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Research Article

Exploring the Antimicrobial Properties of Some Selective Mushroom Mycoflora

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Abstract

Mushrooms have long been recognized for their nutritional and medicinal properties, but their potential as sources of antimicrobial agents remains underexplored. This study investigates the antimicrobial activity of various wild mushroom species against selected bacterial and fungal pathogens. Specifically, the study focused on the inhibition zones of *Staphylococcus aureus*, *Escherichia coli*, *Candida albicans*, *Fusarium oxysporum*, and *Fusarium proliferatum* when exposed to extracts from twelve different mushroom species, including *Schizophyllum commune*, *Laeiporus sulphureus*, *Pleurotus pulmonarius*, and others. The results revealed significant antibacterial and antifungal activities, with *Schizophyllum commune* showing the highest inhibition against *Fusarium oxysporum* (71.42 ± 0.28 mm) and *Microporus xanthopus* demonstrating strong inhibition against *Staphylococcus aureus* (8.46 ± 0.11 mm). Additionally, *Pleurotus sajor-caju* exhibited notable antifungal activity against *Fusarium proliferatum* (76.06 ± 0.14 mm), suggesting its potential as a source of novel antifungal compounds. The study observed a varied range of inhibition zones across different mushroom species, emphasizing the diverse antimicrobial potential within mushroom mycoflora. These findings underscore the importance of further research into the bioactive compounds of mushrooms, which could contribute to the development of new antimicrobial agents. The study not only highlights the significance of mushrooms in combating microbial resistance but also opens avenues for their inclusion in future pharmaceutical applications.

Keywords

Antibacterial, Antifungal, Inhibition Zone, Macrofungi, Medicinal Property

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1. Introduction

Mushrooms, a diverse group of fungi, have been revered for centuries for their culinary delights and medicinal properties. Beyond their nutritional value, mushrooms have gained attention in recent years for their potential therapeutic applications, particularly in combating microbial infections [1]. The mycoflora of mushrooms, comprising a rich assortment of fungal species, has emerged as a promising source of bioactive compounds with antimicrobial properties [2-4]. The rise of antimicrobial resistance poses a significant threat to global public health, underscoring the urgent need for novel antimicrobial agents [5]. Natural products, particularly those derived from fungi, have garnered increasing interest as potential alternatives to conventional antibiotics. Among these, mushroom mycoflora stands out for its diverse array of secondary metabolites, many of which exhibit potent antimicrobial activity [1, 6-8].

One of the key bioactive compounds found in mushroom mycoflora is β-glucans, polysaccharides known for their immunomodulatory and antimicrobial effects [9, 10]. These compounds have been extensively studied for their ability to enhance the body's defense mechanisms and inhibit the growth of various pathogens, including bacteria, fungi, and viruses. Moreover, β-glucans have shown promise in mitigating inflammation and supporting wound healing processes [3, 11, 12]. In addition to β-glucans, mushroom mycoflora produces a myriad of other bioactive compounds, such as phenolic compounds, terpenoids, lectins, and peptides, each with its own unique antimicrobial properties [10]. For instance, phenolic compounds like flavonoids and phenolic acids exhibit antioxidant and antimicrobial activities, while terpenoids such as ganoderic acids demonstrate antiviral and antifungal properties [13, 14]. Lectins, on the other hand, have been shown to agglutinate bacteria and inhibit their growth, making them potential candidates for the development of novel antimicrobial agents [15].

The antimicrobial efficacy of mushroom mycoflora has been demonstrated against a wide range of pathogens, including multidrug-resistant bacteria like Methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *Enterococcus* (VRE), as well as pathogenic fungi like Candida albicans and Aspergillus fumigatus. Moreover, studies have indicated synergistic effects between mushroom mycoflora extracts and conventional antibiotics, suggesting their potential use in combination therapy to enhance antimicrobial efficacy and overcome resistance [16, 17]. Despite the promising potential of mushroom mycoflora as a source of antimicrobial agents, much remains to be explored regarding their mechanisms of action, safety profiles, and clinical applications [15, 18]. Further research is needed to elucidate the specific bioactive compounds responsible for their antimicrobial activity, optimize extraction and purification methods, and evaluate their efficacy in preclinical and clinical settings [19]. In this study, we aim to evaluate the antimicrobial properties and bioactive compounds of mushrooms, highlighting their potential applications in combating microbial infections and addressing the global challenge of antimicrobial resistance [20]. By harnessing the therapeutic potential of these natural resources, we may pave the way for the development of novel antimicrobial agents with improved efficacy and safety profiles.

