ORIGINAL ARTICLE

Agaricales (Gilled Mushrooms) as Biosorbents of Synthetic Dye

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ABSTRACT

Introduction: Due to rapid industrialization and urbanization, abundant industrial effluents are discharged into the environment. Concerns have been raised on dye manufacturing and textile industries due to the detrimental effects of effluents containing dye discharged. Hence, this study aims to examine the adsorption of a common synthetic dye using Agaricales (Gilled Mushrooms) as biosorbents. **Method:** Among the 5 species of Agaricales cultivated in the screening experiment, *Pleurotus ostreatus* (Oyster Mushroom) and *Pleurotus eryngii* (King Oyster Mushroom) were found to have relatively higher growth rates of 86.17% and 77.97% respectively. *Pleurotus ostreatus* was selected for recultivation with addition of 50 ppm of Remazol Brilliant Blue R dye to examine its ability to remove the dye. *Pleurotus ostreatus* showed rapid decolorizing ability within 3 days of cultivation with Ramazol Brilliant Blue R. Therefore, batch analysis was subsequently conducted by varying the experimental parameters. **Results:** AFrom the batch analysis, *Pleurotus ostreatus* achieved the highest dye decolorization in cultivation medium of pH 2, 0.1 mL surfactant, 0 mg/L of sodium chloride and with 8 plugs of biosorbent. Adsorption isotherm studies were also conducted. The adsorption data fitted Jovanovic isotherm the most with highest R^2 value of 0.9949 compared to Langmuir, Freundlich and Harkin-Jura isotherms. **Conclusion:** This study shows the potential of Agaricales, particularly *Pleurotus ostreatus* as biosorbent of synthetic dye due to its high growth rate and efficiency of synthetic dye removal.

Keywords: Agaricales, Adsorption, Biosorbents, Dye, Decolorize, Isotherm

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INTRODUCTION

Rapid industrialization and urbanization have resulted in large amount of industrial effluents being discharged into the environment. There have been increasing public concerns over the textile-manufacturing and dye processing industries as they are the major contributors of dye discharges and these discharges contain high concentrations of synthetic dyes in addition to heavy metals such as cobalt, zinc, copper and lead, which are detrimental to health (1). The findings of the World Bank showed textile industry contributed to about 20% of world water pollution issues (2). It had been estimated that about 15% of leftover liquors from incomplete dyeing processes were released into the environments (3). However, current wastewater treatment methods are only able to remove dye from the effluent by 45% (4). Hence, it is essential to either improve the efficiency of current wastewater treatment methods or seek alternatives to treat residual dye-containing wastewater, which is linked to health and environmental impacts.

The conventional wastewater treatment methods involving the mechanism of coagulation/flocculation, filtration and ion exchange are expensive due to the chemicals required (5). Therefore, much focus has been placed on the derivation of cost-effective biomaterials for the removal of dye from wastewater. It was found that by utilizing fungi as biosorbents, the removal of dye from wastewater can achieve a high efficiency of more than 93% (6, 7). As a promising, environmental-friendly and cost-effective alternative to conventional wastewater treatment, biosorption of dye using bacteria and fungi have been subjected to frequent studies.

Besides dye, fungi have also been found to demonstrate the ability in removing heavy metals. Fungi have complex cell wall structures which provide multiple means for the binding of metal and dye ions protein, chitins, glucans and mannans are examples of organic components found in the cell walls of fungi that confer fungi a huge variety of functional groups for binding of metal and dye ions onto their cell wall (8). Fungal cell

walls are the first component that metal and dye ions interact with before entering cell cytoplasm. Metal and dye ions bind on the surface of the cell walls through stoichiometric interactions, mainly by selectively attaching to functional groups on the cell walls. There are many reactive functional groups on fungal cell walls which offer active binding sites to metal and dye ions. Different biosorbents have different cell wall compositions, hence different functional groups with different affinity for various metal and dye ions (9).

