

Evaluation of antibacterial and antifungal activity of fruiting body extracts of *Trametes versicolor*

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ABSTRACT

The present study was carried out to evaluate the antibacterial and antifungal activity of methanol extract of fruit bodies of *Trametes versicolor* on selected four bacterial pathogens such as *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Staphylococcus aureus* and four fungal strains *Penicillium* spp., *Aspergillus fumigatus*, *A. niger* and *A. flavus*. For antimicrobial test, well diffusion technique was used and the zone of inhibition of microorganisms was measured in mm. The fruit body of *Trametes versicolor* showed potential antimicrobial activities against the selected strains and maximum inhibition was observed in methanol extract against *Staphylococcus aureus*, *Pseudomonas aeruginosa*. The maximum antifungal activity was observed in methanol extract against *Aspergillus fumigatus* and *A. niger*.

Key words: *Trametes versicolor*, Anti-bacterial activity, Anti-fungal activity.

INTRODUCTION

Macro fungi have been proved to be one of the richest sources for producing a large and diverse variety of secondary metabolites with significant bioactivities (H. Mugdha et al., 2010). One macrofungi species can have various bioactive compounds and pharmacological effects (U Lindequist et al., 2005). Multiple drug resistance in human pathogenic microorganisms has been developed, due to over use of antimicrobial drugs against infectious diseases. Search for new antimicrobial substances from various plants is very much popularized as novel antimicrobial chemotherapeutic agents (YS. Karaman et al., 2003. A. Turkoglu et al., 2007). Mushrooms are rich sources of natural antibiotics, and therefore mushrooms extracts have been studied for their antimicrobial

activity. Lignicolous macrofungi have the capability to express significant biological effects, including antibacterial activity (JM. Hur et al., 2004, NK. Ishikawa et al., 2005, F. Kalyoncu et al., 2010) and their secondary metabolites can be easily extracted and identified. According to the recent biological study, more than 75% of screened polypores showed strong antimicrobial activity inhibiting mostly Gram-positive bacterial strains (*B. subtilis*, *S. aureus* and *M. flavus*). It was reported that new sesquiterpenoid hydroquinones produced by some species of the European *Ganoderma* genus, named ganomycins, inhibit the growth of methicillin-resistant *S. aureus* and other bacteria (RAA. Mothana et al., 2000). The fungal metabolites of fruiting bodies frequently differ from those of mycelia of submerged cultures or fermentation broth. Moreover,

biogenetic pathways are rather dependent on their habitats or geographic origin.

The aim of the current study is to evaluate the antibacterial and antifungal activity of *Trametes versicolor* extracts with the help of methanol and aqueous extract against bacterial strains (*Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Staphylococcus aureus*) and fungal species (*Penicillium* sps., *Aspergillus fumigatus*, *A. niger*, and *A. flavus*).

MATERIAL AND METHODS

Collection of Fruiting Body:

A Basidiomycetes species, *Trametes versicolor* was used in this study and collected from Eturnagaram forest, Warangal District. The fruiting body was surface sterilized with disinfectant and a piece of fruiting body was placed in the MEA agar medium in petri dishes. When it starts growth on the medium a small piece of the mycelium was transferred to fresh Malt agar media in tubes. This was regularly carried out until pure culture was obtained. Molecular-based characterization on ribotyping of 18S rRNA was carried at the Scigenome, Kochin, India and sequence were deposited to EMBL database for accession number.

Bacterial Cultures Used:

Staphylococcus aureus, *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa* were procured from Microbial Type Collection (MTCC) center, IMTECH, Chandigarh, India.

Fungal Cultures Used:

Pathogenic fungal strains *Penicillium* sps., *Aspergillus fumigatus*, *A. niger* and *A. flavus* were collected from the Botany Department, Kakatiya University, Warangal, India.