Mushrooms are a group of fungi that has fruiting bodies and are of great importance in the production of extracellular enzymes, agricultural and biotechnological aspects [19]. They are mostly known as food supplements due to their high nutritional contents [1, 19, 21, 22]. Apart from the above-mentioned applications of mushroom, it also has a medicinal importance with its antioxidant and antimicrobial activities [23, 24], immune enhancer [25-27], and are effective for the treatment of diabetes [28, 29] and various types of cancer [30, 31]. Mushrooms, the macrofungi of the fungal kingdom, have captivated human interest for centuries due to their unique nutritional as well as culinary and medicinal properties. Beyond their delightful flavors and textures, mushrooms harbor a treasure trove of bioactive compounds, making them a subject of profound scientific inquiry [1, 4, 8]. From the time immemorial mankind has used traditional medicines for human healthcare with terrestrial plants occupying a significant therapeutic role [32]. Recently, the World health Organization has estimated that approximately 80% of the world's inhabitants still depend on traditional (herbal and fungal) medicines for primary health purposes [33]. Historically, hot-water-soluble fractions (decoctions and essences) from medicinal mushrooms, that is, mostly polysaccharides, were used as medicine in the Far East where knowledge and practice of mushroom was primarily originated [34, 35]. Mushrooms such as *Ganoderma lucidum* (Reishi), *Lentinus edodes* (Shiitake), *Inonotus obliquus* (Chaga) and many others have been collected and used for hundreds of years in Korea, China, Japan and Eastern Russia. Those practices still form the basis of modern scientific studies of fungal medicinal activities. It is notable and remarkable how reliable the facts collected by traditional Eastern medicine are in the study of medicinal mushrooms. The number of mushrooms on earth is estimated at 1,40,000, yet may be only 10% (approximately 14,000 named species) are known. Mushrooms comprise a vast and yet largely untapped source of powerful new pharmaceutical products [34-40].

2. Material and Methods

The samples of mycoflora were collected from Holy Ayodhya region famous for birthplace of Godes Rama. The survey results the collection of macrofungal species which belonging to the different families [41, 42]. Selectively a total of 10 species of macrofungi taken for the evaluation of antimicrobial properties. The antimicrobial activity analysis done by using the previous methods [43, 44]. Fresh collected sample of macrofungi were cut into slice and air dried at 50 ℃ in an oven to constant weight. After that, a fine powder obtains by grinding the samples. A 5 grams sample of each dried mushroom was extracted by stirring with 50 mL of either distilled water or ethyl acetate in a beaker and was continuously shaken for 96 hours to dissolve the constituents' phytochemicals such as tannins, flavonoids, alkaloids, terpenoids and others. The aqueous solution was filtered using filter paper. Qualitative determination of phytochemicals compounds of macrofungi were done by followed the previous methods [3].

2.1. Test Organisms for Antimicrobial Activities

Antimicrobial activities were evaluated with two bacterial pathogens (*Escherichia coli*; *Staphylococcus aureus*), two fungal pathogens (*Fusarium proliferatum*; *Fusarium oxysporum*) and a yeast pathogen (*Candida albicans*). All the tested pathogens were acquired from the Microbial Type Culture Collection (MTCC), Ayodhya, India and were maintained at Microbiology Laboratory, Department of Microbiology, Dr. Ram Manohar Lohia Avadh University on a specific media as per instructions.

2.2. Evaluation for Antifungal Potential

All the isolates were evaluated for their *in vitro* antagonistic potential using dual culture method [45, 46]. Briefly, one 5 mm agar block of fully grown fungal pathogen was placed at the centre of the petridish which contain PDA, and mushroom isolates were kept at the periphery of the plates. Petridishes were incubated at 28 ℃ for 7 days and diameter of the fungus growth was measured and compared with control. All the experiments were carried out in triplicate and the percentage of inhibition was calculated using the following formula:

$$\text{Inhibition } (\%) = \frac{\text{CT}}{\text{C}}\, x\, 100$$

Where, C is the fungal pathogen control and T is the distance of the tested pathogen with the sample.

