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Research Article

ANTIBACTERIAL ACTIVITY AND HEAVY METAL ACCUMULATION OF EDIBLE OYSTER MUSHROOM (*PLEUROTUS SAJOR-CAJU)* GROWN ON TWO SUBSTRATES

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ABSTRACT

The present study aims to investigate the antibacterial activity and heavy metal accumulation of edible oyster mushroom (*Pleurotus Sajor-Caju)* grown on two substrates *Viz.* Paddy Straw and Sugar cane bagasse. Cultivation of mushroom was tried with two different substrates. The disc and well methods were employed using various microorganisms. The studies of antimicrobial activity were based on the zone of inhibition of microbial growth around the disc was found in the range of 0.62±0.27 to 1.760±0.769. In Well method the zone of inhibition was found in the range of 0.632±0.282 to 2.027±0.9065 in the extracts of ethanol and acetone against selected micro organisms. Heavy metals (Lead and Zinc) accumulation was estimated *Viz :* 18.75ppm and 83.38ppm of lead, 33.77ppm and 49.97ppm concentrations of zinc were found both in *Pleurotus* obtained from paddy straw and sugarcane bagasse. The present study aims to introduce cheap substrates for the cultivation of *Pleurotus* Spp in will be helpful to the people with marginal income using this technology. And this study can also be used to develop new drugs from these mushrooms.

Keywords: Antibacterial activity, Heavy metals, *Pleurotus sajor-caju,* Solvent extracts

INTRODUCTION

Edible mushrooms are nutritionally endowed fungi (mostly Basidiomycetes) that grow naturally on the trunks and roots of trees as well as on decaying woody materials [1, 2]. *Pleurotus* species have been used by the people all over the world for their nutritional, medicinal and other beneficial values [3]. Oyster mushrooms are a good source of dietary fiber and other valuable nutrients [4]. The fruiting body of the mushroom is also a potential source of lignin and phenol degrading enzymes [5, 6] analyzed the hypocholesterolemic and antherogenesis inhibition functions in rabbits and rats courtesy of its mycelia secretary function. The mushroom is credited to the third largest macrofungus cultivated for food and industrial purposes world wide. Oyster mushrooms were found to contain antimicrobial and antioxidant potentials [7] .Heavy metal concentrations in mushrooms are considerably higher than those in agricultural crop plants, vegetables and fruits [8]. The fruiting body of the mushroom is also a potential source of lignin and phenol degrading enzymes. The agro industrial solid residues can be utilized for the cultivation of *Pleurotus* spp as it is an environmental protection strategy [9]. The present study is used to carried out antibacterial and heavy metal potential of edible oyster mushroom *Pleurotus sajor-caju* grown on two substrates.

MATERIAL AND METHODS

This study was conducted in the Mushroom Research Centre at Department of Botany, Ayya Nadar Janaki Ammal College Sivakasi during the year 2009-2011.

Preparation of Spawn

A pure culture of *Pleurotus sajor-caju* (Mother spawn) was obtained from the Agricultural College and Research Centre, Madurai, Tamil Nadu, India. The Spawn was prepared from the mother spawn using sorghum grains as substrate. The inoculated grains were incubated at room temperature. After the grains were fully colonized by the mycelia, they were kept in a room temperature (22-30^{0} C).

Substrates of Paddy Straw and Sugar cane bagasse were obtained local area and the materials were cut into pieces (2-4 inches) and the prepared piece bulk was soaked it in cool water for 24 hours, then it was boiled in water for an hour. The substrate material was filtered and dried in a shade to retain 65% moisture content.

The dried substrates of paddy straw and bagasse were packed in 30X60cm polythene bags separately and it is called as a seed bed for cultivating the mushrooms (Polythene bag method). The transparent polythene bag and perforated polythene bags were the best container system for the cultivation of *Pleurotus* spp [10] .

The spawn was spread over the substrates in a polythene bag at 15cm height on first layer. Then these spawn was spread over the substrate for about 4 layers at a height of 10cm height and finally the top layer was covered with the substrates. Twine was used to tie the polythene bag mouth, there are four wholes made on the sides of polythene bag used to sprout the mushrooms, and then it was collected for further analysis.

Assessment of Antibacterial activity

The collected mushrooms were dried in an oven at 15^{0}C for 4 hours[11]. Then it was powdered using mixer grinder. This powder was mixed with ethanol and acetone separately for 24 hours and then extracted using soxhlet apparatus, and then the filtrate was used for the experiment.

The antibacterial activity of the edible oyster mushroom *Pleurotus Sajor-caju* against the pathogens like *Bacillus subtilis, Klebsiella pneumoniae, Pseudomonas aeruginosa* and *Escherichia coli* were analyzed in disc diffusion method [12] and well diffusion method (Modified method of [13].

Analysis of heavy metals

The heavy metals (Lead and Zinc) accumulations in the mushrooms were analysed by using AAS (Atomic Absorption Spectroscopy – Model AA- 6300 Shimadzu, Japan) described by [14].

Fresh Samples of mushrooms from each substrate were collected and dried in an oven at 40^{0}C for 4 hours and powdered. 0.5-2gm of dry powder dissolved in 2ml of concentrated HNO_3 and 1 ml of distilled water and it was used as a test solution to analyze the heavy metals (Zinc and Lead) using AAS.

Statistical Analysis

All the experimental values were reported and expressed as means of SD±SE of five values. Data were evaluated using one way analysis of variance.

