



Biosorption of Copper and Lead from Human Blood Plasma Using *allium cepa*

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Abstract: Lead and Copper are inorganic pollutants that are non-biodegradable. When released into the environment, they have adverse effect on human beings and animals when ingested. *Allium cepa* (Onion) is a vegetable product largely consumed for its potential therapeutic importance. This study investigates the efficacy of *Allium cepa* in removal of Cu^{2+} and Pb^{2+} from human blood plasma *in-vitro*. The adsorption rate was studied at optimum values of pH 6 for Cu^{2+} , and pH 4 for Pb^{2+} , metal ion concentration (50mgL^{-1}), and *Allium cepa* dose 0.60g for Cu^{2+} and 0.40g for Pb^{2+} at physiological temperature (38°C). SEM analysis revealed physical disintegration in the surface morphology of *Allium cepa* biomass after adsorption. Equilibrium sorption occurred at 80 mins with 99.316% removal for Cu^{2+} and 90mins with 99.8914% removal for Pb^{2+} . The adsorption data obtained for Cu^{2+} best fits Temkin isotherm with correlation value (R^2)=0.901 with low binding energy (A_T)= 1.340 Lg^{-1} while the adsorption data for Pb^{2+} best fits Freundlich isotherm with correlation values (R^2)=0.965 at Freundlich coefficient (k_f)= $3.169.27\text{ Lmg}^{-1}$. The result also revealed that physisorption and chemisorption occurred between the metal ions and binding site on the *Allium cepa* biomass as Intraparticle diffusion proved not to be the only rate controlling step.

Keywords: *Allium cepa*, Biosorption, Freundlich Isotherm, Heavy Metals, SEM and Temkin Isotherm

1. Introduction

Chemicals have become an essential part of human life, sustaining activities and development, preventing and controlling many diseases and increasing agricultural yield [1]. Despite their benefits, they can cause adverse effects on human health especially when misused or inappropriately disposed. Heavy metals are among the major constituents of these chemicals. They are harmful because they are non-biodegradable and have the ability to bioaccumulate in different body parts [2]. When heavy metals enter the human system, they are transported to various parts of the body through the blood plasma which is a medium for excretory product transportation [2].

Biosorption is a method that utilizes biological materials to remove heavy metals from aqueous solution. Its

advantages over conventional treatment method include low cost, regeneration of biosorbent, possible metal recovery and high efficiency amongst others [1]. Several studies have reported the potential of biological material to bind heavy metals. Amongst these biomaterials is *Allium cepa* which is highly medicinal [3]. It contains active groups such as carboxylic, sulfate, amino, amide and hydroxyl groups that play an important role in the biosorption process [3]. However, lots of biosorption studies have been carried out. For instance Sargassum seaweed as biosorbent for heavy metals [4]; A comparative study on heavy metals biosorption characteristics of some algae [5]; Adsorption of metals by biomaterials derived from the marine alga *Ecklonia maxima* [6]; Biosorption of cadmium by brown, green and

red seaweeds [7]; Biosorption of lead and nickel by biomass of marine algae [8]; Biosorption of toxic metals from aqueous solutions by bacteria strains isolated from metal-polluted soils [9]; Biosorption of chromium (VI) from aqueous solution by the husk of Bengal gram (*Cicerarientinum*) [10]; Biosorption of copper (II) and cobalt (II) from aqueous solution by crab shell particles [11] amongst others.

Copper and lead were selected for this study been adduced to the wide range application in chemical industries. These metals can accumulate along food chain and are non-biodegradable [9]. Exposure to these metals even at low concentration has adverse effect on human and animal health [2]. This study seeks to investigate the biosorption potential of *Allium cepa* on Cu^{2+} and Pb^{2+} removal from the human blood plasma solution. The effect of pH, biosorbent dosage and metal ion concentration will be optimized and used to study the rate of biosorption. The data obtained will be fitted into Freundlich isotherm, Temkin Isotherm and Intraparticle diffusion model.