2.3. Evaluation for Antibacterial Potential

Antibacterial activity was evaluated by modified agar well diffusion method [47, 48]. Briefly, an aliquot of the tested bacterial pathogens (50 μL) was spread on the surface of the modified LB agar plates using sterile L spreader and 6 mm diameter wells was prepared by using sterile cork borer. The wells were then filled with 100 μL (1×10^4 CFU/mL) of cell free extract of mushroom isolate and the plates were incubated at 37 ℃ for 24 h. All experiments were carried out in triplicates and the zone of inhibition was measured in mm.

3. Results and Discussion

A total of 10 selected species of mushrooms isolates were grown on artificial media and evaluated their antimicrobial properties. The *in vitro* antagonistic activities of the isolates were tested against the selected fungal plant pathogens and the result confirmed diverse antagonistic effects. Of all the isolates, there were maximum isolates showing positive growth inhibitory activity against no less than one or two fungal plant pathogens. The growth inhibitory percentage ranged from 4.26% to 78.33% (Table 1). All the isolates evaluated exhibit positive antifungal activity against *F. oxysporum* except one isolate (*Auricularia auricula judae*). On the other hand, only 4 isolates (*Schizophyllum commune*, *Auricularia auricula judae*, *Microporus xanthopus*, *Xylaria hypoxylon*) showed positive activity against *C. albicans*. The isolates including *Schizophyllum commune*, *Microporus xanthopus* and *Xylaria hypoxylon* exhibited positive antifungal activity against all the pathogens tested in this study. The highest antifungal activity was observed with the isolate *Microporus xanthopus* against *Fusarium proliferatum* with 78.33%; followed by *Pleurotus djamor* with 76.66% against *F. proliferatum* and *Xylaria hypoxylon* against *F. oxysporum* with 66.06% (Figures 1-4).

