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Solvent Based Effectiveness of Antibacterial Activity of Edible Mushroom *Lentinus tuberregium* (Fr.)

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Abstract: In this study, *in vitro* antimicrobial properties of *Lentinus tuberregium* culture filterate extracted using four different solvent systems (Hexane, Dichloromethane, Chloroform and Ethyl acetate) were tested for antimicrobial activity. The activity was evaluated by well diffusion tests using bacteria and yeasts. Vancomycine and fluconazole were used as positive controls for bacteria and yeasts, respectively. The crude extracts of *Lentinus tuberregium* have relatively high antimicrobial activity. Among the four organic extracts ethyl acetate extract showed more effective and inhibited the growth of human pathogenic bacteria and yeast.
Key words: Antimicrobial activity, Macro fungi, Submerged Mycelia.

Introduction

It has been known that macro fungi are used as a valuable food source and traditional medicines since Greek and Roman antiquity [1]. Dioscorides, first century Greek physician, knew that *Laricifomes* (*Fomitopsis*) *officinalis* (Vill.) Kotl. & Pouzar (*Fomitopsidaceae*) can be used for treatment of "consumption", a disease now known as tuberculosis [2]. It is believed that mushrooms need antibacterial compounds to survive in their natural environment. Antimicrobial compounds could be isolated from many mushroom species and some proved to be of benefit for humans [3]. In early studies performed by Anchel, Hervey and Wilkins in 1941, diverse antibiotic activity was detected in basidiocarp or mycelia culture extracts of more than 2000 fungal species [4] In recent *in vitro* studies, screening for the antimicrobial activity of basidiomycete strains, some studies were done both of in basidiocarp and in submerged culture. Antimicrobial activities of basidiomycete strains from different countries were screened in submerged culture [4, 10, 11]. Similarly, Rosa et al. (2003) detected 14 mushroom isolates with significant activity against one or more of the target microorganisms [4]. Zjawiony (2004) observed that 75% of polypore fungi that have been tested show strong antimicrobial activity [7]. Antibacterial activities of mushroom exopolysaccharides such as lentinan (from *Lentinus edodes*), schizophyllan (from *Schizophyllum commune*) and PSK ("Polysaccharide Kureha" from *Trametes versicolor*) have also been reported [2, 8-11]. In this context, the antimicrobial activities of submerged mycelia of newly isolated mushroom strain are reported here.

Determination of antimicrobial activity

In vitro antimicrobial susceptibility studies were performed using the following strains; *Bacillus subtilis* (M-441), *Staphylococcus aureus* (M-96), *Micrococcus luteus* (M-1541), *Escherichia coli* (ATTC 25992), *Candida albicans* (M-227), *Salmonella flerineri* (M-1457) *Salmonella typhi* (M-733) (isolate obtained from MTCC and ATCC Chandighar). Antimicrobial activities of all extracts and fractions were screened by the well diffusion method.

Agar well method

Test microorganisms were activated in Mueller Hinton Broth (37°C, 150 rpm, 24 h). The Mc Farland (No: 0.5) standard is used to adjust the

turbidity to prepare inoculum from overnight grown bacteria and yeast cultures. 100 µl of crude extract (100 mg/ml) were added to each well (6 mm diameter holes cut in the agar gel). The plates were incubated at 37°C for 24 h for bacteria, and at 30°C for 48 h for yeasts. Antimicrobial activity was determined by measuring the radius of the clear inhibition zone around each well [12]. Standard antimicrobial agents, vancomycine and fluconazole (30 µg/disk) were used as positive controls for bacteria and yeasts, respectively. Disks injected with 20 µL of 20% DMSO were observed as negative control. Each experiment was replicated three times and the results were expressed as average values.

Results and Discussion

In this study, antibacterial activity of submerged mycelia newly isolated mushroom strain against test microorganisms was compared with the results of positive control antibiotics of vancomycine and fluconazole. The antibacterial activity of mushroom sample varied according to the solvents. Different solvent extracts of this strain were active against bacteria. These extracts were the most active to inhibit the growth of *Salmonella flexineri*, *Micrococcus luteus* and *Salmonella typhi*. In the present study, *Lentinus tuberregium* showed activity against some of the studied test microorganisms (Table 1). The activities of *Lentinus tuberregium* were higher than positive control (vancomycine or fluconazole) against *Salmonella typhi, Micrococcus luteus*, and *Salmonella flexineri.* But, in the present study, our *Lentinus tuberregium* strain did not show any activity against *E.coli.*

These combine activity of antibacterial increase the chance of the mushroom for medicinal purposes. The fact that the Basidiomycetes have been insufficiently investigated coupled with the broad range of structural types of antibiotics. However, basidiomycetes may be a source of new and useful bioactive compounds. To our knowledge, no investigation has been performed for comparing antimicrobial activity potential of basidiomycetes strains in different life forms. Further studies on isolation and identification of the active compounds may provide a better source for developing new therapeutic agents.

Table-1

Pathogens	Zone of inhibition (mm in diameter)			
	Hexane	**Dichloromethane**	**Ethyl acetate**	**Chloroform**
S.aureus	9	9	11	10
M.luteus	-	10	12	9
P.aureginosa	8	10	8	8
E.coli	-	11	6	7
S.typhi	8	10	12	9
S.flexneri	10	9	13	8

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