2. Materials and Methods

Sample Collection and Preparation

The human blood sample was obtained from the Department of Hematology, Ahmadu Bello University Teaching Hospital, Zaria, Kaduna State, Nigeria. The human blood Plasma was extracted from the human blood sample by centrifugation. The *Allium cepa* was obtained from samara market, Kaduna state, Nigeria. The *Allium cepa* bulb was carefully wash with distilled water and rewashed with de-ionized water, sliced and air dried for a period of 7 days. The dried sample was repeatedly washed with de-ionized water and oven dried for 72 hrs at 60°C to a constant mass, pulverized using agate mortar and sieved with a 0.25-0.50 mm mesh sieve and stored in an air tight plastic container prior to analysis [12].

Scanning Electron Microscope (SEM) analysis of Allium cepa

The surface morphology was determined before and after biosorption using SEM. The adsorbent was glued to 10mm diameter metal mounts and coated with gold under vacuum in an argon atmosphere. The data was recorded over a selected area of the surface of the sample and a two dimensional image was generated as described by [13].

Assessment of Cu^{2+} and Pb^{2+} in Samples

The samples (Human blood plasma and *Allium cepa* biomass) were digested respectively using conventional wet acid digestion as described by [12] and the their filtrates analyzed for Cu^{2+} and Pb^{2+} respectively using Agilent model 3510 Atomic Adsorption Spectrophotometer (AAS).

Biosorption Data Evaluation

The percentage sorption of Cu^{2+} and Pb^{2+} were determined using equation 1

$$\text{Biosorption (\%)} = \frac{C_o - C_e}{C_e} \times 100 \quad (1)$$

Where C_o and C_e are the initial and equilibrium Cu^{2+} and Pb^{2+} concentrations in human blood plasma respectively. The amount of Cu^{2+} and Pb^{2+} removed from the human blood plasma were calculated using equation 2.

$$Q_e = \frac{(C_i - C_e)V}{m} \quad (2)$$

Where q_e is the amount of Cu^{2+} and Pb^{2+} bind onto the biosorbent (mg g^{-1}) at equilibrium, C_i and C_e are the initial and final concentration of the metal ion in the human blood plasma (mg L^{-1}), V is the volume of the human blood plasma (cm^3) and m is the amount (g) of biosorbent used.

Effect of pH

The effect of pH was studied over a pH range of 2 to 10. The study was carried out by introducing 0.60g of biosorbent into separate 250 cm^3 conical flasks containing 20 cm^3 human blood plasma solution spiked with 50 mg L^{-1} Cu^{2+} and 50 mg L^{-1} Pb^{2+} solutions respectively. The pH of the mixture was adjusted to pH 2 with 0.1M HNO_3 . The mixture was agitated for 100 mins at 200rpm using a rotary shaker as described by [3]. The adsorbent was separated from the mixture with Whatman No 11 filter paper. The filtrate was digested and analyzed for residual Cu^{2+} and Pb^{2+} respectively using AAS. An equilibrium pH 6 and 4 was obtained for Cu^{2+} and Pb^{2+} respectively for further study.

Effect of Metal ion concentration

Equilibrium metal ion concentration was studied by introducing 0.60g biosorbent into 250 cm^3 conical flask containing 20 cm^3 human blood plasma spiked 20 mg L^{-1} Cu^{2+} solution. The mixture was adjusted to an equilibrium pH 6 using 0.1M HNO_3 and agitated for 100 mins at 200rpm. The adsorbent was separated from the mixture with Whatman No 11 filter paper. The filtrate was digested and analyzed for residual Cu^{2+} using AAS. This procedure was repeated for Cu^{2+} concentrations of 30 mg L^{-1} , 40 mg L^{-1} , 50 mg L^{-1} , 60 mg L^{-1} and 70 mg L^{-1} respectively. The entire procedure was repeated using equilibrium pH 4 for Pb^{2+} concentration. An equilibrium concentration of 50 mg L^{-1} was obtained for both Cu^{2+} and Pb^{2+} .