Well Diffusion Method:

Antibacterial and Antifungal activity of the plant extract was tested using well diffusion method as suggested by (B. Srinivasulu et al. 2001). The prepared culture plates were inoculated with different bacteria and fungus by using plate method. Wells were made on the agar surface with 8 mm cork borer. The extracts were poured into the well using sterile borer. The plates were incubated at $37\pm 2^{\circ}$ C for 24 hours for bacterial activity and 48 hours for fungal activity. The plates were observed for the zone formation around the wells. The zone of inhibition was calculated by measuring the diameter of the inhibition zone around the well (in mm). The readings were taken in three different fixed directions in all 3 replicates and the average values were tabulated.

Preparation of Mushroom Extracts:

One hundred grams of *Trametes versicolor* fruit body powder was added with 500 ml of methanol. Extraction with solvent methanol was done for 48 h at room temperature. After filtering through filter paper (Whatmann No.1), the supernatant of the methanol extract was recovered. This process was repeated twice and the solvent from the supernatant was evaporated in a rotary vacuum evaporator to obtain the crude extract. These extract was stored at 4° C until used for evaluating the antimicrobial activity.

RESULTS AND DISCUSSION

The methanol and aqueous extract of the *Trametes versicolor* fruit body were screened against four pathogenic bacteria and four fungal pathogens for antibacterial and antifungal activities by agar well diffusion method which showed the zone of inhibition.

Table 1. Inhibition zone of methanol and aqueous extracts (in mm) of fungal fruit bodies against bacterial pathogens.

| Organism | Methanol Extract | Methanol | Aqueous Extract | Standard (Streptomycin) |
|-------------------------------|------------------|----------|-----------------|-------------------------|
| <i>Staphylococcus aureus</i> | 20 | 06 | 02 | 25 |
| <i>Bacillus subtilis</i> | 19 | 05 | - | 26 |
| <i>Escherichia coli</i> | 18 | 04 | 02 | 24 |
| <i>Pseudomonas aeruginosa</i> | 20 | 04 | - | 23 |

Table 2. Inhibition zone of Methanol and Aqueous extracts (in mm) of fungal fruit bodies against fungal pathogens.

| Organism | Methanol extract | Methanol | Aqueous extract | Standard (Nystatin) |
|-------------------------------|------------------|----------|-----------------|---------------------|
| <i>Aspergillus fumigatous</i> | 14 | 04 | 02 | 24 |
| <i>Aspergillus niger</i> | 14 | 04 | - | 23 |
| <i>Penicillium sp.</i> | 12 | 03 | - | 20 |
| <i>Aspergillus flavus</i> | 13 | 02 | 02 | 26 |

The zone of inhibition against various types of pathogenic bacteria and fungi was shown in (Table: 1) and (Table: 2). Methanol and aqueous extract were effective against both bacteria and fungi and aqueous extract was showed less activity against bacteria and fungi comparatively with the methanol extract. However, the data indicates that the extract prepared in organic solvent consistently displayed better antimicrobial activity than that of the aqueous extract.

Among the two extracts prepared, highest antibacterial activity (20 mm Zone of inhibition) was exhibited by the methanol extract against *Staphylococcus aureus*, *Pseudomonas aeruginosa* followed by *Bacillus subtilis* and *Escherichia coli*. There was no activity observed in the aqueous extract against *Bacillus subtilis* and *Pseudomonas aeruginosa* figure 1.

In case of anti fungal activity, highest antifungal activity (14 mm) was exhibited by the methanol

Figure 1: Antibacterial activity of methanol and aqueous extracts of *Trametes Versicolor* against Gram positive & Gram negative bacteria.

