

ANTIMICROBIAL ACTIVITY OF GENUS *TERMINALIA THORELLI*. COMBRETACEAE

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ABSTRACT

The antimicrobial activity of crude and methanol extract of leaves and dry fruit of *Terminalia thorelli* was tested by disc diffusion method, against six human pathogens. Crude aqueous extract of leaves and dry fruit of 5mg concentration showed zone of inhibition ranging from 16-25 mm. *S. aureus* was found to be highly susceptible with highest zone of inhibition suggesting the strong inhibitory activity of these extracts towards the selected bacterial pathogens. The pathogens were more sensitive to the methanol extracts forming 18-28 mm zone of inhibition suggesting that the methanol extract is little more effective than crude extracts against most of these microbes except (entero-pathogen). The minimal inhibitory concentration (MICs) of the extracts of leaves and dry fruit were determined by broth dilution method, ranged from 250 to 1800 mg/ml and 200 to 1600 mg /ml respectively. These results indicate that leaves and dry fruit of *T. thorelli* possess potential antibacterial activity.

KEY WORDS: Anti-microbial activity, Human pathogens, MIC, *T. thorelli*.

INTRODUCTION

Increasing resistance of the pathogenic microbes to chemotherapy lead to the screening of herbal anti-microbial potential from various plants ((Elizabeth, 2005; Murali *et al.*, 2004; Rastogi and Mehrotra, 1999; Dama *et al.*, 1998). *Terminalia thorelli* is one of the exotic immigrant plant belongs to 'Combretaceae' with Indo-Malayan origin. However, it is well adapted now in India and reveals considerable similarities with the other species similarities with the other species of *Terminalia*. The other species from the family *Combretaceae* Viz. *T. chebula*, *T. cattapa* were constantly been used by tribals (Elizabeth, 2005), and these species were thoroughly investigated for their anti-microbial activity of crude and methanol extract of leaves and dry fruit of *T.thorelli* on some pathogenic microbes. The major objective of this study is to identify whether *T. thorelli* show similar anti-microbial properties as that of its Indian counterparts.

MATERIAL AND METHODS

Fresh leaves and dry fruits of *T. thorelli* were collected from the Dr Babasaheb Ambedkar Marathwada University campus, Aurangabad, India. The leaves were sterilized in running water and extracted in distilled water (15 gm in 100ml) and methanol (15 gm 100 ml). Similarly the dry fruits devoid of seeds ground to fine powder and mixed in sterile distilled water to give the concentration of 1gm/5ml stock solution. Then the extracts were stored in refrigerator until further use. The methanol extract of dry fruit was prepared by pulverizing 1 kg of dry fruits in 2.5 lit of absolute methanol for 48 hr. Later the solution was collected and subjected to several cycles of distillation until a thick brown paste was obtained. One gram of residual methanol extract was mixed in 5 ml of methanol to give concentration of 1 ml=0.2 mg of *T. thorelli*.

The bacterial strains used for the study were *Staphylococcus aureus* (ATCC 8145), *Salmonella typhimorium* (ATCC22486), *Escherichia coli* (uropathogen), *Escherichia coli* (entero pathogen), *Pseudomonas aeruginosa* (ATCC 24618) and *Candida albicans* (ATCC2086) procured from MTCC, Chandigarh, India. All chemicals and antibiotics impregnated discs used in the study were procured from Hi Media, Mumbai, India. For testing anti-microbial activity, disc diffusion method was used as given by Elizabeth (2005). For this nutrient agar / or broth was used to culture bacteria and Sabouraud's agar / or broth to cultivate *C. albicans*. Fresh overnight inoculums of each culture (0.1 ml) containing 10⁸ cells was spread on the agar plate, three sterile paper discs (5mm diameter) were placed in each agar plate and on two of them crude and methanol extract of leaves (5mg in 20 ml volume) was placed and on third 20 ml of absolute methanol as a control. One Streptocyclin disc (10µg) was placed as a positive control in each plate.