A wide range of fungal biosorption mechanisms has been identified. From the fungal metabolism dependence perspective, fungal biosorption mechanisms can be divided to 2 major processes which are actively uptake and passive uptake of metal ions (8). Active uptake of metal ions is metabolism-dependent in which energy produced by fungi cell is required to absorb the metal ions. Passive uptake is a metabolism-independent process involving physical adsorption or binding of metals on the extracellular components of fungi (10). The passive uptake of dye ions by physical adsorption was examined in this study.

Agaricales, also known as the gilled mushrooms in the Basidiomycota phylum are commonly used for biosorption of synthetic dyes. Agaricales can be easily differentiated from other phylum of fungi by their fruiting bodies known as basidia which are mostly club-shaped. Basidia are responsible for reproduction of this phylum of fungi. The gill-like structures underneath the basidia give Agaricales the common name of gilled mushrooms (11). Agaricales have received significant attention in the studies of biosorption as they can be easily cultivated, demonstrate rapid growth which enables large amount of biomass to be generated in a relatively short time and provide inexpensive biomaterials for pollutants removal (12).

Fungal biomass has been frequently studied by various researchers due to its potential application as biosorbents for removal of synthetic dyes. The biosorption mechanisms of synthetic dye by using fungi as biosorbents were the focus of several recent studies reviewed (7, 13-16). Fungi can be utilized as biosorbents in either living or dead form. Aksu and Balibek (17) found the dead fungi biomass of *Rhizopus arrhizus* (mucorales order) exhibited the highest removal rate of 87.2% for Yellow RL anionic dye. Biosorption mechanism involving dead biomass is less complex as the dead biomass can be applied in powdered form during biosorption process and dead biomass was often free from contamination (12).

In terms of live fungi biomass, Kabbouta and Taha (7) had studied biosorption of methylene blue azo dye by using *Aspergillus fumigatus* (eurotiales order) in which 93.5% dye removal rate was achieved. In the biosorption study carried out by Bankole, Adekunle & Govindwar (18), *Achaetomium strumarium* (sordariales order) exhibited excellent decolorization of 99.7% on acid red 88 azo dye. On the other hand, Ortiz- Monsalvea et al. (16) found that *Trametes villos* (polyporales order) decolorized acid blue anthraquione dye instead of other azo dyes at an efficiency of 96.8%. It can be concluded that decolorization of synthetic dye is highly dependent on the type of fungi used.

Nor et al (13) conducted an enzymatic study on the biodegradation mechanism of cresol red by *Trichoderma harzianum* (hypocreales order) and found that besides biosorption, the presence of laccase aided in the biodegradation of cresol red into 2,4-dihydroxybenzoic acid and 2-hydroxybenzoicacid which eventually resulted in decolorization of the synthetic dye. Very few studies have been conducted to specifically investigate the potential of Agaricales in adsorbing anthraquinone dye. Remazol Brilliant Blue R (RBBR) dye is a common anthraquinone dye used in textile industries which poses harmful and carcinogenic effects on aquatic lives when it is released into the environment (3). It falls in the category of reactive dyes which are used in approximately 45% of textile manufacturing worldwide (3). Hence, this study aims to examine the biosorption of RBBR anthraquinone dye by using Agaricales as biosorbents.

MATERIALS AND METHODS

The methodology was designed with reference to Nor et al. (13) in which biosorption of Cresol Red using was *Trichoderma Harziamum* sp carried out. Several modifications have been made to impart elements of novelty to this study, which are:

i. The use of Remazol Brilliant Blue R (RBBR) as anthraquinone dye

ii. The use of a different division of fungi (Basidiomycota instead of Ascomycota). Specifically, species under Agaricales of the Basidiomycota are used as potential biosorbents.

iii. The inclusion of isotherm studies namely Langmuir, Freundlich, Harkin-Jura and Jovanovic isotherms.