RESULTS

Disc Diffusion Method

The present study revealed that the antimicrobial activity against human pathogenic microorganisms in the ethanol and acetone extracts of *Pleurotus sajor-caju* grown on two different substrates. In disc diffusion method in ethanol, *Bacillus subtilis* (0.748±0.34) and *E.coli* (0.62±0.27) were more susceptible against *Pleurotus* spp grown on two substrates. Where as *Klebsiella pneumoniae* contains maximum antibacterial activity in the mushroom grown on sugar

cane bagasse (Table 1). In the acetone extracts shows maximum zone of inhibition was found against *Klebsiella pneumoniae* (1.720±0.769) in *Pleurotus* grown on sugar cane bagasse and minimum zone of inhibition against *Pleurotus* grown on paddy straw (Table 2).

Well diffusion method

In the well diffusion method in ethanol solvent shows maximum zone of inhibition against *Pseudomonas aeruginosa* (3.34±1.49) and minimum zone of inhibition against *Bacillus subtilis*(0.8±0.35) both from *Pleurotus* grown on sugar cane bagasse (Table 1). However, in acetone extracts, the maximum (1.469±0.656) and minimum zone of inhibition (0.632 ± 0.282) was found against *Klebsiella pneumonia* *Pseudomonas aeruginosa* found in *Pleurotus* obtained from bagasse (Table 2).

Heavy metal analysis

The ppm level of heavy metals (Zinc & Lead) in *Pleurotus* mushrooms obtained from paddy straw and bagasse were analysed. Among theses two metals (Lead and Zinc) were found in the mushrooms obtained from both substrates. However, the lead concentration was found more (6.754±3.020) in *Pleurotus* obtained from bagasse and minimum (3.15±1.671) in *Pleurotus* from paddy straw. Where as the concentration of zinc found to be maximum in *Pleurotus* from paddy straw (16.676±7.45) and minimum in *Pleurotus* from bagasse (9.994± 4.469).

Table 1: Antibacterial activity of *Pleurotus sajor- caju* in disc and well diffusion method on ethanol extracts

Micro organisms	*Pleurotus* from Paddy Straw		*Pleurotus* from Bagasse	
	Disc Method	Well Method	Disc Method	Well Method
Bacillus subtilis	1.414 ±0.63	1.414 ±0.63	0.748±0.334	0.8±0.357
Escherichia coli	0.62 ± 0.27	1.1135±0.497	0.9197±0.438	1.2±0.536
Klebsiella pneumoniae	1.414±0.63	1.01±0.456	2.027±0.9065	0.8±0.357
Pseudomonas aeruginosa	1.03±0.40	1.09±0.48	0.7683±0.3346	3.34±1.49

Values are the mean value of (SD±SE) n =5

Table 2: Antibacterial activity of acetone extracts of *Pleurotus sajor-caju*. in disc and well diffusion method

Micro organisms	*Pleurotus* from Paddy Straw		*Pleurotus* from Bagasse	
	Disc Method	Well Method	Disc Method	Well Method
Bacillus subtilis	1.166±0.521	1.414±0.632	1.8±0.80	0.894±0.399
Escherichia coli	1.16±0.521	1.378±0.616	1.326±0.593	0.88±0.393
Klebsiella pneumoniae	0.8±0.357	0.748±0.334	1.720±0.769	1.469±0.656
Pseudomonas aeruginosa	1.039±0.464	0.8±0.357	1.01±0.451	0.632±0.282

Values are the mean value of (SD±SE) n =5

DISCUSSION

Medicinally important wild mushrooms were cultivated artificially and they were yielding a diverse source of products[15, 16]. Mushrooms species were sought and cultivated by humans for several purposes like consumption and health benefits[17]. In the present study the cultivation of *Pleurotus sajor-caju* with two substrates i.e paddy straw and sugar cane bagasse was studies. The antimicrobial activity of *Pleurotus sajor-caju* tested against pathogenic microorganisms. The results pronounce similar with the findings of [11] in *Pleurotus eryngii* grown on various substrates. [18] showed that *Pleurotus ostreatus* held highest antimicrobial activity against *Bacillus subtilis, Escherichia coli and Saccharomyces cerevisiae* in petroleum ether extract than the acetone extract. In the present study of *Pleurotus sajor-caju* obtained from paddy straw and sugarcane bagasse shows maximum antimicrobial activity against *Bacillus subtilis* followed by *Klebsiella pneumonia*, this results were also co inside with the findings of [19,20]. The mushrooms exhibited high level of antimicrobial activity due to the presence of secondary metabolites like terpenoids, alkaloids and phenols [21] and various immune enhancing activities [22]. The occurrence of heavy metals like zinc and lead in the mushrooms were analysed in the present study. The concentration of lead was more in *Pleurotus sajor caju* obtained from paddy straw than sugarcane bagasse. This result was co inside with the findings of [23].

The overall results obtained from the present study the antimicrobial activity of *Pleurotus sajor-caju* was evidenced against the tested micro organisms. It may be utilized as one of the resource to control pathogenic forms. As per our studies heavy metals like zinc and lead were also analysed in the *Pleurotus sajor caju* raised from paddy straw and sugarcane bagasse. The concentration of zinc found to be more when compared to the metal lead in the fruit bodies of *Pleurotus sajor-caju*. The concentration levels of heavy metals are found to be below the toxic/tolerable levels and not harmful to humans.

CONCLUSION

The obtained results may be useful in further identification and production of antibacterial compounds, enzymes etc from the edible mushrooms. Further studies going on to produce these antibacterial substances from the various non edible mushrooms like *Ganoderma lucidum, Polyporus spp., and Schizophyllum commune*. etc. Commercial production and marketing of these mushrooms create self employment to rural peoples.

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