The bacterial cultures were incubated at 37° C for 18-24 h and *C. albicans* at room temperature for 48 h. Zone of inhibition was measured. The experiment was done in triplicate and average zone of inhibition was noted. To determine the Minimal inhibitory concentration (MICs) different concentrations of *T. thorelli* crude leaf extract in nutrient broth was serially diluted in triplicates. Control test tubes did not receive any *T. thorelli* extract. Later 10 cells of *S. aureus* in 0.02 ml was added to each test tube and incubated at 37° C for 18-24 h. The lowest concentration of drug inhibits the growth was considered as MIC. Similarly the MIC's of crude and methanol leaf extracts of leaves and dry fruit were determined against each microbe under study.

RESULTS AND DISCUSSION

The results were summarized in table 1 and 2. In the present study both crude and methanol extracts of leaves and dry fruit of *T. thorelii* were strongly inhibitory to *S. aureus* forming large zone of inhibition, closely followed *E. coli* (entero pathogen) and *P. aeruginosa* respectively (Table-1). However, the methanol extract of both leaves and dry fruit is found more effective than crude aqueous extract. The methanol extracts of leaves and dry fruit showed high degree of inhibition towards *S. aureus* and *P. aeruginosa*, indicating the presence of antibacterial principle in the extract.

Table 1. Antimicrobial activity of *T. thorelii* and Zone of inhibition in mm.

Organism	Leaf extract		Fruit extract		Streptomycin
	Aqueous	Methanol	Aqueous	Methanol	
<i>S. aureus</i>	25.0	26.0	24.0	28.0	22.0
<i>S. typhimurium</i>	18.0	20.0	18.0	23.0	14.0
<i>E. coli (UTI)</i>	19.0	24.0	18.5	22.0	00
<i>E. coli (EP)</i>	22.0	24.0	20.0	22.0	00
<i>P. aeruginosa</i>	17.5	18.0	16.0	18.0	00
<i>C. albicans</i>	18.5	20.0	21.0	23.0	19.5

Table 2. Minimum inhibitory concentrations ($\mu\text{g/ml}$) of *T. thorelii* leaves and dry fruit.

Organism	Leaf extract		Fruit extract	
	Aqueous	Methanol	Aqueous	Methanol
<i>S. aureus</i>	280	250	250	200
<i>S. typhimurium</i>	1200	600	1000	800
<i>E. coli (UTI)</i>	1800	400	2000	500
<i>E. coli (EP)</i>	600	1600	400	600
<i>P. aeruginosa</i>	1800	1200	1000	800
<i>C. albicans</i>	1800	1500	1800	1600

Moreover, crude and methanol extract of both leaves and dry fruit of *T. thorelii* showed moderate inhibitory activity against *C. albicans* (Table- 1 and 2). The high degree of inhibition of pathogen is directly supported by low MIC values (Table 2). The MICs against *S. aureus*, *P. aeruginosa* and *S. typhimurium* are comparatively less than the others. This result shows homology of the phytochemicals and active anti-microbial principles present in other *Terminalia* species. Murali, Y. K *et al.* (2004). The similar activity from *T. bellerica* was reported where dry fruit extract showed high inhibition of pathogenic bacteria and can normalize the blood pressure.

The inhibitory activity of *T. thorelii* leaf extract can be attributed to the phytochemicals present in them Rastogi and Mehrotra (1999). It has been reported that the phenolics, tannins and propyl gallate were strong microbial inhibitors. The similar kind of action is predicated in *T. thorelii*. The previous reports of *T. chebula*, *T. cattapa* and *T. bellerica* showed that they have anti-microbial activity in their leaves and fruits. Ritch-Kro *et al* (1996). From the present study, it can be concluded that *T. thorelii* leaves as well as fruits also possess broad spectrum anti-microbial activity than the crude extract. It can also be used as substitute for other Indian *Terminalia* species at commercial level.

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