Screening and selection of Agaricales

Five different Agaricales species consisting of *Hypsizygus tessellatus, Lentinula edodes, Pleurotus eryngii, Pleurotus ostreatus* and *Flammnulina velutipes* were cultivated in separate petri dishes for 5 days in dark conditions at 27 °C. Each petri dish was filled with 5 g/L of yeast extract, 20 g/L of malt extract agar and glucose. The growth of fungi was determined using Equation 1. Agaricales with the highest growth rate were recultivated for 5 days with the addition of 50 mg/L of RBBR dye. The colour changes on the blue agar indicated that the selected Agaricales species were able to decolourize the RBBR dye.

$$\text{Growth rate (\%)} = \frac{\text{Average diameter of fungi}}{\text{Diameter of petri dish}} \times 100 \quad (1)$$

Cultivation in liquid medium

In this stage, the selected Agaricales species that was previously cultivated in solid agar medium was transferred by using cork borer into conical flask filled with 20 mL of liquid medium containing 5 g/L of yeast extract, 20 g/L of glucose solution and 0.025 g/L of RBBR dye. After 15 days of incubation, the liquid medium containing solid fungi biomass was filtered to separate the fungi biomass. The filtrate collected was pipetted into Type 9P semi-micro cuvette to be tested using UV Vis spectrophotometer at maximum absorption wavelength of 592 nm. The absorbance value was recorded and substituted into the linear equation of the calibration curve to obtain the concentration of RBBR dye (refer to Equation 2). The decolorization rate also known as dye removal rate was calculated using the Equation 3.

$$y=mx+c \quad (2)$$

y represents absorbance value
m represents the gradient of the equation
c represents the y-intercept of the graph
x represents the concentration of tested sample

$$\text{Rate(\%)} = \left(1 - \frac{C_1}{C_0}\right) \times 100 \quad (3)$$

C_1 represents the final concentration of the dye solution
C_0 represents the initial concentration of the dye solution

Variation of parameters for biosorption

Parameters that were varied in this study to test their effects on biosorption comprised pH, salinity, dosage of absorbent and the effect of surfactant. The parameters varied are summarized in the Table I. The parameters represented the environmental factors that affect the efficiency of the fungi to absorb RBBR dye.

Table I : The summary of parameters tested in the experiments

Parameters	Chemicals/ materials used	Values/ Concentrations/ Dosages
pH	HCl or NaOH	2, 4, 7, 10
Salinity (mg/L)	NaCl	0.0, 50.0, 100.0, 150.0
Surfactant (mL)	Tween 80	0.1, 0.5, 1, 1.5
Dosage of adsorbent (plugs)	5 mm of grown fungi	2, 4, 6, 8

Isotherm studies

The interaction between the fungus cultivated and the RBBR dye solution can be studied using adsorption isotherm models. Surface characteristics, adsorption capacity and efficacy of adsorption system were taken into consideration in the adsorption isotherm calculations involving Langmuir, Freundlich, Harkin-Jura and Jovanovic isotherms as below:

$$\text{Langmuir isotherm } \frac{C_e}{q_e} = \frac{1}{q_m K_L} + \frac{C_e}{q_m} \quad (4)$$

C_e represents the final concentration of adsorbate
q_e represents the amount of adsorbate in equilibrium
q_m represents the amount of adsorbate in equilibrium
K_L represents the Langmuir constant value

Freundlich isotherm

$$\log q_e = \log K_F + \frac{1}{n} \log C_e \quad (5)$$

K_F represents the capacity of adsorption
n represents the intensity of adsorption
C_e represents the final concentration of adsorbate
q_e represents the amount of adsorbate in equilibrium

Harkin-Jura isotherm

$$\frac{1}{q_e^2} = \frac{B}{A} - \left(\frac{1}{A}\right) \log C_e \quad (6)$$

q_e represents the amount of adsorbate in equilibrium
B represents the Harkin-Jura constant from the plot of $1/qe^2$ versus log Ce
A represents the Harkin-Jura constant from the plot of $1/qe^2$ versus log Ce
C_e represents the final concentration of adsorbate