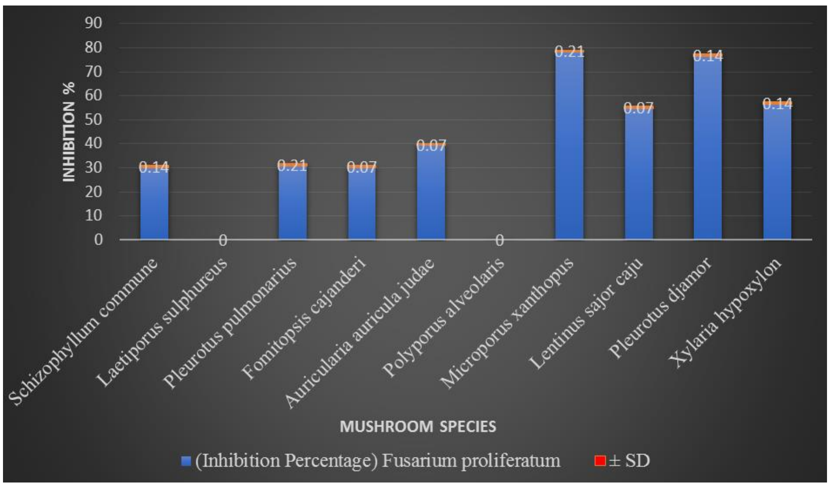

Figure 1. *Antifungal Activity of Wild Mushrooms Against Fusarium proliferatum.*

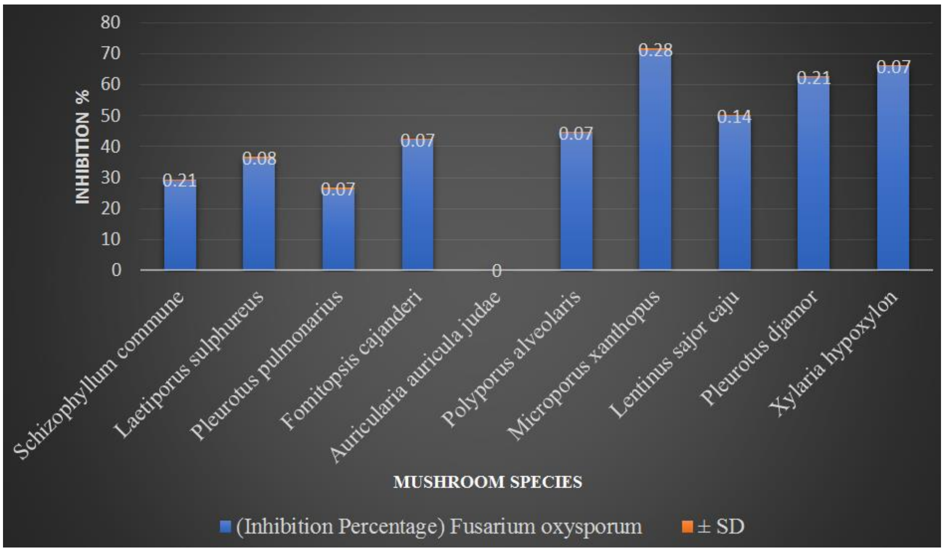

Figure 2. *Antifungal Activity of Wild Mushrooms Against Fusarium oxysporum.*

Figure 2 shows the inhibition percentages of *Fusarium oxysporum* by various mushroom species, along with their standard deviations (SD). *Lentinus sajor caju* shows the highest inhibition percentage (~28%), suggesting its strong antifungal activity against *Fusarium oxysporum*. While other mushrooms, such as *Schizophyllum commune* and *Pleurotus djamor*, exhibit moderate inhibition percentages (~21%). *Polyporus alveolaris* demonstrates no measurable inhibition activity.

Among the mushrooms tested, *Lentinus sajor-caju* showed the strongest antifungal activity, exhibiting the largest inhibition zone, indicating its potent ability to inhibit *Candida albicans* growth. *Pleurotus djamor* also demonstrated significant antifungal effects, although its inhibition zone was smaller compared to *Lentinus sajor-caju*. Both *Schizophyllum commune* and *Polyporus alveolaris* exhibited moderate activity, with smaller inhibition zones. In contrast, *Ganoderma lucidum* and other species showed minimal to no measurable inhibition, suggesting limited or negligible antifungal properties. These findings highlight the potential of certain wild mushrooms, particularly *Lentinus sajor-caju*, as sources of natural antifungal compounds against *Candida albicans*.

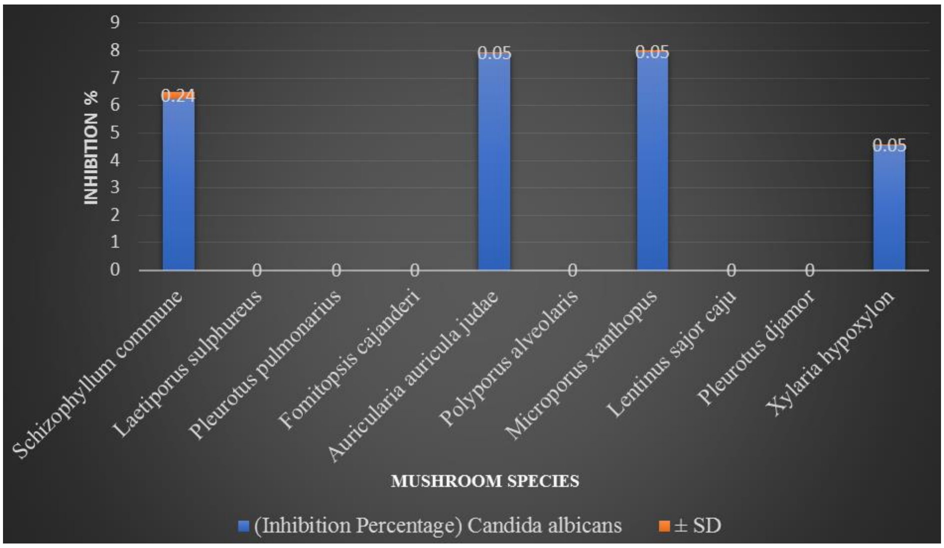

Figure 3. *Antifungal Activity of Wild Mushrooms Against Candida albicans.*

Figure 4 compares the antifungal activity of different mushroom species against various fungal pathogens. The findings indicate significant variation in the inhibitory effects of the mushrooms, with some species showing broad-spectrum antifungal properties while others exhibit more targeted activity. Certain mushroom species, such as *Lentinus sajor-caju* and *Pleurotus djamor*, displayed strong antifungal activity across multiple pathogens, indicating their potential as natural sources of antifungal agents. In contrast, other species, like *Polyporus alveolaris* and *Schizophyllum commune*, showed more limited inhibition or were effective against specific pathogens only. The data highlight the diversity in antifungal potency among wild mushrooms and suggest that certain species could be explored further for their potential in developing natural antifungal treatments.

