Adsorption kinetics, equilibrium and thermodynamics studies for the removal of Cadmium (II) Ions from aqueous solution by *Aspergillus nidulans*

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Abstract- The biosorption of Cd (II) ions from the aqueous solutions by *A. nidulans* strain isolated from contaminated soil was studied in batch system. The effect of pH (3, 4, 5 and 6), temperature (25, 30, 35, 40 and 45ºC) initial concentration (100 – 500 mg/ L) of Cd (II) ions on biosorption were investigated. The maximum biosorption capacity of *A. nidulans* was 258 mg/L at 350 mg/L of Cd (II) ions. The experimental results suggest that the second – order equation is the most appropriate equation to predict the biosorption capacity by *A. nidulans*. Biosorption equilibrium data was best described by Langmuir isotherm model followed by Freundlich model. The free energy change ($\Delta G°$) obtained for the biosorption of Cd (II) ions at the temperature of 298, 303, 308, 313 and 318 K was – 633, - 644, - 655, - 665 and – 676 by the fungal biomass at pH – 3. The high negative value of change in Gibbs free energy indicates the spontaneity and feasibility of the biosorption of Cd (II) by kJ/mol by *A. nidulans*.

Index Terms- *A. nidulans*, Cd (II) ions, Thermodynamics, Kinetic characterization and pH

I. INTRODUCTION

Cadmium is one of the components of the earth's crust and present everywhere in the environment. The natural occurrence of cadmium in the environment results mainly from gradual phenomenon such as rock erosion and abrasion that estimate for 15, 000 mt per annum. It is one of the toxic heavy metals with a greatest potential hazard to humans and the environment. Cadmium is used in a wide variety of industrial processes such as alloy preparation, metal plating and electronics (Akus, 1998). The major sources of Cd (II) release into the environment through wastewater streams are electroplating, smelting, paint pigments, batteries, fertilizers, mining and alloy industries (Iqbal and Edyvean, 2005) which might lead to cadmium releasing into the environment by waste streams (Holan et al., 1993). Cadmium has a half-life of 10-30 years in the human body. Therefore, their toxic effects are especially pronounced in animals of higher trophic levels, particularly in humans. It causes kidney damage, bone diseases and cancer. Chronic exposure to elevated levels of cadmium is known to cause renal dysfunction, bone degeneration, liver damage (Iqbal et al., 2007). During recent years, the study of microorganisms has contributed important insights into the basic problems an emerging technology that has received more attention in the development of biosorbents with high affinity and specificity

(Murugesan and Maheswari, 2007). Bioremediation is an emerging form of technology that uses microbes to remove or stabilize contaminants may offer a low cost and ecologically valuable means for the mitigation of heavy metal toxicity in the environment. Fungi cell wall contains large quantity of polysaccharides and proteins which offer many functional groups (such as carboxyl, hydroxyl, sulphate, phosphate and amino groups) for binding metal ions (Bai, *et al*., 2012; Chen *et al*., 2012). However, the efficiency of fungal cell as sorbents varies greatly and depends on the physiological states, surface properties of cells, pH and other physicochemical parameters of the metal solution (Sag and Kutsal, 1996). In this investigation the fungal biomass of *A. nidulans* was isolated from the contaminated soil were evaluated for their sorption efficiencies of Cd (II) ions from aqueous solution using batch systems.

II. MATERIALS AND METHODS

2.1. Isolation and Identification of Cd (II) ion resistant fungi

10 g soil sample were added into 90 ml sterile distilled water and agitated for uniform microbial suspension. Serial dilutions were performed by decimal dilutions made up to 10^{-7} from these dilutions 1ml aliquots were poured into sterile pertriplates with 15 – 20 ml of sterile Sabourd Dextrose Agar medium (Hi – media, Mumbai, India) supplemented with chlorotetracycline (10 mg l^{-1}). Plates were incubated at room temperature for 3 – 5 days. Fungal isolates were identified using the characteristics structures seen in culture which includes colonial morphology, hyphae, a-sexual spores, reproductive bodies and conidia arrangements (Alexopoulous *et al*., 1996). Slide culture techniques were used to observe morphological characteristics of fungi (Aneja, 2001).

2.2. Biosorption Studies

The biosorption of Cd (II) ions on the isolated fungi from aqueous solution containing metal ions was investigated in batch biosorption equilibrium experiments. The metal concentration of 20–500 mg L^{-1} was prepared by diluting stock solution (1000 ppm) in deionized water. The diluted solution was sterilized by filtration through a flow pore filter with a 0.45 μm pore size and was used for further preparation of metal concentration. The fungal biomass was harvested from the medium by filtration through whatman No.1 filtration paper. The

filtered fungal biomass was re-suspended in purified water for washing and again filtered as above to make sure that no media remain on the cell surface. The fungal biomass was dried in a hot air oven at 40º C for 24 h to remove the water content in the fungal biomass. Biosorption studies were done using biomass as a function of various parameters such as pH, temperature, time and initial metal ion concentration.

