



Sustainable Clean-Up of Xenobiotic Compounds in the Environment using *Phanerochaete Chrysosporium*

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Abstract

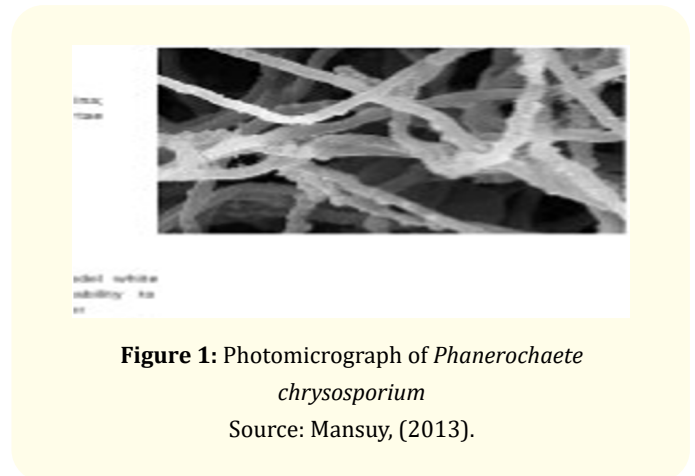
The presence of xenobiotics in the environment pose a lot of health hazards to the wellbeing of living things/ The use of *Phanerochaete chrysosporium* to sustainably free the environment from these recalcitrant xenobiotic compounds have been investigated using standard microbiological techniques. Samples used included commercially purchased dyes (congo red, malachite green and chrystal violet) and textile industry effluent in Lagos, Nigeria. Malt extract agar medium enriched with urea and glucose as nitrogen and carbon sources respectively and diluted in 1L of distilled water was used to culture the fungus. Eighty millilitres of different concentrations of dye solutions in quadruplets were poured in four Erlenmeyer flasks. Results obtained from this study showed that, the pH and presence of co-substrates in the medium affected the rate of decolourization of the dyes and breakdown of the lignolytic substances. More nitrogen in the medium brought about increased decolourization, while more glucose retained the dye colour. The contribution of *Phanerochaete chrysosporium* to sustainable environment through its activity of biodegradation cannot be over-emphasised.

Keywords: Xenobiotics; Bio-Degradation, *Phanerochaete chrysosporium*; Environment; Sustainable

Introduction

Phanerochaete chrysosporium is a white rot fungus, with specialized ability to degrade the aromatic polymer and lignin, while leaving the white cellulose, nearly untouched [1]. It is a member of the Basidiomycota and bears its merotic spores externally in a structure called Basidium [2]. *Phanerochaete chrysosporium* releases extracellular enzymes to break up lignin into utilizable forms [3]. Lignin bonds are broken down by non-specific oxidizing agents present in the enzymes produced by these fungi [4]. The genome of *P. chrysosporium* has been completely sequenced [5].

Genome Structure *Phanerochaete chrysosporium* consists of approximately 29.6 million base pairs arranged in ten liner chromosomes which is the genome [6]. Most of the organism's information is provided by the genome [4]. In biotechnology, *P. chrysosporium* has enhanced the elucidation of P450 mono-oxygenase genes. This has given insight to complex protein interactions. And the interactions have revealed components responsible for the de-



grading extracellular enzymes production [7]. Alternative splicing during transcription, has been facilitated by P450 mono-oxygenase genes, thus, explaining this organism's evolution of diverse metabolic activities [8].



Figure 2: *Phanerochaete chrysosporium* degrades lignin, leaving the white cellulose untouched.

Source: Mansuy, (2013).

Cell Structure and Metabolism: *P. chrysosporium* is a crust fungus, with branched septate hyphae and chlamydiospores at the tips. Its reproductive bodies are flat and fused [9]. It has asexual blastoconidia present in conidiophores [4]. Extracellular enzymes such as lignin peroxidase are involved in degradation of lignin [10]. *Phanerochaete chrysosporium* grow best at moderate temperature [3]. It has a unique capacity of degrading complex lignin from plants. Complex lignin molecules are broken down saprophytically [1].

Pathology: *Phanerochaete chrysosporium* is neither zoonotic nor does it cause disease in man [5]. Dead plants which serve as optimal substrate have been decomposed by this fungus. Its activity is detected by white cellulose patches formed as a result of disappearance of lignin from plant structure [11]. Most aromatic dye pollutants create aesthetic problems due to their recalcitrant nature. (Combes and Haveland Smith;1982). Similarly, many synthetic dyes used in most industries are almost not biodegradable and cumbersome to eliminate [3,7,10]. Conventional textile effluent treatments pose serious sludge disposal problems, so degradation ability of *P. chrysosporium* during fermentation by extracellular enzymes is of importance [8].