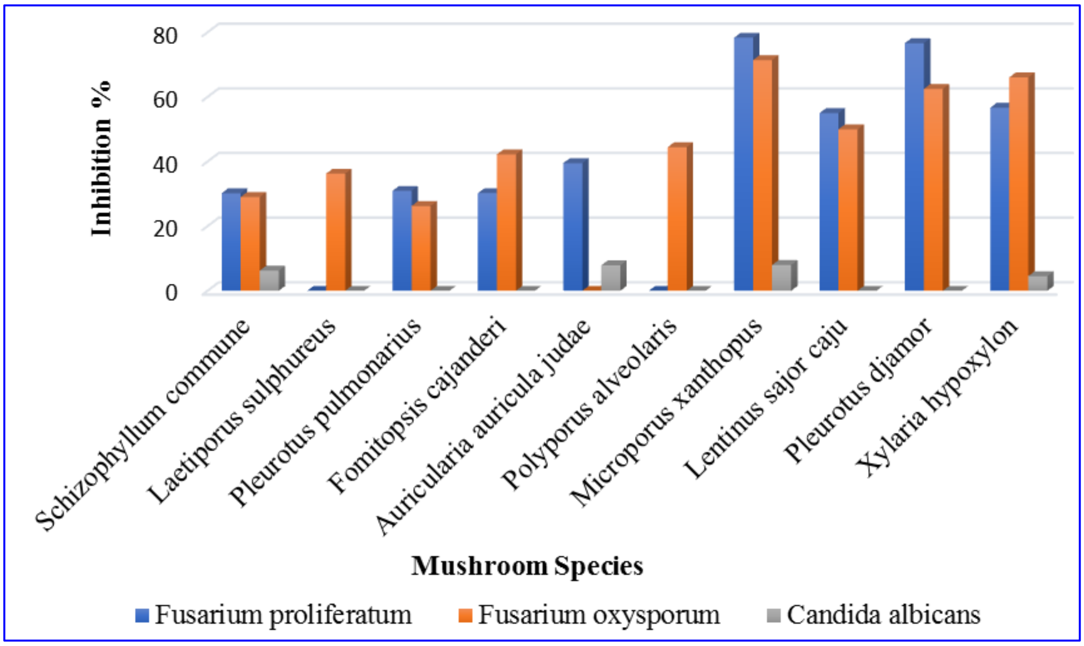

Figure 4. *Comparison of Antifungal Activity of Different Mushroom Species Against Fungal Pathogens.*

Similar to the figure, the table 1 presents a comparative analysis of the antifungal and antibacterial activities of various mushroom species against fungal and bacterial pathogens. Among the tested mushrooms, *Microporus xanthopus* exhibited the highest antifungal activity, with inhibition percentages of 78.33% against *Fusarium proliferatum* and 71.42% against *Fusarium oxysporum*. It also showed moderate activity against *Candida albicans* (7.96%). *Pleurotus djamor* and *Lentinus sajor-caju* demonstrated significant antifungal effects, with inhibition percentages of 76.66% and 55.00%, respectively, against *Fusarium proliferatum*, and 62.49% and 49.99%, respectively, against *Fusarium oxysporum*. In contrast, other species such as *Schizophyllum commune*, *Laetiporus sulphureus*, and *Pleurotus pulmonarius* exhibited more variable or less pronounced antifungal activity. Regarding antibacterial properties, *Schizophyllum commune* showed the strongest inhibition against *Staphylococcus aureus* (9.61 mm), while *Laetiporus sulphureus* and *Pleurotus pulmonarius* displayed moderate antibacterial activity with inhibition zones of 5.50 mm and 8.26 mm, respectively. Other mushrooms, such as *Polyporus alveolaris*, *Fomitopsis cajanderi*, and *Auricularia auricula judae*, demonstrated limited antibacterial effects, particularly against *Escherichia coli* and *Staphylococcus aureus*. Overall, *Microporus xanthopus* and *Pleurotus djamor* emerged as the most potent antifungal species, whereas *Schizophyllum commune* exhibited the strongest antibacterial activity. These findings suggest that certain wild mushrooms possess valuable antimicrobial properties and could serve as potential natural sources for the development of antifungal and antibacterial agents.

