tiliaceous</i>			<i>N. arbortristis</i>			<i>S. rhombifolia</i>			<i>T. procumbens</i>		
	A= 100, B= 300, C= 500 (mg/ml DMSO) Dilutions											
	A	B	C	A	B	C	A	B	C	A	B	C
<i>A. alternata</i>	–	–	–	–	–	–	–	–	–	7± 0.14	9±0.05	10±0.25
<i>A. flavus</i>	–	–	–	–	–	–	–	–	–	9±0.24	11± 1.3	13±1.4
<i>A. niger</i>	–	–	–	–	–	–	–	–	–	–	–	–
<i>M. phaseolina</i>	–	–	–	–	–	–	–	–	–	–	–	–
<i>R. solani</i>	–	–	–	–	–	–	10±0.25	12±1.2	14 ±1.00	–	–	–

Mean±S. D of Diameter of the zone of inhibition in (mm)

Fig 1



No results or no zone of inhibition with all three concentrations with *H. tiliaceous* against *A. alternata*

### 3. Results

The antifungal activity was determined by measuring the diameter of zone of inhibition in milli meters (mm). The antimicrobial activities of methanolic extracts were represented in Table 1 according to the results *T. procumbens* was some extant good against *A. alternata* and *A. flavus* with all concentration where as *S. rhombifolia* is significant against *R. solani*. No inhibition was found with methanolic extracts of *H. tiliaceus* and *N. arbortristis* with all three 100, 300, 500 mg/ml DMSO Dilutions.

### 4. Discussion

The variation of susceptibility of the tested microorganisms could be attributed to their intrinsic properties that are related to the permeability of their cell surface to the extracts. Negative results do not indicate the absence of bioactive constituents, nor is that the plants *H. tiliaceus*, *N. arbortristis* are inactive. Active compound(s) may be present in insufficient quantities in the crude extracts to show activity with the dose levels employed. Lack of activity can thus only be proven by using large doses. On the other hand, if the active principle is present in high quantities, there could be other constituents exerting antagonistic effects of the bioactive compounds. It is not surprising that there are differences in the antimicrobial activities of plant groups, due to the phytochemical differences between species.

### Conclusion

Plants are being used as medicines by mankind since the ancient times and they are being taken as a good source of drugs (Deshwal and Siddiqui, 2011a).

Plants are being used as medicines by mankind since the ancient times and they are being taken as a good source of drugs [16] but selected plant Species *H. tiliaceus* and *N. arbortristis* may not be useful in controlling diseases against *A. alternata*, *A. flavus*, *A. niger*, *M. phaseolina*, *R. solani*. This study give an idea to avoid work of activity of these medicinal plants extracts on mentioned fungal for screening.

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