Response Surface Methodology for Optimization of Zinc Biosorption By Grewia Orbiculata L.

Rakesh Namdeti*, King Pulipati

Environmental Pollution Control Engineering Laboratory, Department of Chemical Engineering, Andhra University College of Engineering, Visakhapatnam - 530003, Andhrapradesh, INDIA.

*Corresponding Address:

rakesh_nandeti@yahoo.co.in

Research Article

Abstract: Response surface methodology was used to study the effect of various parameters like initial zinc ion concentration, pH, Temperature, biosorbent dosage and to optimize process conditions for the maximum removal of zinc. Batch mode experiments were conducted to determine the isotherms (Langmuir, freundlich, temkin, dubinin radushkevich models), kinetics (Pseudo-first and pseudo second order) and thermodynamic parameters like standard Gibbs free energy (ΔG^0), enthalpy (ΔH^0), entropy (ΔS^0). A 2^4 full factorial central composite design (CCD) using response surface methodology (RSM) was employed for obtaining the mutual interaction between the variables and optimizing these variables. The most influential factor on each experimental design response was identified from the analysis of variance (ANOVA). The model was statistically tested and verified by experimentation. The response surface methodology indicated that 53.07 mg/l initial zinc concentration, 5.86 pH, 0.27 g biosorbent dosage and a temperature of 39.42 ^{0}C were optimal for biosorption of zinc by *Grewia Orbiculata* L., when 75.86 % of the zinc metal is removed from the solution.

Keywords: Biosorption, Grewia Orbiculata L., CCD (Response surface methodology), Isotherms, Kinetics, Thermodynamics.

1. Introduction

Research and development in effluent treatment processes has gained much importance in the recent years as environmental legislations for industrial discharges have become quite stringent. Heavy metal contamination of surface waters due to industrial effluents is a major problem, which needs to be tackled due to ecotoxicological effects of metals along with their accumulation in food chain (EPA, 1976). Zinc is the one of the toxic metals that is available in industrial effluents involved in acid mine drainage, galvanizing plants, natural ores and municipal waste water treatment plants (L Nortan et al., 2004). Conventional technologies like oxidation/reduction, ion exchange, and membrane transfer methods available to tackle the problem of heavy metal contamination of waste waters have a major constraint that they are not so effective and economical for effluents containing concentration of metals less than 100mg/L (B. Volesky, 1990). Hence, there is a need to develop newer techniques, which are cost effective, environment friendly and have high efficacy for metal removal from waste waters with relatively lower metal concentration (S.Tunali et al., 2006). Biosorption is an innovative technology using inactive and dead biomasses to remove heavy metals from aqueous solutions. This biological phenomenon could be explained by considering different kinds of chemical and physical interactions among the functional group present in the cell wall and heavy metals in solution (A.Esposito et al., 2002) . The biosorbent used in this study is Grewia Orbiculata leaves powder, in which the adsorption takes place on surface of insoluble cell walls of the leaves. The insoluble cell walls of the Grewia Orbiculata leaves are largely made up of cellulose and hemi celluloses, lignin, condensed tannins and structural proteins (H.Christ Ray et al., 1981). Batch mode experiments were conducted to determine the biosorption isotherms, kinetics and thermodynamic parameters. The conventional method used for optimization is the “one factor at a time” method in which one independent variable is varied while fixing all others at a specific level. This may lead to unreliable results and less accurate conclusions. This method requires a large number of experiments which might result in predicting the false optimum values. These drawbacks can be eliminated by optimizing all the affecting parameters collectively by Central Composite Design (CCD) (Box GEP et al., 1951) using Response Surface Methodology (RSM). A detailed account of this technique has been outlined (Cochran WG et al., 1957). This optimization process involves three major steps: performing the statistically designed experiments, estimating the coefficients in a mathematical model and predicting the response and checking the adequacy of the model. This technique has been applied in biosorption studies for optimization of different parameters (Farshid G et al., 2008; Preetha B et al., 2007; Kumar R et al., 2009). The present study also applied

CCD to optimize zinc biosorption in an aqueous solution onto *Grewia Orbiculata* L. powder.

2. Material And Methods

2.1 Preparation of biosorbent

The *Grewia Orbiculata* L. leaves were collected near K.L University campus of Guntur, Andhra Pradesh, India. Leaves were washed with deionized water several times to remove dirt particles. Then the dried leaves were powdered using domestic grinder and the powder size of 75-212 µm, which were used as biosorbent without any pretreatment for zinc adsorption.