Effect of Biosorbent dosage

Equilibrium biosorbent dosage was studied by introducing 0.20 g of biosorbent into 250 cm^3 conical flask containing 20 cm^3 human blood plasma spiked with equilibrium copper concentration (50 mg L^{-1}). The mixture was adjusted to equilibrium pH 6 and agitated for 100 mins at 200 rpm. The adsorbent was separated from the mixture with Whatman No 11 filter paper. The filtrate was digested and analyzed for residual copper using AAS. This procedure was repeated for biosorbent dosage of 0.40 g, 0.60g, 0.80g and 1.00 g. This procedure was repeated for Pb^{2+} using equilibrium pH 4 and equilibrium concentration (50 mg L^{-1}). An equilibrium biosorbent dose of 0.60 g for Cu^{2+} and 0.40 g for Pb^{2+} was obtained.

Rate of biosorption

Equilibrium sorption rate was investigated by introducing 0.60 g of biosorbent into 250 cm^3 conical flask containing 20 cm^3 human blood plasma spiked with equilibrium Cu^{2+}

concentration (50 mgL^{-1}), adjusted to equilibrium pH 6 using 0.1HNO_3 and agitated for 10 mins at 200 rpm with a rotary shaker [14]. The procedure was repeated for 30 mins, 40 mins, 50 mins 60 mins, 70 mins, 80 mins 90 mins 100 mins, 110 mins and 120 min. The procedure was repeated for Pb^{2+} at equilibrium of pH 4, biosorbent dose 0.40 g and Pb^{2+} concentration 50 mgL^{-1} .

3. Result and Discussion

Scanning Electron Microscope (SEM)

SEM micrograph of the native and heavy metal (Copper and Lead) treated biosorbent are presented in Figures 1, 2 and 3. The result revealed an uneven surface texture with a lot of irregularities in the surface morphology for the native biosorbent. The SEM micrograph Cu^{2+} treated biosorbent showed remarkable physical disintegration resulting to emergence of protrusions and rough surface area which is an evidence of high metal uptake. The SEM micrograph for Pb^{2+} treated biosorbent showed more disintegration and rough surface than Cu^{2+} treated biosorbent been added to the high surface interaction between the binding sites on the surface of the *Allium cepa* biomass and Pb^{2+} [15].

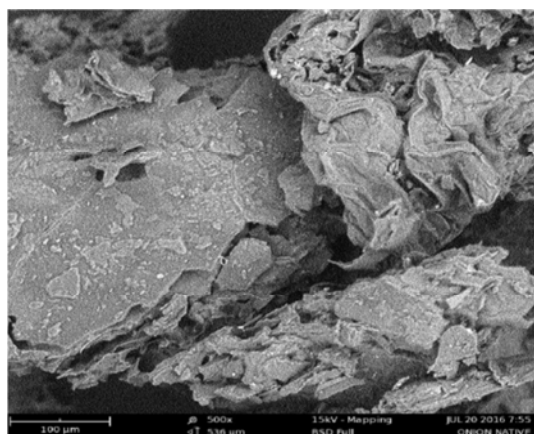


Figure 1. SEM micrograph of *Allium cepa* Biomass.

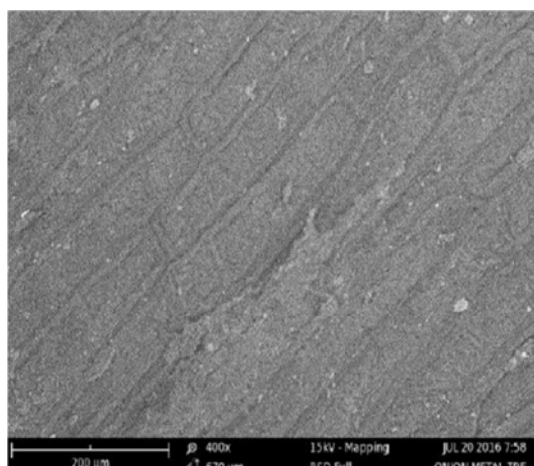


Figure 2. SEM micrograph for copper treated biosorbent.