Jovanovic isotherm

$$ln\, q_e = \ln q_{max} - K_J C_e \quad (7)$$

q_e represents the amount of adsorbate in equilibrium
q_{max} represents the maximum amount of adsorbate in equilibrium
K_J represents the Jovanovic constant

RESULTS

Screening of fungi species

In order to obtain the yield of active cell from the cultivation, harvesting of cell should take place at the end of the log phase (19). The growth rate of each Agaricales species is tabulated in Table II. Throughout the 5-day observation on *Hypsizygus tessellatus, Lentinula edodes, Pleurotus eryngii, Pleurotus ostreatus, Flammnulina velutipes* cultivated, it was found that both *Pleurotus ostreatus* and *Pleurotus eryngii* exhibited the growth rates of 86.17% and 77.97% respectively. The growth rates of both species were quite similar because both species belong to same genus of *Pleurotus.* As *Pleurotus ostreatus* demonstrated the highest growth rate, it was selected for recultivation with RBBR dye. Figure 1 shows 5-day observation on the growth of *Pleurotus ostreatus*.

Recultivation of fungi in solid medium

Recultivation of *Pleurotus ostreatus* was conducted to determine its dye decolorization ability. The fungus' ability to decolorize dye can be distinguished by visual inspection of colour changes of the agar medium as illustrated in Figure 2. Based on Figure 2, *Pleurotus ostreatus* exhibited excellent RBBR dye decolorization ability within 3 days of observation.

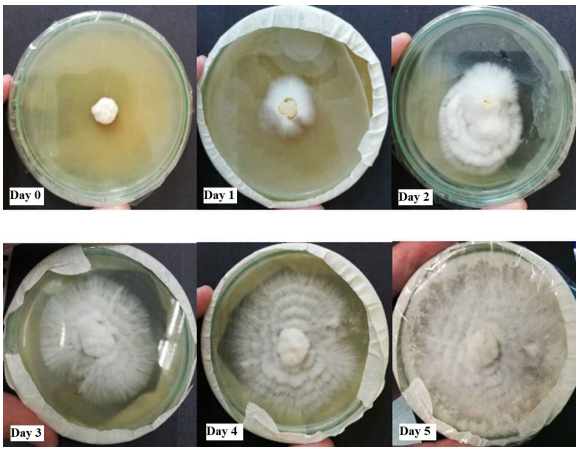

Figure 1 : 5 days observation on the growth of *Pleurotus ostreatus*. 5 different types of Agaricales were cultivated in malt extract (ME) agar plates separately. Observations were made on the growth of mycelium of each Agaricales species throughout 5 days. The Agaricales species with the widest mycelium coverage within the shortest timeframe was selected.

Table II : Growth rates of fungi over 5 days

No	Growth rate (%)		
	Day 1	Day 3	Day 5
B01	0.00	8.72	56.00
B02	0.00	5.45	72.00
B03	0.00	2.35	32.56
B04	0.00	7.28	45.00
Average			51.39
S01	0.00	24.00	74.14
S02	0.00	10.85	75.56
S03	0.00	5.00	59.00
S04	0.00	45.56	83.00
Average			72.93
KO1	0.11	30.00	62.45
KO2	1.83	89.00	89.00
KO3	0.84	45.89	76.42
KO4	0.96	66.00	84.00
Average			77.97
O1	2.35	72.00	98.17
O2	0.00	45.83	75.89
O3	1.43	65.00	84.35
Average			86.14
E1	0.00	0.00	0.00
E2	0.00	5.00	5.00
E3	0.00	15.00	17.89
E4	0.00	32.00	36.00
Average			14.72

