Biological Treatment of Cadmium-Contaminated Solution Using White-Rot Fungi *Phanerochaete Chrysosporium* and *Trametes Versicolor*

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Abstract

The toxicity of cadmium (Cd^{2+}) as heavy metal in certain forms and concentrations has drawn attention on various proposed methods to treat cadmium-contaminated solutions. The biological method has been of interest lately due to its environmental friendliness, cost-effectiveness and low energy consumption. This paper, therefore, reports the findings obtained from treating cadmium-contaminated solutions using white-rot fungi *Phanerochaete chrysosporium* (Pc) and *Trametes versicolor* (Tv). The influence of biomass concentration (0.5-1 g), contact period (0-24 hrs), and initial Cd^{2+} solution concentration (50-200 ppm) on the fungi's Cd^{2+} biosorption ability was assessed. *T. versicolor* was found to have higher biosorption capacities with 45-80% of the Cd^{2+} removed, as against *P. chrysosporium* removing 25-55% Cd^{2+} from contaminated solution. The ability of both fungi to biosorp Cd^{2+} increased as the initial Cd^{2+} concentration increased. The kinetic rates were modelled using the pseudo-first-order and pseudo-second-order. The pseudo-second-order model was found to explain the adsorption kinetics most effectively for both *T. versicolor* and *P. chrysosporium*, indicating chemisorption. This study concludes that the white-rot fungi; *P. chrysosporium* and *T. versicolor* have the potential to be a low-cost and ecologically-friendly solution for the treatment of cadmium-contaminated water and wastewater.

Keywords: *Phanerochaete chrysosporium*, *Trametes versicolor*, Fungal Biomass, Cadmium, Concentration, Kinetic model

1 Introduction

Cadmium is a soft, ductile, silver-white, lustrous, electropositive metal with an atom. wt. of 112.4, density of 8.64 g cm^{-3}, and melting pt. of 321 ºC. Like Zn and Hg, Cd is a transition metal in Group II-B of the periodic table (Adriano, 2001). It is of great importance in areas such as electroplating and galvanisation of other metals due to its relatively high resistance to corrosion (Xu *et al.*, 2012). Cadmium is also used extensively as a pigment because of its ability to produce brilliant yellow, orange, and red colours. Common uses of cadmium today are in batteries, alloys, and plastic stabilisers, among others (Turner, 2019).

In as much as cadmium is of great use in the manufacturing industries, it has drawn much concern from researchers due to its serious side effects on mankind and the environment at large. It is continually discharged into the general environment causing pollution to land, sea and air. Cadmium is said to be nephrotoxic, thus; it causes kidney tubular damage. It occurs naturally in the environment as well as a pollutant emanating from industrial and agricultural sources (Järup and Åkesson, 2009). Water pollution of cadmium is mainly from the manufacture and discharge of cadmium-containing articles like paints, pigments, batteries and phosphate fertilizers (Hayat *et al.*, 2019). According to Yang and Massey, (2019),

ingestion and inhalation are the primary routes of exposure to cadmium. Järup and Åkesson (2009) also indicated that, for the non-smoking population, the main source of cadmium intake is food and water which plays a vital role in the food chain.

The potential risks from cadmium exposure are now tightly controlled by occupational exposure standards, regulations for cadmium in ambient air, water and soil, legislation covering cadmium emissions, labelling and disposal of cadmium-containing products, and impurity levels in other products such as fossil fuels, fertilizers and cement. Thus, cadmium release into the environment must be regulated to meet acceptable limits. Sorption of cadmium from the environment is one of the regulatory methods which can be done both chemically and biologically but biological treatment is gaining acceptance due to its environmental friendliness, low cost, high efficiency, biosorbent regeneration ability and the possibility of metal recovery (El-Sheekh *et al.*, 2019; Crini *et al.*, 2019).