Table 1. *Antimicrobial activity of wild mushrooms against selected bacterial and fungal pathogens.*

Mushroom Species	**Antifungal Activity (Inhibition Percentage ± SD)**			**Antibacterial Activity (Inhibition Zone (mm) ± SD)**	
	Fusarium proliferatum	***Fusarium oxysporum***	***Candida albicans***	***Escherichia coli***	***Staphylococcus aureus***
Schizophyllum commune	30.22 ±0.14	29.05 ±0.21	6.25 ±0.24	-	9.61 ±0.02
Laetiporus sulphureus	-	36.26 ±0.08	-	5.50 ±0.26	6.36 ±0.11

Mushroom Species	Antifungal Activity (Inhibition Percentage ± SD)			Antibacterial Activity (Inhibition Zone (mm) ± SD)	
	Fusarium proliferatum	*Fusarium oxysporum*	*Candida albicans*	*Escherichia coli*	*Staphylococcus aureus*
Pleurotus pulmonarius	30.93 ± 0.21	26.23 ± 0.07	-	-	8.26 ± 0.15
Fomitopsis cajanderi	30.23 ± 0.07	42.23 ± 0.07	-	-	6.33 ± 0.15
Auricularia auricula judae	39.53 ± 0.07	-	7.90 ± 0.05	-	4.43 ± 0.20
Polyporus alveolaris	-	44.45 ± 0.07	-	-	4.26 ± 0.15
Microporus xanthopus	78.33 ± 0.21	71.42 ± 0.28	7.96 ± 0.05	-	8.46 ± 0.11
Lentinus sajor caju	55.00 ± 0.07	49.99 ± 0.14	-	4.43 ± 0.20	7.06 ± 0.15
Pleurotus djamor	76.66 ± 0.14	62.49 ± 0.21	-	-	7.60 ± 0.10
Xylaria hypoxylon	56.66 ± 0.14	66.06 ± 0.07	4.53 ± 0.05	-	-

Furthermore, we studied the antibacterial potential of the isolates with the selected bacterial pathogens including *E. coli* and *S. aureus*. In all, only 2 isolates (*Laetiporus sulphureus* and *Lentinus sajor caju*) out of 10 isolates exhibited positive results against both pathogens and these two isolates shows positive inhibitory activity against *E. coli* pathogen. Isolates *Xylaria hypoxylon* only reported as non-inhibitory activities against both tested pathogens. Except isolate *Xylaria hypoxylon*, all nine isolates (*Schizophyllum commune*, *Laetiporus sulphureus*, *Pleurotus pulmonarius*, *Fomitopsis cajanderia*, *Auricularia auricula judae*, *Polyporus alveolaris*, *Microporus xanthopus*, *Lentinus sajor caju* and *Pleurotus djamor*) exhibited positive inhibitory activity against *S. aureus*. The highest inhibitory activity showed by isolates *Schizophyllum commune* (9.61 mm) and followed by *Microporus xanthopus* (8.46 mm) *Pleurotus pulmonarius* (8.26 mm) against tested pathogen *S. aureus* (Figures 5-7).