Note: B= Hypsizygus tessellatus (Buna Shimeji Mushroom)
S= Lentinula edodes (Shiitake Mushroom)
KO= Pleurotus eryngii (King Oyster Mushroom)
O= Pleurotus ostreatus (Oyster Mushroom)
E= Flammulina velutipes (Enokitake Mushroom)

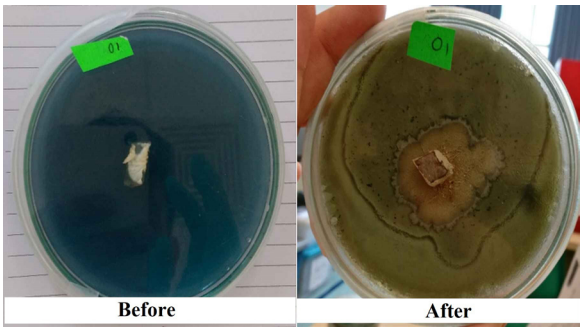

Figure 2 : Visual changes during recultivation of *Pleurotus ostreatus* with dye. Pleurotus ostreatus was selected to be recultivated with addition of RBBR dye to determine its decolorization ability.

Effect of surfactant

The effect of surfactant on decolorization of RBBR dye by *Pleurotus ostreatus* was studied throughout a 15-day incubation. Tween 80 was used as surfactant in this experiment. Different volumes (0.1 mL, 0.5 mL, 1.0 mL, 1.5 mL) of Tween 80 were added to the dye-containing culture media in this experiment. The surfactant was added into the liquid culture media containing dye and fungi biosorbent. Results from the experiment showed that with the addition of 0.1 mL, 0.5 mL, 1.0 mL and 1.5 mL Tween 80, the dye decolorization rates were 83.36%, 67.22%, 63.02% and 53.93% respectively as plotted in graph of dye removal rate vs surfactant (Figure 3a).

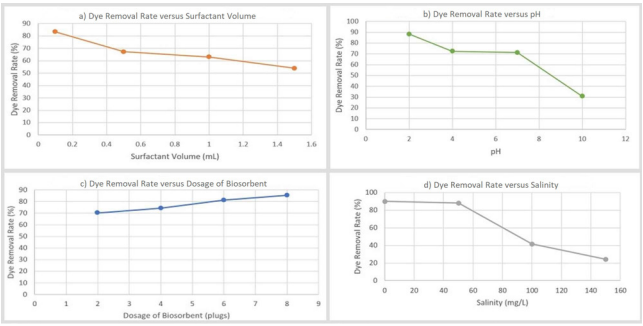

Figure 3 : Graph of dye removal rate versus four experimental parameters. (a) shows dye removal rate versus surfactant volume, (b) shows dye removal rate versus pH, (c) shows dye removal rate versus dosage of biosorbent, and (d) shows dye removal rate versus salinity.

Effect of pH

The effect of varying pH on the decolorization of RBBR dye by *Pleurotus ostreatus* was conducted at pH 2, 4, 7 and 10 over 15-day incubation. The removal rates of RBBR by *Pleurotus ostreatus* were calculated to be 88.23%, 72.44%, 71.18% and 30.73% respectively for pH 2, 4, 7 and 10. The results were shown in Figure 3b – plots of dye removal rate versus pH.

Dosage of biosorbent

The dosage of biosorbent was measured as number of plugs of *Pleurotus ostreatus*. Cork borer was used to cut 5mm plugs of *Pleurotus ostreatus* in the petri dishes, which were transferred into conical flasks filled with

culture media and dye solutions. The dosages of the biosorbent used in this experiment were 2, 4, 6 and 8 plugs respectively. The removal rates of RBBR dye by *Pleurotus ostreatus* were 70.23%, 74.27%, 81.15% and 85.48% respectively for dosages of 2, 4, 6 and 8 plugs. The results were plotted in Figure 3c – dye removal rate versus dosage of biosorbent.