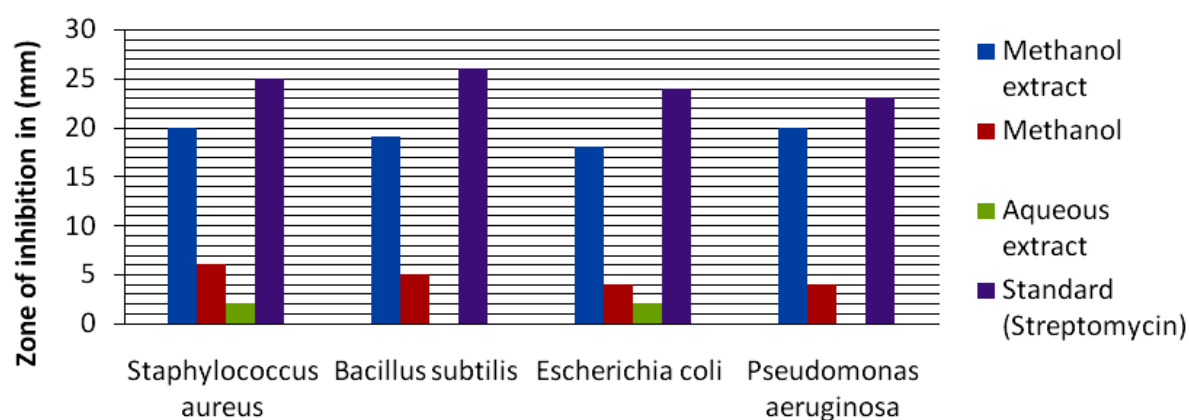
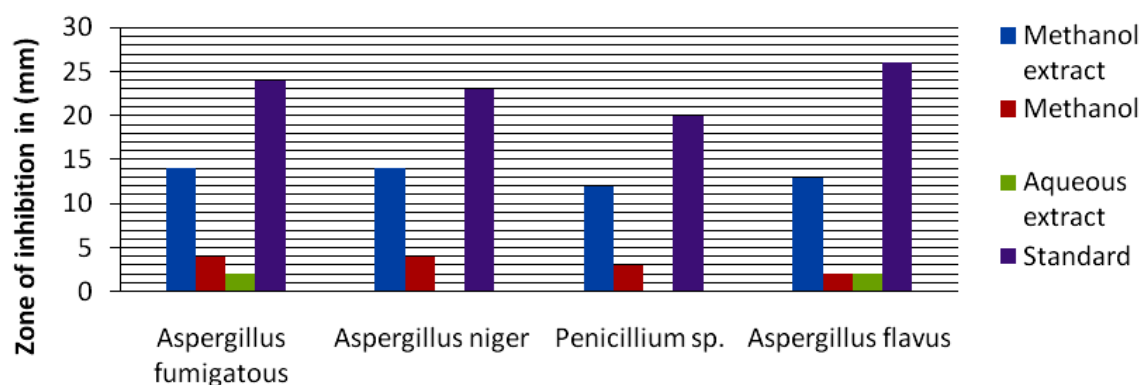


Figure 2: Anti fungal activity of methanol and aqueous extracts of *Trametes Versicolor* against fungal pathogens.



extract against *Aspergillus fumigatus*, *A. niger* followed by *A. flavus* and *Penicillium sp.* There was no activity observed in the aqueous extract against *Aspergillus niger* and *Penicillium sp* figure 2.

Metabolites obtained from microbes and their derivatives play an important role in the development of medicines. The use of these metabolites has grown extensively over the past century, starting with the Fleming's discovery of penicillin (1924). Metabolites obtaining from fungi were the main targets of antimicrobial screening these studies were interrupted for a short time by Waksman's discovery of streptomycin (1945) originating from Actinomycetes. However, in recent years the trend has changed and fungal metabolites have again attracted the attention of pharmacological research. Mushroom based products either from the mycelia or fruiting bodies are consumed in the form of capsules, tablets or extracts (B. Nitha *et al.*, 2006).

In current study, the selected macrofungus showed antibacterial and antifungal activity in high level which was screened against the selected bacterial and fungal strains. The methanol extract showed the maximum inhibition and the aqueous extract showed less zone of inhibition.

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