The organisms used in environmental biosorption mainly include microbes, algae and plants. However, with the characteristics of quick reproducible velocity and easy control, microbes are preferable to use in environmental treatment (Zeng *et al.*, 2010). Both whole cell and mycelium of white-rot fungi have found great application in heavy metals biosorption because of their low substrate specificity (Ofori-Sarpong, 2020) exhibited by the oxidative and extracellular ligninolytic systems which allow them to transform or destroy a wide range of contaminants in the environment (Chen *et al*., 2022). Among these are the fungi *Phanerochaete chrysosporium* and *Trametes versicolor*. The ability of both fungi to produce a variety of enzymes like manganese peroxidase and lignin peroxidase which have biodegenerative properties that are capable of sorbing heavy metals from wastewater has been extensively studied (Subbaiah *et al.*, 2011; Zhao *et al.*, 2016; Noormohamadi *et al.*, 2019). This research, therefore, sought to investigate the biosorption abilities of the aforementioned fungi in a cadmium-contaminated solution and the effects of environmental parameters like time, initial metal concentration and biomass dosage on cadmium sorption.

2 Materials and Methods Used

2.1 Materials Used

Spore suspension of the fungi, millet to serve as growth media for the fungi, cadmium chloride ($CdCl_2$), hydrochloric acid (HCl), and calcium carbonate ($CaCO_3$) (all of reagent grade) were obtained from the Minerals Engineering Laboratory of the University of Mines and Technology (UMaT), Tarkwa.

2.2 Culturing of Fungi

Fungus spore suspension was made by adding the sterilised deionised water to the spore stock to the 250 ml mark. 10 g of commercial millet and 10 ml of deionised water were measured into sterilised Erlenmeyer flasks and sealed with aluminium foil. The mixtures were then heated till near water absence was achieved, cooled and inoculated with about 5 drops of the spore suspension of the fungus. The mixture was then covered with aluminium foil and shaken adequately to ensure uniform growth. The fungus was allowed a growth period of one week at a temperature between 25ºC and 30ºC for *Trametes versicolor*. The same procedure was employed in the growth of *Phanerochaete chrysosporium* but at a temperature of around 37ºC in an MRC LOM-150-2 refrigerated shaker incubator. Figure 1 depicts the visual growth progress of the cultures.

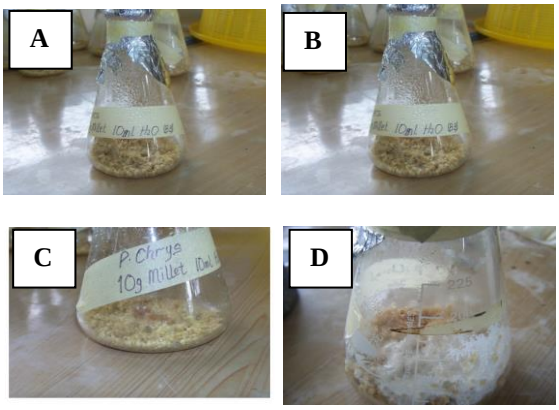

Figure 1 (A) A Day-old Inoculation and (B) A Week Old *T. versicolor* Growth; (C) A Day Old Inoculation and (D) A Week Old *P. chrysosporium* Growth.

2.3 Preparation of Cadmium Solution and Sorption Experiment

Three (3) different concentrations of Cd^{2+} (II) solutions at 50 ppm, 100 ppm, and 200 ppm

respectively were analytically prepared by dissolving 50 mg, 100 mg and 200 mg $CdCl_2$ with 5% HCl solution and diluting with deionised water to the 1000 ml mark in a volumetric flask. pH adjustment was done with the 5% HCl and $CaCO_3$ and maintained within 5.5-6.

Batch sorption experiments were carried out by suspending 0.5 g and 1.0 g of each culture in 50 ml of Cd^{2+} solution with an initial concentration of 100 ppm to check the effect of culture mass on sorption. One gram of each culture mass was measured and contacted with 50 ppm, 100 ppm, and 200 ppm while maintaining the optimal pH of the systems at a range of 5.5-6 to assess the effect of metal concentration on sorption. At predetermined time intervals of 0.5 hr, 1 hr, 3 hrs, 12 hrs, and 24 hrs, the residual Cd^{2+} concentration in the supernatant was isolated using a pipette, and equal volumes of 10 ml were then analysed with Atomic Adsorption Spectrometer (AAS). Metal solution without biomass served as control.