Effect of salinity

The effect of salinity on decolorization of RBBR dye by *Pleurotus ostreatus* was conducted over 15 days of incubations where the concentrations of sodium chloride, NaCl in the culture media were 0 mg/L, 50 mg/L, 100 mg/L and 150 mg/L respectively. Dye decolorization rates recorded were 89.89%, 88.15%, 41.59%, and 24.44% respectively. The results are plotted in Figure 3d – dye removal rate versus salinity.

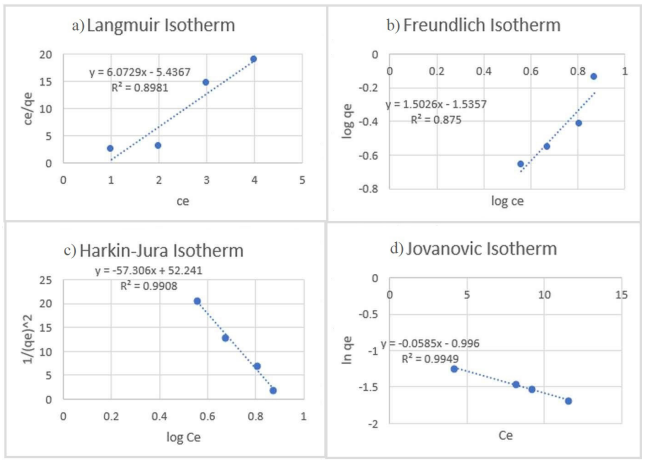

Figure 4 : Graph of isotherms for biosorption of RBBR by *Pleurotus ostreatus*. Four isotherms were examined in this study, i.e. (a) Langmuir, (b) Freundlich, (c) Harkin-Jura, and (d) Jovanovic isotherms. The biosorption data fitted Jovanovic isotherms the most with the R2 closest to 1.

Isotherm studies

Figure 4 displays isotherms for biosorption of RBBR by *Pleurotus ostreatus*. RBBR dye adsorption with *Pleurotus ostreatus* was analysed with Langmuir, Freundlich, Harkin-Jura and Jovanovic isotherms respectively. It was found that the biosorption data fitted the Jovanovic isotherm the most with R^2 value (0.9949) closest to 1.

DISCUSSION

Screening of fungi species

Room temperature of 27 °C was the optimum temperature for the growth of *Pleurotus sp.* This is supported by Hoa and Chun-Li (20) where experiments conducted on the effect of temperature on the growths of *Pleurotus ostreatus* and *Pleurotus cystidiosus* showed that the mycelium grew significantly under incubation at 27 °C. Furthermore, yeast extract is commonly added into culture medium to boost its nutrient contents. In this experiment, the amount of yeast extract used was 0.4% which fit into the optimum range reported by Owaid, Al-Ani and Muslat (21) whose study showed that *Pleurotus sp* grew best in yeast (*Saccharomyces cerevisiae*) extract with concentrations between 0.3% to 0.5%.

Recultivation of fungi in solid medium

The malt extract agar is commonly used to support the inoculation of most fungi and bacteria species (22). The presence of carbon and nitrogen in the agar promoted dye decolorization by fungi. Owaid, Al-Ani and Muslat (21) suggested that dye decolorization rate can be improved by raising the dosage of carbon and nitrogen sources, although there is possibility that catabolic repression may happen (6), leading to increased biomass yield instead of higher rate of dye decolorization.

This study found that not all the fungi species can decolorize RBBR dye in a short period of time due to the differences in their ability to break the azo bond. Azo bond breaking is the first step of dye decolorization and this is often influenced by aromatic functional groups on the bonds. Interactions between the functional groups on azo bonds and biosorbent give rise to azo bond breaking (23). *Pleurotus ostreatus* was able to decolorize RBBR dye probably due to its ability to interact with the functional groups on azo bonds.