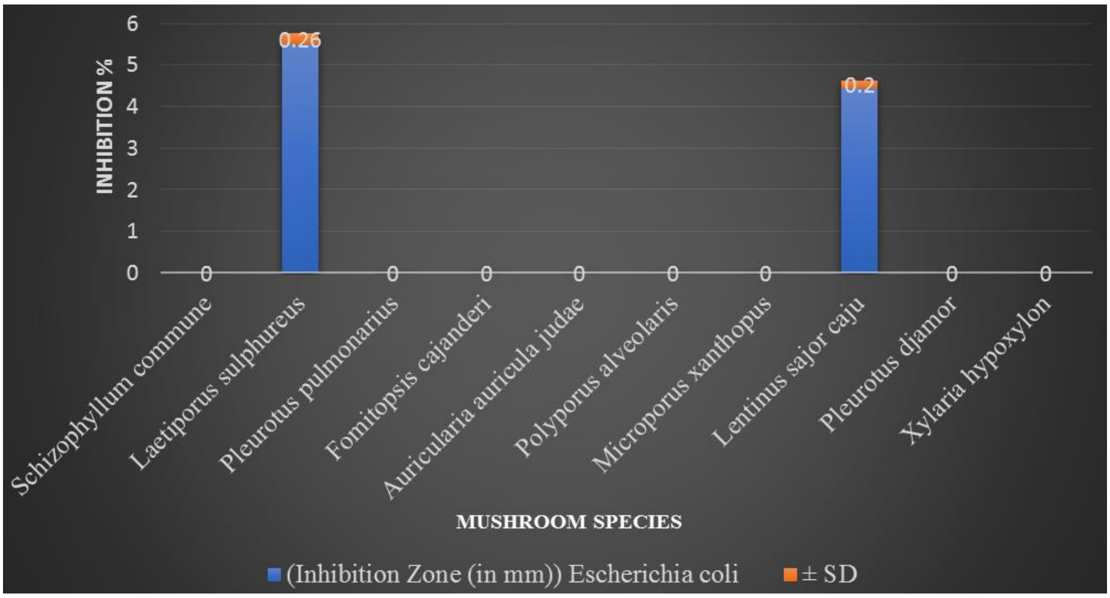

Figure 5. *Antibacterial Activity of Wild Mushrooms Against Escherichia coli.*

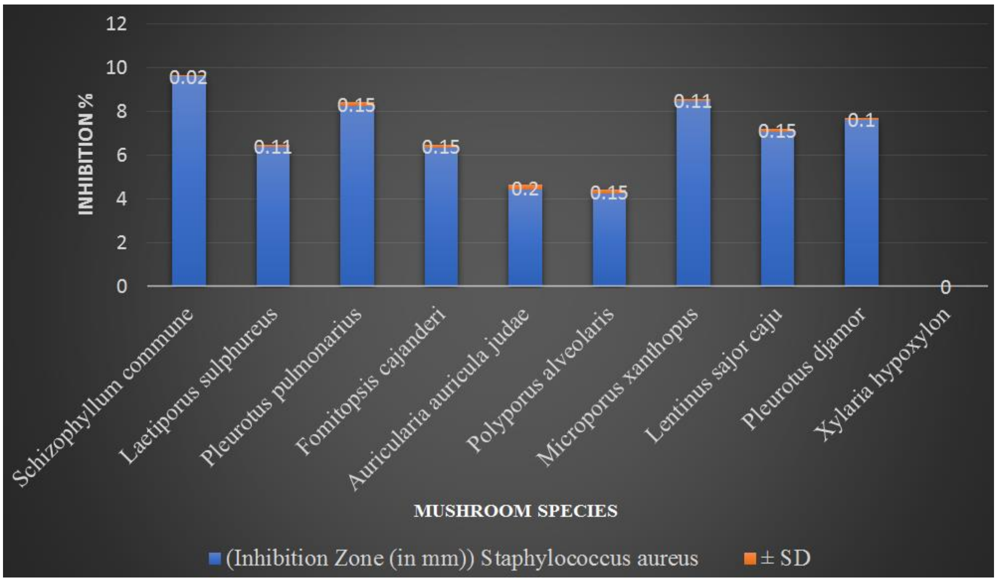

Figure 6. *Antibacterial Activity of Wild Mushrooms Against Staphylococcus aureus.*