The effect of pH on the biosorption rate in aqueous solution (100ml) by the resistant fungi with Cd (II) ion was investigated in the pH range (3, 4, 5 and 6) (which was adjusted with HCl or NaOH at the beginning of the experiment). The general experimental procedure was repeated for various values of temperature such as (25, 30, 35, 40 and 45º C) respectively. The pH was maintained at 3 (optimum). The effect of the initial Cd (II) ion concentration on the biosorption medium (100ml) was studied at different pH and temperatures described above except that the concentration of heavy metal ion in the adsorption medium was varied between 100 to 500 mg l^{-1}. After the desired incubation period the aqueous phases were separated from the fungal biomass by centrifuged at 1000 rpm. The concentrations of remaining metal ions in biosorption medium were determined by Atomic Absorption Spectrophotometer.

2.3. Data analysis

The amount of adsorbed Cd (II) ions per unit biosorbent (mg metal ions/g dry biosorbent) was obtained by using the following expression (Demirbas *et al*., 2004).

$$q = \frac{[(C_o - C_I)\, V]}{M} \qquad (1)$$

where q is the amount of Cd (II) ions adsorbed onto the unit amount of the adsorbents (mg/g) and C_o and C_I are the concentrations of the Cd (II) ions in the aqueous solution (mg/l) before and after biosorption respectively ; V is the volume of the aqueous phase and M is the amount of the adsorbents (g)

2.4. Pseudo – first and second order equation

The study of sorption kinetics describes the adsorbate uptake rate and evidently this rate controls the residence time of adsorbate at the solid – liquid interface (Demirbas *et al*., 2004). The kinetics of Cd (II) ions sorption on fungal adsorbents was analyzed using the pseudo first order (Lagergren, 1891) and pseudo second order.

The Pseudo first order equation (Lagergren, 1891) is generally expressed as follows,

$$\log(q_{eq} - q_t) = \frac{\log q_{eq} - (k_1 t)}{2.303} \qquad (2)$$

A plot of Log $(q_{eq} - q_t)$ against t should give a straight line to confirm the applicability of the kinetic model. In a true first - order process q_{eq} should be equal to the intercept of a plot of Log $(q_{eq} - q_t)$ against t

If the rate of sorption is a second order mechanism, the pseudo second order chemisorption kinetics rate equation (Ho and Mckay, 1998) is expressed as Eq, 4

$$\frac{t}{q_t} = \frac{1}{(k_2 q^2 eq)} + \frac{1}{q_{eq}}\, t \qquad (3)$$

A plot of $t\,/\,q_t$ vs t should give a linear relationship for the applicability of the second order kinetics. The rate constant (k_2) and adsorption at equilibrium (q_{eq}) can be obtained from the intercept and slope respectively.

2.5. Adsorption isotherms

Sorption data is analyzed by widely used sorption models, such as the Langmuir or Freundlich isotherm, which relate sorption density qe (metal uptake per unit weight of sorbent) to equilibrium sorbate concentration in the bulk fluid phase, Ce.

$$\frac{C_e}{q_e} = \frac{1}{q_m k_a} + \frac{C_e}{q_m} \qquad (4)$$

Where qe is the amount adsorbed per unit mass of adsorbent (mg/g), Ce the equilibrium concentration of the adsorbate (mg/l), qm the equilibrium sorption capacity for complete monolayer (mg/g) and Ka the sorption equilibrium constant (l/mg). When Ce/qe was plotted against Ce, a straight line with slope 1/Ka qm and an intercept of 1/qm were obtained

The Freundlich equation is described by the following equation

$$q_{eq} = K_F\, Ce^{1/n} \qquad (5)$$

q_e – Metal uptake at equilibrium concentration (mg / g) ; C_e – Equilibrium metal ion concentration, (mg / g) ; K_F – Freundlich's constant of adsorption capacity ; n – Freundlich's constant of adsorption intensity. The K_F was estimated from the y – intercept and n was calculated from the slope.

