Explore–Journal of Research
Peer Reviewed Journal
ISSN 2278 – 0297 **(Print)**
ISSN 2278 – 6414 **(Online)**
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https://www.patnawomenscollege.in/journal

Mycoremediation Potential of Wild *Pleurotus* Species in Environment Pollutant Degradation

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Received : October, 2025
Accepted : January, 2026
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Abstract : *Through this research work, the potential of wild Pleurotus species for mycoremediation of environmental pollutants was investigated. The study was aimed at performing a comparative analysis of different pollutant degradation using the same species under the same experimental framework. Degradation of a textile dye (methylene blue), a phenolic compound (phenol), and a heavy metal (lead) was examined. Unlike previous studies, that focused primarily on a single pollutant degradation, this research was aimed at performing a comparative analysis of different pollutant degradation using the same species under the same experimental framework. Early-stage liquid mycelial inoculum was used to allow direct interaction with pollutants, and the experiment was conducted without the addition of external enzymes or chemical stimulants to assess the fungus's intrinsic pollutant degrading potential. Visual observations and spectrophotometric analysis confirmed that Pleurotus mycelium exhibited measurable pollutant degradation across all three samples. Variations in degradation rate and removal efficiency were observed among the different pollutants, highlighting the potential of wild Pleurotus species as economical and naturally efficient agents for sustainable pollutant remediation.*

Keywords: *Mycoremediation, Pollutant degradation, Liquid mycelial inoculum, Spectrophotometric analysis*

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Introduction:

Environmental pollution caused by the discharge of synthetic dyes, phenolic compounds, and heavy metals into aquatic ecosystems without adequate treatment continues to pose a serious global threat to both environmental and public health. Conventional physical and chemical treatment methods, although effective, are often associated with high operational costs, energy demands, and the generation of secondary pollutants (Singh & Singh, 2021). Biological remediation approaches, particularly fungal-based treatment systems, have gained significant attention as sustainable and cost-effective alternatives for wastewater and soil remediation. Recent studies have highlighted the efficiency of white-rot fungi in degrading a wide spectrum of recalcitrant organic pollutants due to

their extracellular oxidative enzyme systems and high metabolic adaptability (Sharma & Thakur, 2022). Among these, *Pleurotus* species are especially promising because of their rapid mycelial growth, high biomass yield, and demonstrated ability to detoxify textile dyes, phenolic compounds, and heavy metals in aqueous environments (Das & Mukherjee, 2022). Submerged cultivation systems using liquid mycelial inoculum have further been reported to enhance pollutant–fungus interaction, resulting in improved degradation efficiency compared to solid-state biomass applications (Kumar et al., 2022). Additionally, several recent investigations confirm the capability of *Pleurotus* species to tolerate and remove heavy metals such as lead through biosorption and intracellular sequestration mechanisms (Mehta et al., 2023).

However, most existing studies focus on the degradation of a single pollutant and frequently rely on externally added enzymes, chemical inducers, or pre-conditioned fungal biomass, which may not reflect realistic environmental conditions. Therefore, the intrinsic biodegradation potential of fungi under natural, unstimulated conditions remains insufficiently explored.

To address these gaps, the present study evaluates the comparative degradation of a textile dye (methylene blue), a phenolic compound (phenol), and a heavy metal (lead) using a single wild *Pleurotus* strain under a uniform experimental framework. Earlystage liquid mycelium was employed as the inoculum, and no external enzymatic or chemical stimulants were added, allowing assessment of the fungus's true innate pollutant-degrading capacity under environmentally relevant conditions.

Materials and Methods:

Methylene blue (1% aqueous solution), phenol crystals, and lead acetate powder were used as the pollutant sources. Phenol crystals were melted prior to preparation, while lead acetate was used in its powdered form. Potato Dextrose Broth (PDB) served as the growth medium. Wild *Pleurotus* species liquid mycelium, obtained from a certified microbiology culture laboratory, served as the fungal inoculum. All experimental setups were carried out in 250-mL sterilized Erlenmeyer flasks.

Preparation of Potato Dextrose Broth (PDB): Preparation of PDB followed standard microbiological practice described by Cappuccino and Sherman (2014). Approximately 300 mL of PDB was prepared by dissolving the appropriate amount of PDB powder in distilled water. The medium was sterilized by autoclaving at 121 °C and 15 psi for 15 minutes (Cappuccino & Sherman, 2014). After cooling, 100 mL of sterile PDB was dispensed into each 250-mL flask, and each flask was inoculated with 5 mL of *Pleurotus* liquid mycelium. The inoculated flasks were incubated in a shaker incubator at 25–28 °C and 120 rpm for 3–4 days to allow active mycelial growth.