2.3 Analysis

The amount of cadmium adsorbed in percentage and the specific amount of cadmium biosorbed by the biomass were estimated using Equations 1 and 2.

$$\%\ Cd\ Removal = \frac{(C_i - C_f)}{C_i} * 100\% \qquad (1)$$

Where: C_i = initial metal concentration (mg/L) and C_f = metal concentration at the time of isolation (mg/L).

$$q = \frac{(C_i - C_f)}{m} * v \qquad (2)$$

Where: q = amount of cadmium biosorbed by biomass (mg/g), C_i = initial cadmium concentration (mg/L), C_f = concentration at the time of isolation of supernatant solution (mg/L), v = initial volume of cadmium solution (L), m = mass of biomass (g).

Equation 2 indicates the effect of biomass on sorption.

Results from the AAS analysis for the sorption test were modelled using the pseudo-first-order and pseudo-second-order kinetic models as shown in Equations 3 and 4 respectively.

$$\log(q_e - q_t) = \log(q_e) - \left(\frac{k_1}{2.303}\right) t \qquad (3)$$

$$\frac{t}{q_t} = \frac{1}{k_2 {q_e}^2} + \left(\frac{1}{q_e}\right) t \qquad (4)$$

Where,

q_t and q_e = solid-phase ion concentrations at any time (t) and at equilibrium (mg/g), respectively.

k_1 = pseudo-first-order adsorption rate constant.

k_2 = rate constant of the second-order equation

Furthermore, the sum of squared errors (SSE) were calculated using Equation 5.

$$SSE = \sum \frac{(q_{exp} - q_{cal})^2}{q_{exp}^2} \qquad (5)$$

Where q_{exp} = experimental biosorption capacity and

q_{cal} = modelled biosorption capacity

3 Results and Discussions

3.1 Effect of Sorbent Dose on Cd^{2+} Removal

The experiments were conducted with different weights of *Trametes versicolor* and *Phanerochaete chrysosporium* fungi (0 g, 0.5 g, 1.0 g) keeping the concentration of Cd^{2+}, the volume of adsorbate solution and contact time constant. Figure 2 depicts the effect of the sorbent dose of each fungus on Cd^{2+} removal. *P. chrysosporium* biosorped 26%-34% Cd^{2+} from the solution with the highest attained at 30 mins when 0.5 g of fungal biomass was added. When 1 g of Pc was dosed, 26-45% Cd^{2+} was removed from the solution with the optimum of 45.7% attained at 3 hrs. At all sorbent doses of *P. chrysosporium*, there was a drop in the percentage of Cd^{2+} biosorped across the kinetic period. *T. versicolor* on the other hand had 58–63% and 57-65% respectively for sorbent dosages of 0.5 g and 1.0 g. There was a slight increase in Cd^{2+} removal when the mass of fungal biomass was increased. Subbaiah *et al.* (2008) reported a similar trend and attributed the occurrence to the number of adsorption sites available due to an increase in the amount of sorbent. Within the 24 hr period, the concentration of the setup without any fungal biomass added remained unchanged. This confirms

the biosorption ability of the white rot fungi (*P. chrysosporium* and *T. versicolor*). However, *T. versicolor* proved a better biosorbent of Cd^{2+} than *P. chrysosporium*.

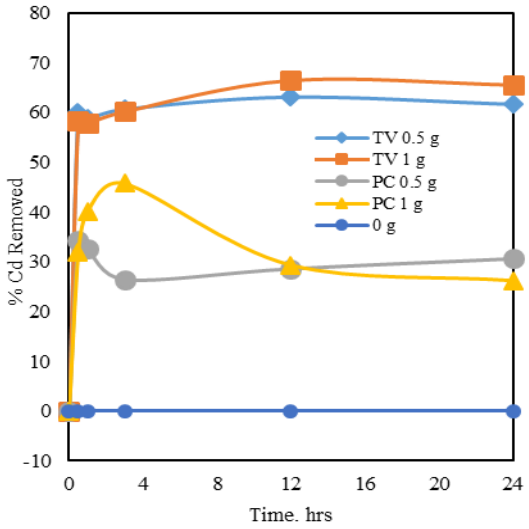

Figure 2 Effect of Sorbent Mass on Cd^{2+} Removal

3.2 Effect of Cd^{2+} Concentration on Cd^{2+} Removal

With the mass of 1 g maintained, the concentration of Cd^{2+} was varied at 50 ppm, 100 ppm and 200 ppm. The percentage of Cd^{2+} removed from solution increased with increasing concentration of Cd^{2+} for *P. chrysosporium* with the highest Cd^{2+} removed attained at 3 hrs for 100 ppm and 200 ppm and 4 hrs for 50 ppm. Consequently, there was no significant increase in Cd^{2+} removed after the optimum time but a decrease for the 100 ppm setup as shown in Figure 3.