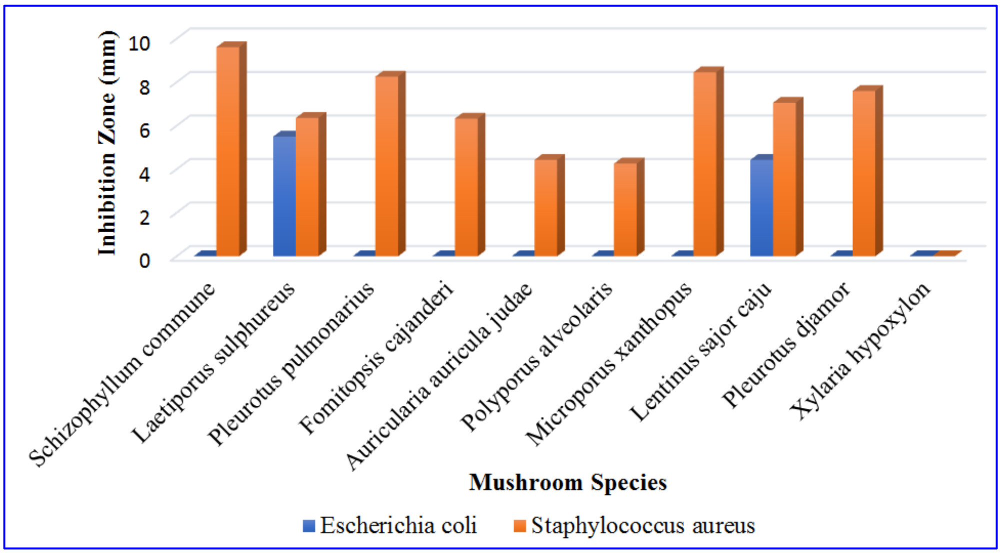

Figure 7. *Comparison of Antibacterial Activity of Different Mushroom Species Against Bacterial Pathogens.*

4. Conclusion

The present study investigated the antimicrobial activity of ten wild mushroom species against a range of bacterial and fungal pathogens, revealing significant potential for some species in inhibiting microbial growth. Among the tested mushrooms, *Microporus xanthopus* exhibited the most potent antifungal activity, particularly against *Fusarium proliferatum* and *Fusarium oxysporum*, with inhibition percentages of 78.33% and 71.42%, respectively. Similarly, *Pleurotus djamor* showed strong antifungal effects, particularly against *Fusarium proliferatum* with a 76.66% inhibition rate. In terms of antibacterial activity, *Schizophyllum commune* and *Pleurotus pulmonarius* demonstrated noteworthy inhibition against *Staphylococcus aureus*, with inhibition zones of 9.61 mm and 8.26 mm, respectively. Conversely, *Laetiporus sulphureus* and *Lentinus sajor-caju* showed limited antibacterial effects against *Escherichia coli*, with inhibition zones of 5.50 mm and 4.43 mm, respectively. The variation in antimicrobial efficacy across different species highlights the selective nature of mushroom-derived compounds in targeting specific pathogens. This study underscores the importance of wild mushrooms as a valuable source of bioactive compounds with significant antimicrobial properties, suggesting their potential application in developing natural antimicrobial agents. Further research, including the isolation and characterization of active compounds, is warranted to fully explore the therapeutic potential of these mushroom species.

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Author Contributions

Balwant Singh: Conceptualization, Investigation, Methodology, Writing – original draft

Sneha Dwivedi: Conceptualization, Investigation, Methodology, Writing – original draft

Mukul Machhindra Barwant: Conceptualization, Data curation, Formal Analysis, Methodology, Resources, Writing – original draft

Vinay Kumar Singh: Conceptualization, Formal Analysis, Funding acquisition, Investigation, Methodology, Writing – original draft

Alok Kumar Singh: Data curation, Formal Analysis, Funding acquisition, Project administration, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing

Shailendra Kumar: Conceptualization, Formal Analysis, Funding acquisition, Methodology, Writing – review & editing

Mudasir Ahmad Dar: Formal Analysis, Project administration, Software, Visualization, Writing – review & editing

Declarations

*Ethics approval and consent to participate***:** This study does not contain any experiments with human participants or animals.

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Data Availability Statement

All data generated or analyzed during this study are included in this published article.

Conflicts of Interest

The authors declare no conflicts of interest.

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Research Fields

Balwant Singh: Mycology, Fungal diversity, Antibiotics, Anti microbials, Mushrooms, Fungus

Sneha Dwivedi: Microbiota, Bacteria, Fungi, Fungus potential, Microbes

Mukul Machhindra Barwant: Plants, Fungi, Bioactive compounds, Fungal diversity, Mushrooms

Vinay Kumar Singh: Microbiota, Fungus species, Fungi, Antimicrobials, Mushrooms

Alok Kumar Singh: Fungi, Fungus species, Microbials, Antimicrobials, Mushrooms

Shailendra Kumar: Geology, Fungus species, GIS, Antimicrobials, Mushrooms

Mudasir Ahmad Dar: Microbes, Gut microbiome, Lignocellulose, Gut bacteria, Symbiosis, Fungal diversity