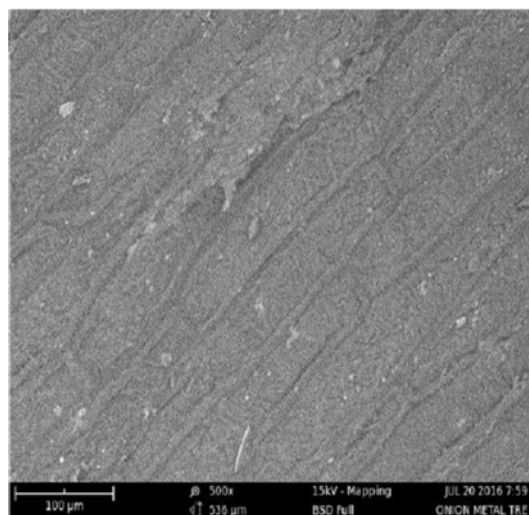


Figure 3. SEM micrograph for lead treated biosorbent.

Equilibrium Adsorption pH

Effect of pH on Cu^{2+} and Pb^{2+} sorption is presented in Figure 4. The result revealed an increase in percentage removal of Cu^{2+} and Pb^{2+} respectively. The equilibrium sorption was observed at pH 6 for Cu^{2+} and pH 4 for Pb^{2+} . After equilibrium pH for the metal ions was attained, desorption occurred with further increase in pH of the solution which indicates high level of interaction between the metal ions and binding sites on the *Allium cepa* biomass as the solution tends towards acidic medium.

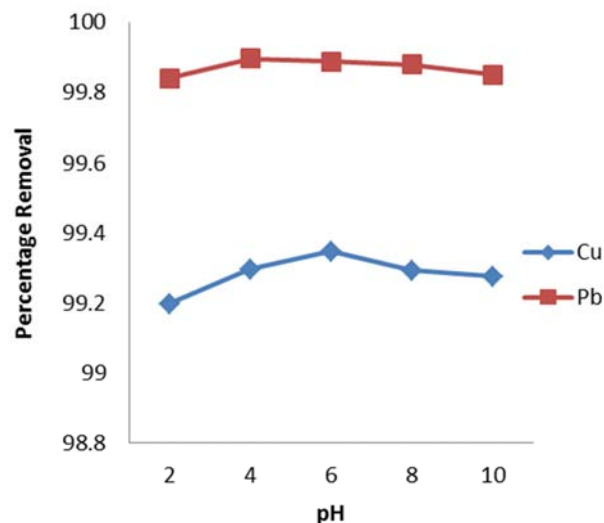


Figure 4. Percentage removal of Copper and Lead as a function of pH.

Effect of Metal ion concentration

The effect of concentration on Cu^{2+} and Pb^{2+} sorption is presented in Figure 5. The result showed an increase in percentage removal from 20 mgL^{-1} to 50 mgL^{-1} . The equilibrium concentration for Cu^{2+} and Pb^{2+} sorption occurred at 50 mgL^{-1} with percentage removal of 99.19% and 99.896 % respectively. The data obtained showed a high interaction between the metal ions and the biosorbent [14].

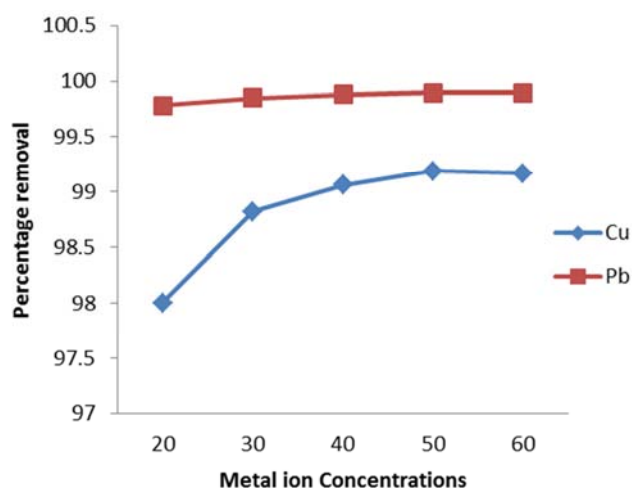


Figure 5. Percentage removal of Copper and Lead as a function of concentration.