The effect of concentration on the effect of *T. versicolor* is depicted in Figure 4. Cd^{2+} removal increased significantly in the first 30 mins for all concentrations with a reduced removal rate for Cd^{2+} concentrations at 50 ppm and 100 ppm. Cd^{2+} concentration at 200 ppm, however, continually increased in removal rate till the 12th hour. The maximum removal of Cd^{2+} for 50 ppm, 100 ppm and 200 ppm setups were 47.52%, 65.55% and 83. 91% respectively. Ultimately, the Cd^{2+} removal ability of *T. versicolor* increased with elevating concentration. The possible contribution of high adsorption at high concentration is attributed to mechanisms such as intraparticle diffusion, electrostatic interaction and ion exchange of fungi biomass (Subbaiah *et al.*, 2008). According to Alothman *et al.* (2020), carboxyl and phosphate groups of the cell walls of fungus are arranged such that the cell wall attains a negative charge and, as such, positive metals bind to the cell walls of the fungus. Again, the fungi secrete some enzymes which are capable of degrading recalcitrant compounds (Prakash, 2017).

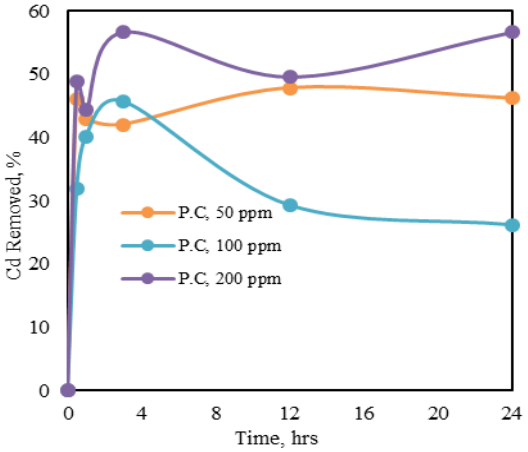

Figure 3 Effect of Concentration on Cd^{2+} Removal by *Phanerochaete Crysosporium*

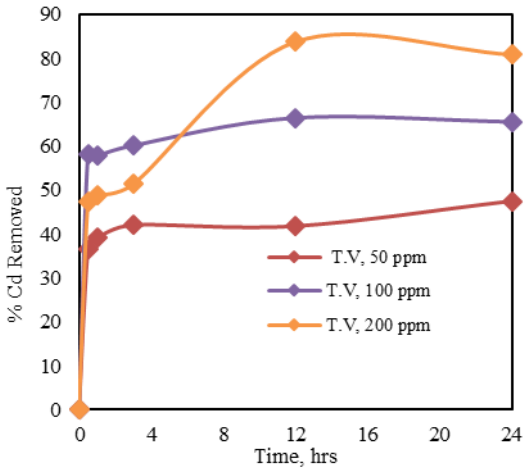

Figure 4 Effect of Concentration on Cd^{2+} Removal by *Trametes Versicolor*

3.3 Comparing the Sorption Ability of *T. Versicolor* and *P. Chrysosporium*

The sorption capacity of *T. versicolor* and *P. chrysosporium* were assessed, and as represented in Figure 5, *T. versicolor* recorded Cd^{2+} removal of 66.5%, 47.53% and 81.0% respectively for initial Cd^{2+} concentration of 50 ppm, 100 ppm and 200 ppm. *P. chrysosporium*, however, removed lower Cd^{2+} percentages at all concentrations. At 50 ppm, 100 ppm and 200 ppm, *P. chrysosporium* removed 26.2%, 46.19% and 56.69% of Cd^{2+} respectively. From the results obtained, *T. versicolor* has proven to have a higher sorption capacity than *P. chrysosporium*.