2.6. Thermodynamics of biosorption of Cd (II) ions

In the present study, the biosorption experiments were carried out in the temperature (25, 30, 35, 40 & 45ºC). The values of the thermodynamic parameters such as ΔGº, ΔHº and ΔSº, describing Cd (II) ions uptake by *A. nidulans* were calculated using the thermodynamic equations (Fraiji *et al*., 1992). The biosorption process can be regarded as a heterogeneous and reversible process at equilibrium. The change in Gibbs free energy of the biosorption process is thus given as

$$\Delta G° = -RtlnK_c \qquad (6)$$

Where ΔGº is the standard Gibbs free energy change for the biosorption (J / mol), R the universal gas constant (8.314 J/mol/K) while T is the temperature (K). The effect of temperature on the biosorption of Cd (II) ions by *A. nidulans* from thermodynamics,

$$\Delta G° = \Delta H° - T\Delta S° \qquad (7)$$
$$\text{or}$$
$$\Delta G° = -\Delta S° (T) + \Delta H° \qquad (8)$$

A plot of T against ΔGº gives a straight line with slope –ΔSº and an intercept of ΔHº was obtained

III. RESULTS

### 3.1. Identification of cadmium resistant fungi

The ability of isolated cadmium resistant fungal biomass was to determine biosorbs Cd (II) ions from aqueous solution. In this preliminary screening the total count of fungus in the soil sample was ranged from $123 \pm 7.31 \times 10^2$ to $3.7 \pm 0.23 \times 10^7$ CFU g^{-1}. Fungi with different colony morphology were selected, purified and stored in the Sabourd Dextrose Agar

medium (Hi – media, Mumbai, India) supplemented with chlorotetracycline (10 mg ml^{-1}l). The dominant cadmium resistant isolate was grown well at 35° C and they show green brown shade with velvety texture on sabourd dextrose agar plate. The colony consisted of a cinnamon brown color of conidiophore and narrows down towards the foot cell. Conidia are short and columnar, globose and are rough. The sterigmata developed in two series as primary (5 – 6 µm x 2-3 µm) and secondary (narrower) sterigmata. Based on the microscopic observation the isolate was identified as *A. nidulans*. The resistant fungal mycelium was cultivated in fungal broth media amended with various concentration of Cd (II) ion. The resistant fungal mycelium isolated from the soil was cultivated in fungal broth media amended with various concentration of Cd (II) ion (100– 500 mg l^{-1}).

3.2. *Biosorption rate of Cd (II) ion at different pH*

The biosorption capacity of Cd (II) ion at different pH by *A. nidulans* from solution containing 200 mg l^{-1} of Cd (II) ion is shown in Fig - 1. As seen from the figure the saturation level was obtained after about 240 min. Biosorption capacity of *A. nidulans* increased at different pH (3, 4, 5 and 6) with increasing the time. As seen from Fig – 1 the amount of biosorbed Cd (II) ion at pH 3, 4, 5 and 6 was found to be 107, 96, 75 and 60 mg g^{-1} respectively after 240 min.

Fig – 1 Biosorption rate of Cd (II) ions at different pH by *A. nidulans* from aqueous solution. Adsorption conditions: Initial concentration of Cd (II) ions - 200 mg l^{-1}; Biomass concentration – 0.1g
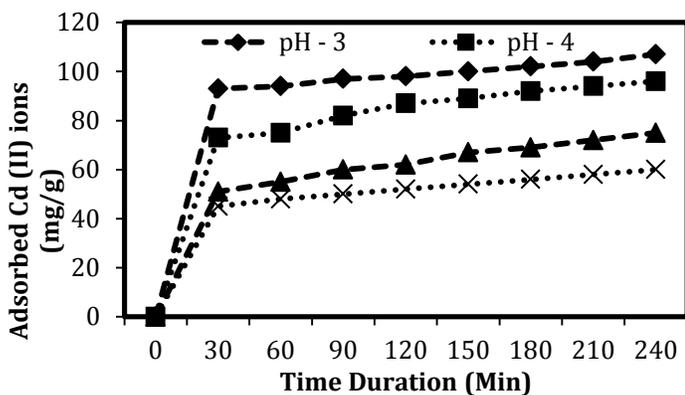
3.3. *Effect of initial Cd (II) ion concentration on biosorption*

The biosorption capacity of fungal biomass at equilibrium concentration of Cd (II) ions in heavy metal solution is shown in Fig – 2. The biosorption capacity of the tested adsorbent was presented as a function of the initial concentration of Cd (II) ions (100 – 500 mg l^{-1}). Biosorption capacity of *A. nidulans* increased with increasing initial concentration of Cd (II) ions in the solution and reached a saturated value of 350 mg l^{-1}. As seen from the Fig – 2 the amount of biosorbed Cd (II) ions on the *A. nidulans* was found to be 258 mg g^{-1}.