Materials and Methods

Collection and transport of materials

Three (3) commercially most utilized dyes in various industries were used as samples for the analysis. These dyes were congo red, malachite green and crystal violet. They were purchased locally from the market in Lagos, Nigeria. The effluent from a textile industry was collected using sample bottles and transported to Mi-

crobiology Departmental laboratory for analyses. All samples were subjected to standard microbiological analyses.

Fungal Strain

Phanerochaete chrysosporium pure culture was obtained from the stock culture in the Department of Microbiology, University of Calabar and was sub-cultured in a medium of malt extract agar medium diluted in 1 litre of distilled water with pH adjusted to 7.0 and growth at 35°C according to the method described by Suzuki, et al [12].

Selection of dye concentration

The optimum dye concentration was selected after seven-days of experiment using four concentrations (i.e. 10, 20, 30 and 50 ppm) of each dye according to the method described by Singh and Chen [13]. Erlenmeyer flasks were used in quadruplets for the analysis. Each flask contained 80 ml of dye solutions, which were of different dye concentrations. Mycellial discs of 2 cm diameter of *P. chrysosporium* was cut out from a pure culture and inoculated in each flask.

Results

The effect of *Phanerochaete chrysosporium* on different concentrations of the three dyes is presented in the table 1 below.

Dye concentration (ppm)	Initial	After 7 days
Congo red		
10	41.50 (1.34)	0.00 (0.00)
20	126.33 (1.35)	62.3 (4.15)
30	682.37 (1.94)	332.50 (11.11)
50	1240.42 (3.96)	1227.36 (11.91)
Malachite green		
10	1531.82 (2.80)	207.08 (9.64)
20	1540.63 (1.73)	578.6 (12.36)
30	2237.64 (2.24)	976.00 (15.60)
50	3561.31 (4.22)	3492.42 (16.35)
Crystal violet		
10	217.25 (1.10)	205.39 (4.69)
20	916.60 (1.15)	392.82 (12.66)
30	1740.80 (2.13)	951.57 (17.98)
50	2694.57 (2.90)	2736.33 (15.87)

Table 1

Discussion

The sustainable use of *Phanerochaete chrysosporium* for the degradation of xenobiotic dyes has been investigated using standard microbiological techniques. The results obtained in this study, after seven (7) days of treatment, have shown that decolourization of dyes was inversely proportional to the concentration of dyes. Papinutti and Forchiassin [9] have reported that higher concentration of malachite green dye, affected both growth and decolourizing ability of the fungus. The results obtained also corroborated by that, obtained by Suzuki, *et al.* [12], where it was reported that the ability of *P. chrysosporium* to degrade azo dyes had a nexus with the ability of these organisms to synthesize lignin-degrading enzymes [1]. Similarly, when *P. chrysosporium* was cultured under ligninolytic and non-ligninolytic conditions, catalyzed by lignin peroxidases, which brought about sequential N de-methylation of parent compound. This effect resulted in metabolism and degradation of dyes into distinct metabolites. This result is similar to that reported by Cashban, *et al* [14].

Urea sterilization and addition in the form of nitrogen and co-substrates, with *P. chrysosporium* enhanced pH and decolourization of the dye solutions while absence of nitrogen affected the decolourization process adversely, to as low as 20% after seven days of incubation. The presence of glucose in the form of carbon affected it adversely. *Phanerochaete chrysosporium* treated dye solutions recorded low pH, (2.24) at 30 DAT in non-sterilized and glucose added solutions (GAS) for malachite green. During this process, organic acids were elaborated by *P. chrysosporium*. Dye colour was enhanced by addition of co-substrates and sterilization. Addition of 10.0% nitrogen source, decolourization of dye by the organism increased significantly. Similarly, addition of glucose, as carbon source impacted positively on decolourization efficiency of *Phanerochaete chrysosporium* [15,16].

Conclusion

Results obtained from this study, have shown that the use of *Phanerochaete chrysosporium*, with carbon and nitrogen sources greatly enhanced the removal of dye and reduction in chemical oxygen demand (COD) of dye-containing industrial effluents. Thus, the potentials of *Phanerochaete chrysosporium* fungus to sustainably degrade xenobiotics in the environment cannot be over-emphasised.

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