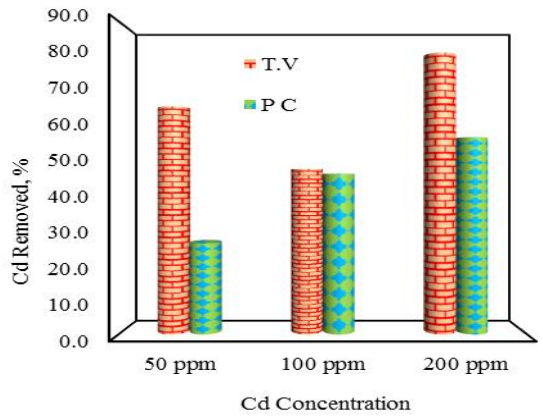

Figure 4 Comparing the Sorption Capacity and Uptake of *T. Versicolor* and *P. Chrysosporium*

In a biosorption study involving Pb^{2+}, Cr^{3+} and Zn^{2+} with *P. chrysosporium* and *T. versicolor*, Solis *et al.* (2015) reported a better biosorption ability and uptake by *T. versicolor* than *P. chrysosporium*. Several researchers have investigated the effect of pH on the biosorption by fungi and reported that maximum biosorption occurs at pH 5-7, the pH range within which this research was conducted. However, Kiran *et al.* (2012) reported an optimum activity of *P. chrysosporium* at pH 4.0-4.5 when employed in the decolourisation of reactive dye. with marked suppression above 5.5 and below 3.5. This may explain the reduced Cd^{2+} removal ability by *P. chrysosporium*.

3.4 Biosorption Kinetics of Cd^{2+}

The data obtained was modelled using the pseudo-first-order and pseudo-second-order kinetic models. The rate constants and correlation coefficients for each model and fungal biomass are presented in Table 1. In comparison, the correlation coefficients, R^2 of the pseudo-second-order model are very high for both fungi. Again, the sum of squared errors (SSE) were calculated to predict the best-fit kinetic model. Lower values of SSE indicate a better fit to the sorption data of a particular model and indicate the sorption mechanism. It is observed from Table 1 that the pseudo-second-order model gives a better fit than the pseudo-first-order model. Subbaiah *et al.* (2011) reported similar trends based on modelled data. This indicates that the sorption of Cd^{2+} by *T. versicolor* and *P. chrysosporium* is a chemisorption and monolayer coverage process.

4 Conclusions

This study focused on the biosorption ability of Cd^{2+} by *T. versicolor* and *P. chrysosporium*. Results from the study indicated an increased sorption ability of Cd^{2+} with increasing biomass sorbent and concentration for both fungi. However, *T. versicolor* proved a better biosorbent than *P. chrysosporium* at all Cd^{2+} concentrations. The kinetic data modelling revealed that the biosorption mechanism is chemisorption since the kinetic data best fitted the pseudo-second-order model. Based on the findings, it can be inferred that *T. versicolor* and *P. chrysosporium* are viable and cost-effective biomass for the biotreatment of Cd^{2+} ions from wastewater due to their high biosorption competency.

Table 1 Pseudo-first-order and Pseudo-second-order Models for the Biosorption of Cd^{2+} onto *T. versicolor* and *P. chrysosporium* Biomass

Fungi	Ini. Conc.	Pseudo-first order					Pseudo-second order				
	C_o(ppm)	$q_{e(Exp)}$	$q_{e(Cal)}$	K_1(min^{-1})	R^2	SSE	$qe_{(Exp)}$	$qe_{(Cal)}$	K_2($mgg^{-1}min^{-1}$)	R^2	SSE
Trametes versicolor	50	1.288	0.533	0.003	0.682	2.002	1.288	1.300	0.026	0.999	0.0001
	100	3.278	0.801	0.003	0.581	9.555	3.278	3.290	0.027	1.000	0.0000
	200	8.100	5.272	0.004	0.940	0.288	8.100	8.402	0.002	0.997	0.0013
Phanerochaete chrysosporium	50	1.155	0.049	-0.004	0.203	500.156	1.155	1.165	0.215	0.999	0.0001
	100	1.311	1.087	0.000	0.136	0.042	1.311	1.296	-0.019	0.996	0.0001
	200	5.669	1.745	0.001	0.258	5.057	5.669	5.612	0.009	0.995	0.0001

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