Effect of surfactant

Based on Figure (3a), the decolorization rates decreased with increasing surfactant volume during the 15-day incubation. Surfactants contain hydrophobic and hydrophilic parts. At the interface of air and water, surfactants tend to align themselves such that the hydrophilic part is in the water and hydrophobic part is in the air. This reduces surface tension as the intermolecular forces between water molecules are weakened (24). Once the surface tension is reduced, it would be easier for adsorbents to adsorb dye in wastewater. However, in the case of solid biosorbent in this study, the surfactants tended to have adverse effect on adsorption. An explanation is that, at the interface of water and biosorbent, the surfactant molecules tend to align themselves in a way that the hydrophilic part is in the water and the hydrophobic part binds to the biosorbent which eventually decreases the surface area available for adsorption of dye compounds. Therefore, increasing volume of Tween 80 reduced dye decolorization in this experiment due probably to adsorption of Tween 80 molecules onto biosorbent, thus reducing its surface area for dye adsorption (25).

This finding is identical to that of Syaffiudin et. al (14) who studied the ability of *Trichoderma Harzianum* and *Acremonium Spinosum* to decolorize mordant orange-1 dye. The results of Syaffiudin et. al (14) showed that the volume of Tween 80 added was inversely proportional to decolorization rate of mordant orange-1. However, it was directly proportional to biomass production rate. Nor et al. (13) also reported that decolorization of Cresol Red by *Trichoderma harzianum* was the highest (88%) with low surfactant of 0.1mL Tween 80. In contrast,

Kristanti et. al (5) reported that the addition of Tween 80 as surfactant aided in the decolorization of Cresol Red dye by *Absidia spinosa* as it raised biomass production, thus raising the surface area of biosorbent available for adsorption.

Effect of pH

Based on Figure 3(b), both normal and acidic conditions were suitable for the decolorization of RBBR dye by *Pleurotus ostreatus.* It was because the majority of fungi species grow vigorously in acidic conditions. However, *Pleurotus ostreatus* was able to tolerate extreme pH of 2 in this experiment. Several studies pointed out that pH is closely related to fungal growth, enzymatic activities and survival rate of the macro fungi (14, 26, 27). pH can, therefore, affect the fungal enzymatic activities and the biosorption behavior. According to Levin, Papinutti and Forchiassin (28), pH ranging from 3 to 7 is ideal for the enzymatic activities of laccases and lignin peroxides found in fungi. Since acidic condition is more favorable to fungi, fungi biomass production and enzyme secretion which aid in the biosorption and breakdown of dye are increased in acidic environment. On the aspect of electrostatic interaction, the positively charged surfaces of biosorbent surface attract the negatively charged dye anions. This mutual attraction facilitates dye decolorization. In a low-pH, there is higher concentration of H^+ ions available for binding the negatively charged organic dye molecules onto the cell walls of fungi (29).

Bankole, Adekunle and Govindwar (18) also showed that isolated Achaetomium Strumarium exhibited the highest Acid Red 88 decolorization rate of 98.99% at pH 4. Under neutral condition of pH 7, the decolorization rate dropped to 76.77%. This finding is further supported by Ortiz-Monsalvea et al. (16) demonstrating that Trametes Villosa achieved the highest decolorization rate of 94.03% in an environment with pH 5. However, the decolorization rate gradually declined to 53.11% in pH 9. This indicated that alkaline condition exhibited adverse effect on the ability of Trametes Villosa to remove dye as the growth of Trametes Villosa was greatly reduced in alkaline condition. Hence, the dye decolourization rate of Trametes Villosa was the lowest in alkaline condition.

Dosage of biosorbent

Based on Figure 3(c), it can be concluded that dosage of biosorbent is directly proportional to dye decolorization rate. The increasing decolorization rates reported with increasing dosages from 2 to 8 plugs could be attributed to the increasing surface area available for the binding of organic dye molecules as more biosorbent was added (30). The increase in surface area basically resulted in more active binding sites available to bind dye anions onto the biosorbent.