Equilibrium Biosorbent dosage

The result in Figure 6 revealed that maximum sorption of copper and lead occurred at 0.60 g and 0.40 g at percentage sorption of 99.34 % and 99.89 % respectively. The result showed that at high biosorbent dosage the available metal ions are insufficient to cover the entire exchangeable site on the biosorbent therefore resulting to low metal sorption [11].

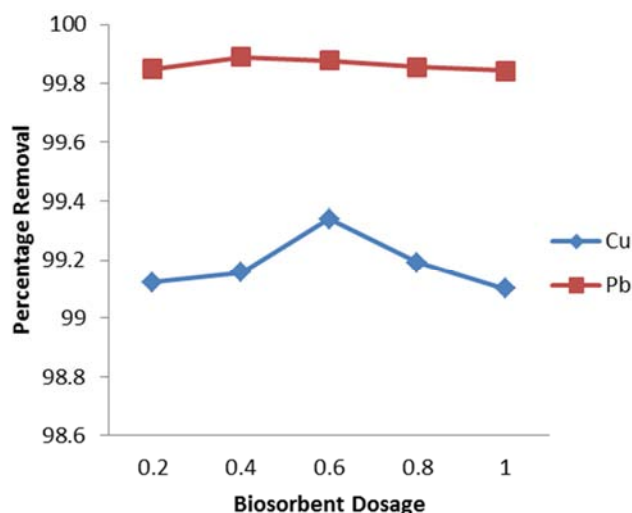


Figure 6. Percentage removal of Copper and Lead as a function of Biosorbent dosage.

Rate of Biosorption

The effect of contact time on Cu^{2+} and Pb^{2+} sorption are presented in Figure 7. The result showed a significant increase in Cu^{2+} sorption with equilibrium sorption at 80 mins (99.316 % removal) while Pb^{2+} sorption showed a slight increase with equilibrium sorption at 90 mins (99.8914% removal). Further increase in contact time resulted to release of the adsorbate (Desorption), been adduced to unavailability of free binding sites for metal ion uptake [16].

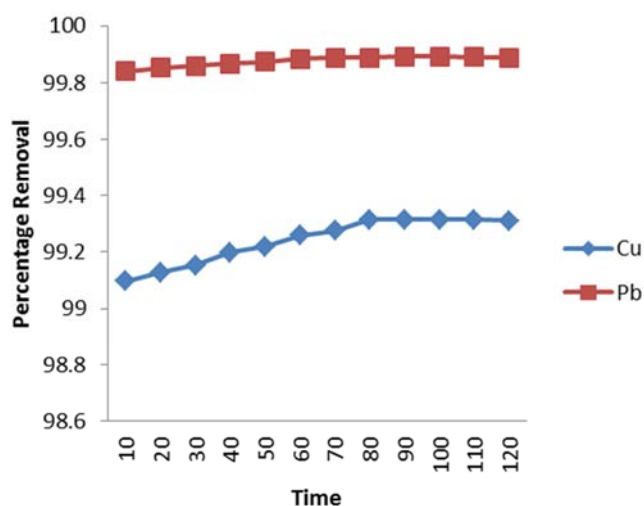


Figure 7. Percentage removal as a function of Time.

Biosorption Isotherm

The biosorption isotherm reveals the relationship between equilibrium concentration of adsorbate in the solution and the biosorbent at constant temperature [2]. The data obtained from the percentage removal of Cu^{2+} and Pb^{2+} with contact time was fitted into Freundlich and Temkin isotherms respectively.