2.2 Chemical

Analytical grades of $ZnSO_4.7H_2O$, HCl and NaOH were purchased from Merck (Mumbai, Maharastra, India). Zinc ions were prepared by dissolving its corresponding sulphate salt in distilled water. The pH of solutions was adjusted with 0.1 N HCl and NaOH.

2.3 Biosorption experiments

Biosorption experiments were performed at room temperature (30 ± 1^0C) in a rotary shaker at 180 rpm using 250 mL Erlenmeyer flasks containing 30 mL of different zinc concentrations. After 1 hr of contact (according to the preliminary sorption dynamics tests), with 0.1 g *Grewia Orbiculata* L. leaves biomass, equilibrium was reached and the reaction mixture was centrifuged for 5 min. The metal content in the supernatant was determined using Atomic Absorption Spectrophotometer (GBC Avanta Ver 1.32, Australia) after filtering the adsorbent with 0.45 µm filter paper. The amount of metal adsorbed by *Grewia Orbiculata* L. leaves was calculated from the differences between metal quantity added to the biomass and metal content of the supernatant using the following equation:

$$q = \left(C_0 - C_f\right)\frac{V}{M} \qquad (1)$$

Where q is the metal uptake (mg/g); C_0 and C_f the initial and final metal concentrations in the solution (mg/L), respectively; V the solution volume (mL); M is the mass of biosorbent (g).The pH of the solution was adjusted by using 0.1N HCl and 0.1N NaOH.

The Langmuir (I.Langmuir, 1916) sorption model was chosen for the estimation of maximum zinc sorption by the biosorbent. The Langmuir isotherm can be expressed as

$$q = \frac{Q_{\max} b C_{eq}}{1 + bC_{eq}} \qquad (2)$$

Where Q_{max} indicates the monolayer adsorption capacity of adsorbent (mg/g) and the Langmuir constant b (L/mg) is related to the energy of adsorption. For fitting the experimental data, the Langmuir model was linearized as

$$\frac{1}{q} = \frac{1}{Q_{\max}} + \frac{1}{bQ_{\max} C_{eq}} \qquad (3)$$

The freundlich (H.M.F Freundlich, 1906) model is represented by the equation:

$$q = KC_{eq}^{\frac{1}{n}} \qquad (4)$$

Where K (mg/g) is the Freundlich constant related to adsorption capacity of adsorbent and $1/n$ is the Freundlich exponent related to adsorption intensity (dimensionless). For fitting the experimental data, the Freundlich model was linearized as follows:

$$\ln q = \ln K + \frac{1}{n}\ln C_{eq} \qquad (5)$$

The Temkin (C.Aharoni et al., 1977) isotherm has generally been applied in the following form:

$$q = \frac{RT}{b_T}\ln\left(A_T C_{eq}\right) \qquad (6)$$

Where A_T (L/mg) and b_T are Temkin isotherm constants.

The Dubinin–Radushkevich (D–R) (T.Santhi et al., 2010) model was also applied to estimate the porosity apparent free energy and the characteristics of adsorption. The D–R isotherm dose not assumes a homogeneous surface or constant adsorption potential. The D–R model has commonly been applied in the following Eq. (7) and its linear form can be shown in Eq. (8):

$$q_e = Q_m \exp(-K\varepsilon^2) \qquad (7)$$

$$\ln q_e = \ln Q_m - K\varepsilon^2 \qquad (8)$$

where K is a constant related to the adsorption energy, Qm the theoretical saturation capacity, ε the Polanyi potential, calculated from Eq. (9).

$$\varepsilon = RT\,(1+1/C_e) \qquad (9)$$

The slope of the plot of ln qe versus ε^2 gives K (mol^2 $(kJ^2)^{-1}$) and the intercept yields the adsorption capacity, Qm (mg g−1). The mean free energy of adsorption (E), defined as the free energy change when one mole of ion is transferred from infinity in solution to the surface of the solid, was calculated from the K value using the following relation 10:

$$E = 1/\sqrt{2K} \qquad (10)$$

The calculated value of D–R parameters is given in Table 1. The values of E calculated using Eq. (10) is below 1 KJ mol−1, which indicating that the physico-sorption process plays the significant role in the biosorption of zinc metal.

2.4 Biosorption Kinetics

The kinetic studies were carried out by conducting batch biosorption experiments with different initial zinc concentrations. Samples were taken at different time periods and analyzed for their zinc concentration.