Kabbouta and Taha (7) demonstrated that the biosorption of Methylene Blue dye increased with the dose of *Aspergillus fumigatus* as biosorbent. It was due to the augmentation in surface adsorption as the dosage of biosorbents increased. Bankolea, Adekunleb and Govindwarc (18) found that the highest decolorization rate of 98.99% of acid red 88 dye by *Achaetomium Strumarium* at maximum dosage exhibited the lowest adsorption capacity of 20.54. The authors agreed that the dosage of adsorbent is directly proportional to decolorization. However, it also resulted in lower adsorption capacity as larger dosage of adsorbent provided more surface area for adsorption and led to lower saturation on the surface area of biosorbent at constant dye concentration.

Effect of salinity

Figure 3(d) shows that dye removal rates gradually decreased with increasing salinity. Hence, it can be concluded that the rate of dye removal by *Pleurotus ostreatus* is inversely proportional to salinity. The trend can be best explained by the ionic strength of dye solution caused by increasing salt concentration. Ionic strength is dependent on ion concentration of a solution. It is directly proportional to salt concentration. Equilibrium uptake of ions from dye solution by biosorbent is highly influenced by ionic strength.

The presence of salt in dye solution introduced both positively charged sodium cations (Na+) and negatively charges chloride anions (Cl-) to solution. The chloride anions altered the ionic strength of the solution and competed with the dye anions for active binding sites on the biosorbent. Complexes can also form between chloride anions and dye cations, hence affecting the biosorption process significantly (17). From the perspective of activity coefficient, the biosorbent and dye ions tend to be surrounded by a layer electrostatic interaction. The electrostatic interaction increases with ionic strength and prevents the dye ions from binding onto the surfaces of the biosorbent (31).

The result of this experiment can be supported by Aksu and Balibek (17) who found that dried *Rhizopus arrhizus* exhibited highest Yellow RL anionic dye biosorption rate without the presence of salt and the biosorption rate gradually decreased from 87.2% to 67.0% with increasing salinity. In contrast, Nor et al (13) found that *Trichoderma harzianum* was able to adapt to high salinity conditions where biosorption rate was the highest (73%) in salinity as high as of 100g/L. Razmovski and Siban (15) also showed that the biosorption of copper (II) ions and chromium (VI) ions by dried tea fungus increased with salinity.

Isotherm studies

The biosorption data fitted the Jovanovic isotherm well (Figure 4d). Jovanovic isotherm assumes monolayer adsorption without interaction (32). Although Jovanovic isotherm is similar to Langmuir isotherm, it is different

from Langmuir isotherm in terms of surface binding vibration. A study on the equilibrium of biosorption of copper (II) ions by Mussel shells conducted by Farouq and Yousef (33) also showed that Jovanovic isotherm explained the biosorption data the best with the highest R^2 value of 0.9212. In contrast, Kabbouta and Taha (7) showed that the biosorption of methylene blue dye by *Aspergillus fumigatus* followed the Langmuir isotherm with an R^2 value of 0.9906. Hence, it can be concluded that adsorption isotherms vary with different biosorbents due to the differences in the structures of biosorbents as well as dyes.

CONCLUSION

In conclusion, this study demonstrates the potential use of Agaricales as biosorbents for decolorization of synthetic dye. It identifies the optimal conditions for the removal of RBBR dye by an Agaricales fungus called *Pleurotus ostreatus* and shows *Pleurotus ostreatus* as a promising candidate for biosorbent of synthetic dye due to its high growth rate. This study contributes significantly to the advancement of dye wastewater treatment using biomaterials particularly fungi which are inexpensive and easy to grow. Further study can focus on applications of the biosorbent to a pilot-scale dye wastewater treatment plant to identify the most feasible methods of applying the biosorbent, the optimal operational conditions and the effectiveness of the biosorbent at pilot-scale. This study did not include characterization studies of functional groups on the biosorbent which are responsible for the binding and removal of dyes. Therefore, a comprehensive surface characterization of the biosorbent can be a potential extension to this study.

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