Freundlich Isotherm

This isotherm model describes non ideal sorption onto heterogeneous surface involving multilayer sorption [15]. The isotherm model is expressed in equation 3

$$q_e = K_f C_e^{1/n} \quad (3)$$

The linearized form is expressed in equation 4

$$\log Q_e = \log K_f + 1/n \log C_e \quad (4)$$

Where Q_e is the amount of adsorbate adsorbed per unit mass of biosorbent, K_f is the freundlich constant measuring adsorption capacity (L/mg), C_e is equilibrium concentration of the adsorbent in solution (mg/L), n is constant related to adsorption efficiency and adsorption intensity of the biosorbent. K_f is dependent on the units upon which Q_e and C_e are expressed [16].

The Freundlich isotherm for Cu^{2+} and Pb^{2+} sorption are presented in Figures 8 and 9. The result revealed that experimental data obtained for Cu^{2+} and Pb^{2+} sorption conforms to Freundlich isotherm. However the level of conformity is more in Pb^{2+} as indicated by the correlation coefficient value $R^2 = (0.965 \text{ and } 0.217)$ for Pb^{2+} and Cu^{2+} respectively. The values of k_f (0.3908 L/mg and 3,169.57 L/mg) for Cu^{2+} and Pb^{2+} respective indicates that there was a high adsorption of the Pb^{2+} per cite and a low adsorption of Cu^{2+} onto the biosorbent surface. The values of n (5.714 and 0.403) for Cu^{2+} and Pb^{2+} respectively indicates a favourable Cu^{2+} sorption and unfavourable Pb^{2+} sorption since favourable sorption occurs at $1/n < 1$ [16].

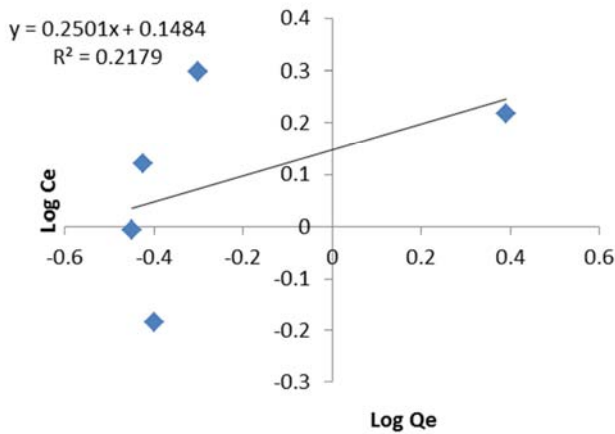


Figure 8. Freundlich isotherm for Cu^{2+} sorption.

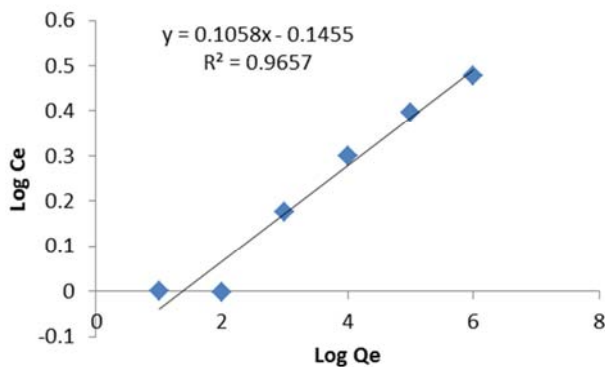


Figure 9. Freundlich isotherm for Pb^{2+} sorption.

Temkin Isotherm

Temkin isotherm takes into cognizance the occupation of the more energetic adsorption sites [17]. Besides the low and large magnitude of concentrations, the model assumes that heat of adsorption by all molecules in a layer would decrease linearly rather than logarithmically with coverage [18].