Preparation of Pollutant Stock Solutions: Preparation of pollutant stock solutions was based on standard protocols used in fungal biodegradation studies (Saratale et al., 2020). Stock solutions (1000 ppm) of each pollutant were prepared as follows: for methylene blue, 10 mL of the 1% aqueous dye solution was diluted to 100 mL with distilled water; for phenol, 100 mg of melted phenol crystals was dissolved in 100 mL of distilled water; and for lead, 100 mg of lead acetate powder was dissolved in 100 mL of distilled water. All stock solutions were sterilized by autoclaving at 121 °C for 15 minutes. Working solutions of 50 ppm were then prepared from each stock by diluting 5 mL of stock to 100 mL with distilled water.

Experimental Setup: Each experimental flask contained 100 mL of PDB previously inoculated with actively growing *Pleurotus* liquid mycelium. To this, 50 mL of the respective 50 ppm working pollutant solution was added aseptically, resulting in a final reaction mixture for fungal treatment. Based on the type of pollutant added, three experimental flasks were prepared: methylene blue with fungal inoculum (MB), phenol with fungal inoculum (PH), and lead with fungal inoculum (Pb). These codes (MB, PH, and Pb) are used throughout the study to denote the respective treatment flasks. All experimental flasks were incubated under identical shaking conditions at 25–28 °C and 120 rpm for a period of 12 days.

Sampling and Analytical Measurements: At each sampling interval (Day 0, Day3, Day 6, Day 9, and Day 12), 5 mL of culture sample was withdrawn from each flask and centrifuged at 5000 x g for 10 minutes to

separate the fungal biomass. The clear supernatant obtained after centrifugation was used for analytical assessment of pollutant degradation by the fungal mycelium. Methylene blue degradation was monitored spectrophotometrically at approximately 665 nm, while phenol concentration was measured at approximately 270 nm using a UV–Visible spectrophotometer. Lead concentration was determined using Atomic Absorption Spectroscopy (AAS) with calibration standards of 2, 5, and 10 ppm. A decrease in absorbance or pollutant concentration was interpreted as evidence of fungal degradation or biosorption activity.

Calculation of Degradation Efficiency: The efficiency of pollutant degradation by the fungal mycelium was calculated as: Degradation (%) = $(C_0-C_t)/C_0 \times 100$ where C_0 is the initial concentration at Day 0 and C_t is the concentration at a given sampling time.

Statistical Analysis:

All experimental measurements were carried out at defined sampling intervals (Day 0, 3, 6, 9, and 12), and degradation efficiency was calculated using standard biostatistical methods. Data were organized and expressed as percentage removal values for comparative evaluation. For analytical comparison of degradation trends among different pollutants over the incubation period, one-way Analysis of Variance (ANOVA) was considered as a statistical approach to assess variation in mean degradation efficiency across treatment groups. Statistical interpretation was performed to evaluate relative differences in pollutant removal patterns under identical experimental conditions.

Results and Discussion:

The treated samples were analyzed at regular sampling intervals (Day 0, 3, 6, 9, and 12) to evaluate pollutant degradation. For methylene blue and phenol, absorbance was measured using a UV–Visible spectrophotometer, while lead concentration was determined using Atomic Absorption Spectrophotometry (AAS). At each interval, 3 mL of treated sample was withdrawn, clarified to remove fungal biomass, and the supernatant was analyzed for residual pollutant concentration. A progressive decline in absorbance or concentration was observed for all three pollutants, indicating effective fungal-mediated degradation or removal.

The comparative degradation data summarized in Table 1 indicate that methylene blue exhibited the highest removal efficiency (64.8%). This enhanced degradation can be attributed to the strong interaction between *Pleurotus* mycelium and dye molecules, resulting in effective fungal-mediated decolorization, a phenomenon widely reported for *Pleurotus* species (Kumar & Chandra, 2018; Kapahi & Sachdeva, 2017). Lead showed moderate removal (~50%), which may be explained by fungal biosorption and intracellular accumulation mechanisms commonly associated with *Pleurotus* mycelium during heavy metal remediation (Kapahi& Sachdeva, 2017). In contrast, phenol showed comparatively lower degradation (38.4%), possibly due to its higher chemical stability and reduced susceptibility to fungal enzymatic attack under the same experimental conditions, as previously observed in fungal bioremediation studies (Harms et al., 2011).