3.4. *Biosorption kinetic and equilibrium modeling*

The comparison of experimental biosorption capacity and the theoretical values at different pH and temperatures were estimated from the first and second order rate equations are represent in Table 1a&1b. The theoretical q$_{eq}$ values estimated

from the first order kinetics model gave significantly different values when compared to experimental values and the correlation coefficients for the linear plots of log (qe-qt) against t were also found to be slightly lower than 0.995 for both pH and temperatures. The correlation coefficients for the linear plots of t / qt against t for the second order equation was greater than 0.995 for *A. nidulans* for contact times of 270 min at different pH and temperatures. The Langmuir constants (q$_m$ and k$_d$) along with correlation coefficients (R^2) have been calculated from the plots for biosorption of Cd (II) ion on the biosorbents and the results is given in Table 2. The maximum capacity q$_m$ determined from the Langmuir isotherm defines the total capacity of the biosorbents for Cd (II) ion. It is clear that this increase in the q$_m$ value is due to an increase in the adsorptive sites on the biosorbents

Fig – 2 Biosorption capacities of Cd (II) ions by *A. nidulans*. Biosorption conditions: volume of biosorption medium – 100ml; biomass concentration 0.1g; pH – 3; temperature - 35˚C
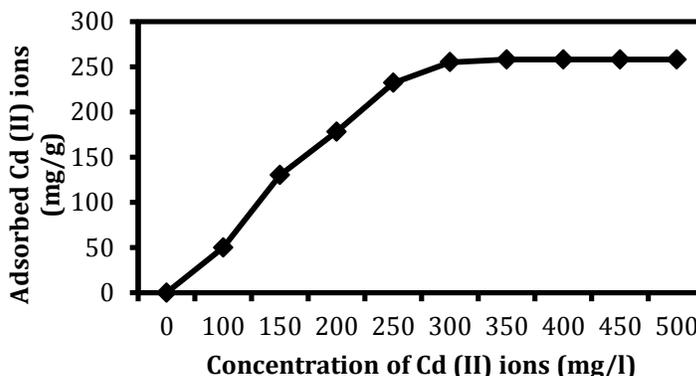
3.5. *Thermodynamics of biosorption of Cd (II) ions*

The thermodynamic of biosorption of Cd (II) ions by *A. nidulans* the value of the entropy and enthalpy was 0.007 J/mol/K and 2.127 J/mol/K, respectively. The free energy change (ΔG°) obtained for the biosorption of Cd (II) ions by *A. nidulans* at the temperatures of 298, 303, 308, 313 and 318 K was found to be – 633, - 644, - 655, - 665 and – 676 kJ/mol respectively at pH – 3 (Fig – 3). The negative values of ΔG° indicate the feasibility of the process and indicate spontaneous nature of the sorption. The positive values of ΔS° show the increased randomness at the solid / solution interface during the sorption also reflects the affinity of the sorbent for Cd (II) ions.

Fig – 3 The free energy plot for the biosorption of Cd (II) ions using *A. nidulans* at pH - 3; initial metal ion concentration of 200 mg l^{-1}
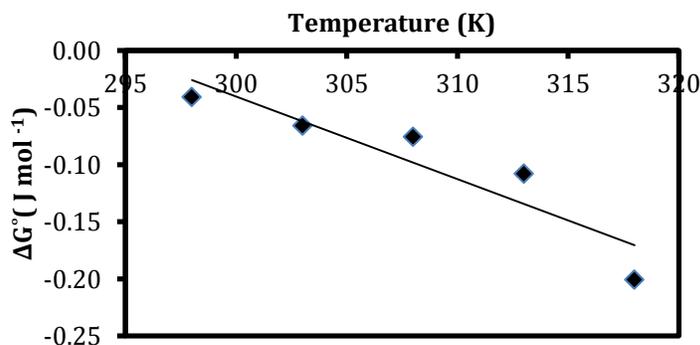
Table 1a The Pseudo first kinetics for biosorption of Cd (II) ion from aqueous solution by *A. nidulans* at different pH

Biosorbent at different pH	Experimental q_{ex} (mg g^{-1})	Pseudo first order		
		q_{eq} (mg g^{-1})	$k_1 \times 10^{-3}$ (min^{-1})	R^2
pH - 3	105	277	4.74	0.938
pH - 4	96	185	8.43	0.948
pH - 5	80	208	7.65	0.952
pH - 6	92	222	6.20	0.937

Table 1b The Pseudo second order kinetics for biosorption of Cd (II) ion from aqueous solution by *A. nidulans* at different pH