2.5 Thermodynamic Studies

The thermodynamic effect was studied by agitating 20 mg l^{-1} of zinc solution with 0.1 g of adsorbent at temperature 30, 35, 40, and 45 ◦C for a time period of 15 min.

2.6 Central composite design (CCD)

With the identification of the parameters having the statistically significant influence on the response a CCD (Box gep et al., 1951) was used to optimize the levels of these parameters. The full CCD, based on three basic principles of an ideal experimental design, primarily consists of (i) a complete 2^n factorial design, where n is the number of test parameters, (ii) n_o center points ($n_o \geq 1$) and (iii) two axial points the axis of each design parameters at a distance of $2^{n/4}$ from the design center. The total number of design points is $N= 2^n+2n+n_o$. For statistical calculations, the parameters X_i are coded as x_i according to Eq. (11):

$X_i = X_i - x_i / \Delta x_j$ (i=1, 2, 3, ….k) (11)

where x_i is dimensional value of an independent parameter, X_i is the real value of an independent parameter, x_i^- is the real value of the independent parameter at the center point and x_j is the step change. The second degree polynomials (Eq. 12) are calculated with the statistical package STATISTICA 6.0 (Stat-Ease Inc., Tulsa, OK, USA) to estimate the response of the dependent variable:

$$Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_4X_4 + b_{11}X_1^2 + b_{22}X_2^2 + b_{33}X_3^2 + b_{44}X_4^2 + b_{12}X_1X_2 + b_{13}X_1X_3 + b_{14}X_1X_4 + b_{23}X_2X_3 + b_{24}X_2X_4 + b_{34}X_3X_4 \quad (12)$$

Where Y is predicted response, X_1, X_2, X_3,X_4 are independent parameter,b_0 is offset term, b_1, b_2, b_3, b_4 are linear effects , b_{11}, b_{22}, b_{33}, b_{44} are squared effects and b_{12}, b_{13}, b_{14}, b_{23}, b_{24}, b_{34} are interaction terms.

3. Results and Discussion

3.1 The effect of contact time

The data obtained from the biosorption of zinc ions on the *Grewia Orbiculata* L. showed that a contact time of 15 min was sufficient to achieve equilibrium and the adsorption did not change significantly with further increase in contact time. Therefore, the uptake and un - adsorbed zinc concentrations at the end of 15 min are given as the equilibrium values (q_e, mg/g; C_{eq}, mg/L), respectively (Fig. 1) and the other adsorption experiments were conducted at this contact time of 15 min (pH 6).

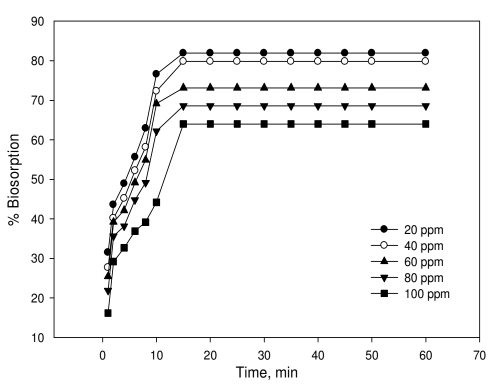

Figure.1.Effect of contact time on biosorption of zinc by *Grewia Orbiculata* L. for various concentrations and 0.1g/30ml of biosorbent concentration

3.2 Effect of pH

The effect of pH on % biosorption was investigated and results were shown in Figure 2. It reveals that % biosorption increased from 15.5 % to 81.9 % as pH is increased from 2 to 6 and then decreased beyond pH value of 6 reaching 76.3 % for pH value of 7. Low pH depresses biosorption of zinc, which may be due to competition with H^+ ions for appropriate sites on the biosorbent surface. However, with increasing pH, this competition weakens and zinc ions replaces H^+ ions bound to the biosorbent or forming part of the surface functional groups such as OH, COOH, etc. (R.Gong et al., 2005; F.A Abu Al-rub et al., 2004; P.X.Sheng et al., 2004; G.Ozdemir et al., 2004; A.Iyer et al., 2004)

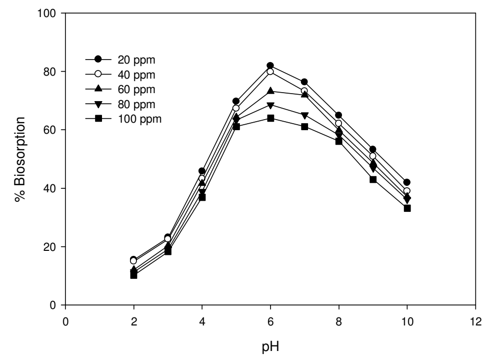

Figure.2. Effect of pH on zinc biosorption by Grewia Orbiculata L. for different concentrations of metal and 0.1g/30ml of biosorbent concentration