Table 1. Comparative degradation profile of three pollutants treated with fungal mycelium across five sampling intervals

Day	Methylene Blue Absorbance (660nm)	%Removal (MB)	Phenol Absorbance (270nm)	%Removal (Phenol)	Lead Conc. (mg/L)	%Removal (Pb)
0	0.35	0%	0.26	0%	5.00	0%
3	0.26	24.2%	0.23	11.5%	4.10	18.0%
6	0.19	43.2%	0.19	23.8%	3.30	34.0%
9	0.14	58.5%	0.17	33.8%	2.78	44.4%
12	0.12	64.8%	0.16	38.4%	2.50	50.0%

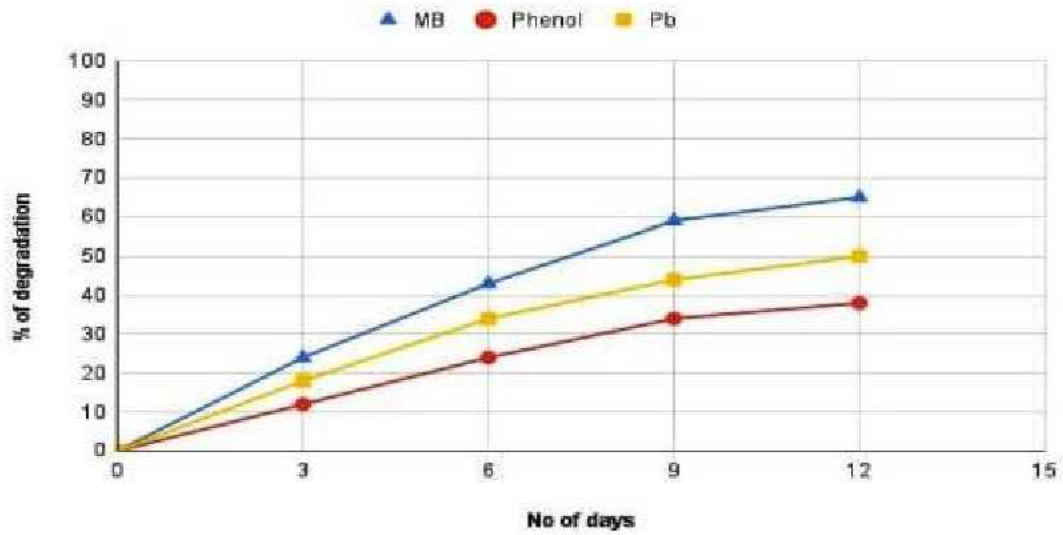


Fig. 1. Time-dependent comparative degradation of methylene blue, phenol, and lead by wild *Pleurotus* mycelium

In the Fig. 1, the graph illustrates a time-dependent degradation trends of all three pollutants over the 12-day incubation period. Methylene blue showed the steepest increase in removal efficiency, followed by lead, while phenol displayed a more gradual decline. The progressive reduction in pollutant concentration across all treatments confirms active interaction between Pleurotus mycelium and the contaminants. The observed differences in degradation profiles reflect pollutant-specific responses to fungal metabolism and biosorption processes, consistent with earlier reports on Pleurotus-based mycoremediation systems (Kumar & Chandra, 2018).

Conclusion:

The present study demonstrates the mycoremediation potential of wild Pleurotus mycelium for degrading chemically diverse environmental pollutants under a uniform experimental framework. Significant reductions in methylene blue, phenol, and lead were observed over a 12-day treatment period, confirming effective fungal-mediated degradation. Methylene blue showed the highest degradation efficiency, followed by lead, while phenol exhibited comparatively lower removal, indicating pollutant-specific variation in degradation behavior. The use of early-stage liquid mycelial inoculum without external enzymatic stimulation highlights the feasibility of a low-cost and naturally driven fungal treatment system.

From an environmental perspective, this study supports the application of wild fungal resources as sustainable alternatives to conventional chemical treatments. Such mycoremediation strategies can reduce treatment costs, minimize secondary pollution, and promote eco-friendly wastewater management. The findings hold practical significance for environmental protection and public health by contributing to safer water resources and sustainable pollution control practices.

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