This is as expressed in equation 5

$$q_e = RT/b_T \ln(AC_e) \quad (5)$$

The linearized Temkin isotherm is presented in equation 6

$$q_e = B \ln A_T + B \ln C_e \quad (6)$$

where $RT/b_T = B$, T (K) is the temperature, R is the ideal gas constant ($8.314 \text{ J mol}^{-1} \text{ K}^{-1}$), A (L/g) is the equilibrium binding energy, B (J/mol) is a constant related to heat of sorption and b_T is the Temkin isotherm constant. The linear plot of q_e versus $\ln C_e$ is the Temkin isotherm for the biosorption data presented in Figure 10 and Figure 11 for sorption of Cu^{2+} and Pb^{2+} respectively. From the result obtained $b_T = (52.0307$ and $86.220)$ for Cu^{2+} and Pb^{2+} sorption respectively, which suggest a high heat of adsorption [19]. Equilibrium binding energy $A_T = (1.340 \text{ Lg}^{-1}$ and $3.90 \text{ Lg}^{-1})$ for Cu^{2+} and Pb^{2+} respectively, indicates low binding energy between the metal ions and the active site on the biosorbent [20]. The correlation coefficient $R^2 = (0.901$ and $0.508)$ values for Cu^{2+} and Pb^{2+} respectively indicating conformity to

Temkin isotherm, with Cu^{2+} showing better fit as indicated by the $R^2 = 0.901$.

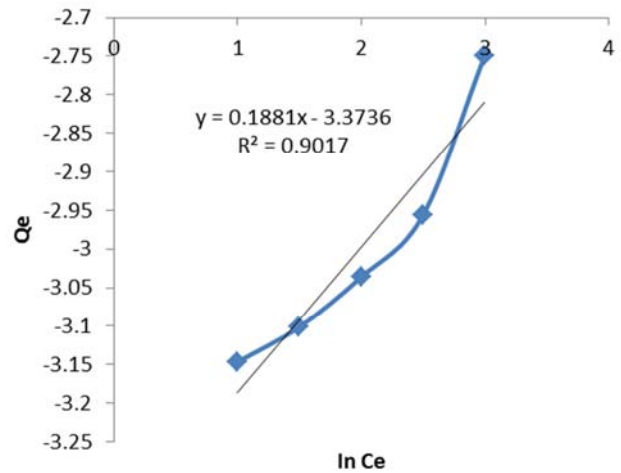


Figure 10. Temkin Isotherm for Cu^{2+} sorption.

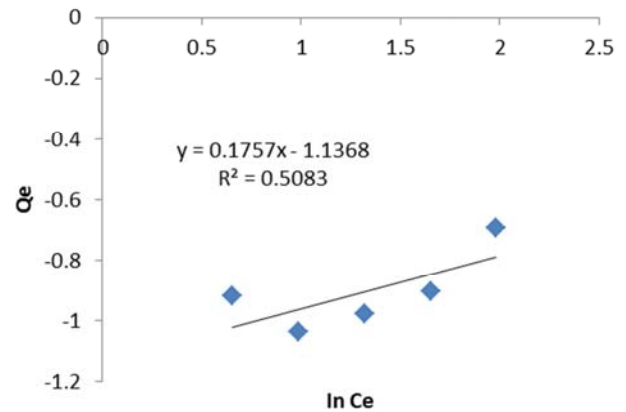


Figure 11. Temkin Isotherm for Pb^{2+} sorption.

Kinetic Study

Intraparticle Diffusion

Intraparticle diffusion is employed to identify diffusion mechanism and rate controlling steps that affect the adsorption process [21]. The rate of Intraparticle diffusion is calculated as expressed in equation 7.

$$qt = k_{ID} t^{1/2} + C_{ID} \quad (7)$$

where, C_{ID} is the intercept and K_{ID} is the Intraparticle diffusion rate constant ($\text{mg/g min}^{1/2}$). A plot of uptake (qt) versus ($t^{1/2}$) should be linear if Intraparticle diffusion is taken place in the adsorption process and if the line passes through the origin then Intraparticle diffusion is the rate determining step [22]. The experimental data obtained for Intraparticle diffusion of Cu^{2+} and Pb^{2+} sorption are presented in Figures 12 and 13. The result revealed that a plot of qt versus $t^{1/2}$ did not pass through the origin suggesting that Intraparticle diffusion is not the rate determining step [23]. However the data obtained conforms to Intraparticle diffusion as indicated by correlation coefficient value $R^2 = (0.900$ and $0.832)$ for Cu^{2+} and Pb^{2+} respectively, with Cu^{2+} showing best fit as

indicated by the $R^2=0.900$ for Cu^{2+} sorption. The positive intercept $C_{iD}=(1.649$ and $1.663)$ indicates some degree of boundary layer control [24].