Biosorbent at different pH	Experimental q_{ex} (mg g^{-1})	Pseudo second order		
		q_{eq} (mg g^{-1})	$k_2 \times 10^{-3}$ (min^{-1})	R^2
pH - 3	105	108	0.83	0.998
pH - 4	96	102	1.82	0.997
pH - 5	80	81	4.27	0.992
pH - 6	92	63	5.01	0.995

Table – 3 Langmuir and Freundlich Isotherm model constant and correlation co efficient for biosorption of Cd (II) ion from aqueous solution by *A. nidulans*

Langmuir Constant		Freundlich Constant	
q_m (mg g^{-1})	588	K_F	1.15
$k_d \times 10^{-4}$ (M)	3.12	n	0.76
R^2	0.859	R^2	0.984

IV. DISCUSSION

The maximum biosorption capacity of *A. nidulans* from solution containing 200 mg l^{-1} of Cd (II) ion is at pH – 3. The biosorption of Cd (II), Pb(II) and Cu (II) on inactivated *P.chyrsosporium* was pH dependent and maximum biosorption was obtained at pH – 6 (Say *et al*., 2001) . The uptake of cadmium by *Spirulina platensis* at a period of 24 hrs showed up take to be maximum during the initial period of contact at 1.6 mg/L Cd (II) ions (Murugesan *et al*., 2008).The biosorption equilibrium time of chromium (IV) on the dead and immobilized biomass of *R. arrhizus* was 2h (Prakasham *et al*., 1999). The lead biosorption rate on *P. chrysosporium* is fast and reached saturation value within 2h

(Yetis *et al*., 2000). Ana Claudia C. Nascimento *et al*., (2015) revealed that 53%, 56.8% and 66.9% of 1mM of cadmium was removed by *Aspergillus nidulans* at pH values of 4.0, 5.0 and 6.0, respectively by native washed biomass. Maheswari and Murugesan (2011, 2016) studied the equilibrium biosorption of As(III) ions for all the fungal biomass preparations were similar at pH 4. From this study the biosorption capacity of *A. nidulans* increased with increasing initial concentration of Cd (II) ions in the solution and reached a saturated value of 350 mg l^{-1}. The higher biosorption capacity was obtained in fungal biomass may be increases the availability of the binding sites by fixing the soluble protein in the cell wall after denaturation with heat (Maheswari and Murugesan, 2009b, 2010). The biosorption capacity of dead *Fusarium flocciferum* was 19.2 mg Cd (II) g^{-1} dry biomass (Delgado *et al*., 1998). The fungal biomass of white rot fungus *Phanerochaete chrysosporium* used for heavy metal removal from artificial waste water had a cadmium capacity of the dry fungal biomass of 23.4 mg Cd/g (Say *et al*., 2001). The adsorption capacity of *Rhizopus arrhizus* was 78 mg for Fe (III), 71 mg for Pb (II) and 62 mg for Cd (II) /g dry biomass (Ozer *et al*., 1997). Raman Kumar *et al*, (2014) showed the maximum uptake (13.15 mg/g) of Cd was observed in *Rhizopus arrhizus* and the minimum uptake of Cd (0.51 mg/g) was observed in *Penicillium chrysogenum*. *A. niger* and *A. parasiticus* species were reduced the Cr(VI) concentration of 96.3 % and 91.6 % respectively from an initial concentration of 20 mg/Lin 72 h (Shugaba *et al*., 2012). The percentage removal of chromium was around 89% and 85% with initial metal ion concentration of 5mg/L for *Aspergillus oryzae* and *A.sojae* respectively. The percentage removal decreased as the concentration increased and it is due to lack of active sites (Reya Issac et al., 2012). *Aspergillus* sp. was more tolerant to Cu, Cd and Ni metal ions than other fungi species such as *Curvularia, Acrimonium* and *Pithyum* (Akhtar *et al*., 2013). The theoretical q_{eq} values for different pH and temperatures were very close to the experimental q_{eq} values in the case of pseudo second order kinetics. These results suggest that the second – order mechanism is predominant and the chemisorptions may be the rate limiting step that controls the biosorption process (Allen *et al*., 2005). Therefore the results of this investigation showed that the biosorption systems best described in pseudo second order kinetic model. The living organisms induce the production of metallothioneins which are protein that contain large amounts of cystein and bind heavy metal ions in order to respond to the effects of heavy metals (Hafez *et al*., 1997). The Langmuir constant (k_d) estimated from the intercept is a measure of the stability of the complex formed between metal ions and adsorptive surface layer of the biosorbents under specified experimental conditions. The Freundlich constants K_F and n shows easy separation of metal ions from aqueous medium and indicate favorable adsorption (Cetinkaya *et al*., 1999). In the present investigation it is clear that freundlich model fits well than Langmuir isotherm model.

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