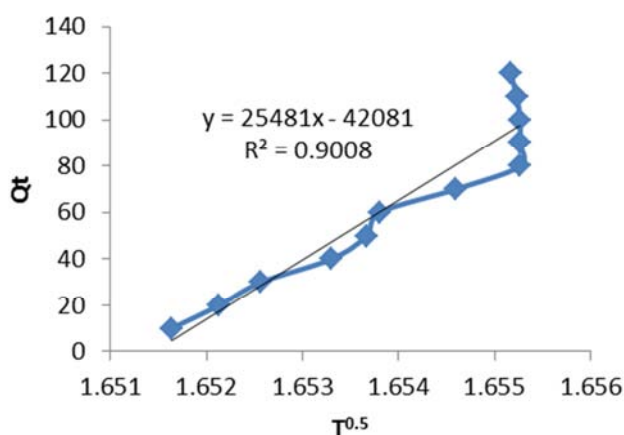


Figure 12. Intraparticle diffusion model for Cu^{2+} sorption.

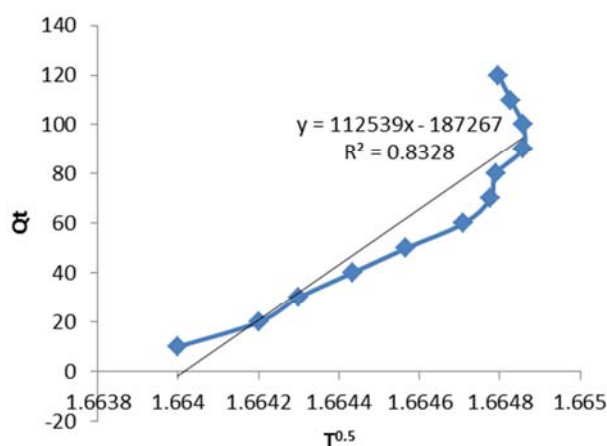


Figure 13. Intraparticle diffusion model for Pb^{2+} sorption.

Table 1. Isotherm and Kinetic Parameters for Biosorption of Cu^{2+} and Pb^{2+} .

Models	Parameters	Values	
		Cu^{2+}	Pb^{2+}
Freundlich	K_f	0.3908	3,169.57
	N	5.714	0.403
	R^2	0.217	0.800
Temkin	b_T	86.220	52.0307
	A_T	1.340	1.340
	R^2	0.901	0.900
Intraparticle diffusion	K_i	0.000	0.000
	C_{iD}	1.649	1.663
	R^2	0.900	0.832

4. Conclusion

This study has revealed that *Allium cepa* biomass has favourable properties for removal of Cu^{2+} and Pb^{2+} from the human blood plasma in-vitro. The biosorption of Cu^{2+} and Pb^{2+} was dependent on the pH, concentration of Cu^{2+} and Pb^{2+} , amount of *Allium cepa* biomass used and contact time. The biosorption of Cu^{2+} and Pb^{2+} was found to be equilibrium at pH 6 for Cu^{2+} and pH 4 for Pb^{2+} , metal ion

concentration of 50 mgL^{-1} , biosorbent dose of 0.60g for Cu^{2+} sorption and 0.40g for Pb^{2+} sorption, contact time of 80 mins for Cu^{2+} and 90 mins for Pb^{2+} . Temkin and intraparticle diffusion showed fitted best for the adsorption data of Cu^{2+} while Freundlich isotherm showed best fitted best for the adsorption data of Pb^{2+} . The information obtained can be useful in heavy metal removal from the human body in-vivo using natural products. However there is need for further study to investigate the effectiveness of *Allium cepa* biomass in removing copper and lead from human blood plasma in-vivo.

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