3.3 Effect of metal ion concentration

Fig. 3 shows the effect of metal ion concentration on the adsorption of zinc by *Grewia Orbiculata* L. The data shows that the metal uptake increases and the percentage adsorption of zinc decreases with increase in metal ion concentration. This increase is a result of increase in the driving force, i.e. concentration gradient. Though an increase in metal uptake was observed, the decrease in percentage adsorption may be attributed to lack of sufficient surface area to accommodate much more metal available in the solution. The percentage adsorption at higher concentration levels shows a decreasing trend whereas the equilibrium uptake of zinc displays an opposite trend. At lower concentrations, all zinc ions present in solution could interact with the binding sites and thus the percentage adsorption was higher than those at higher zinc ion concentrations. At higher concentrations, lower adsorption yield is due to the saturation of adsorption sites. As a result, the purification yield can be increased by diluting the wastewaters containing high metal ion concentrations.

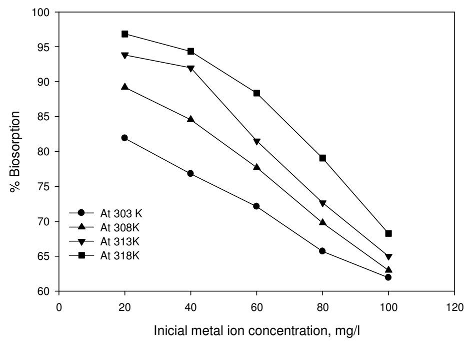

Figure.3. Effect of metal concentration on the biosorption of zinc by Grewia Orbiculata L. at different temperatures & 0.1g/30ml of biosorbent concentration

3.4 Effect of adsorbent dosage

Fig. 4 shows the effect of adsorbent dosage on the % removed at equilibrium conditions. It was observed that the amount of zinc adsorbed varied with varying adsorbent dosage. The amount of zinc adsorbed increases with an increase in adsorbent dosage from 0.1 to 0.5 g. The increase in the adsorption of the amount of solute is obvious due to increasing biomass surface area. Similar trend was also observed for zinc removal using *Azadirachta indica* as adsorbent (K.G.Battacharya et al., 2004).

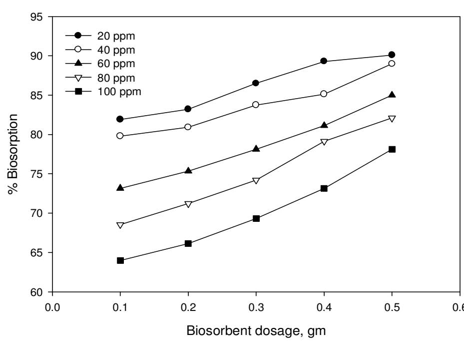

Figure.4. Effect of *Grewia Orbiculata* L. dosage on biosorption of zinc for various concentrations

3.5 Effect of temperature

The effect of temperature on biosorption of zinc was studied between 303 K to 318 K and results were depicted in Figure 5. The results show that biosorption increased from with the rise in temperature.

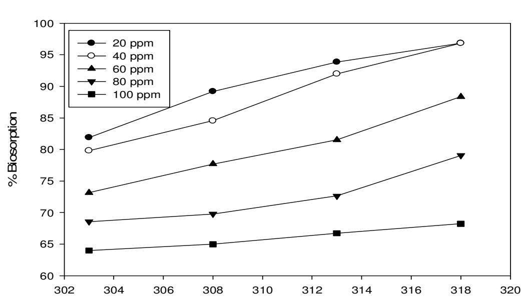

Figure.5. Effect of temperature on biosorption of zinc by *Grewia Orbiculata* L. at different temperatures and 0.1g/ml of biosorbent concentration

4. Biosorption equilibrium

The equilibrium biosorption of zinc on the *Grewia Orbiculata* L. as a function of the initial concentration of zinc is shown in Fig. 6-9. There was a gradual increase of adsorption for zinc ions until equilibrium was attained. The Langmuir, Freundlich, Temkin and Dubinin–Radushkevich models are often used to describe equilibrium sorption isotherms. The calculated results of the Langmuir, Freundlich, Temkin and Dubinin–Radushkevich isotherm constants are given in Table 1.

It is found that the adsorption of zinc on the *Grewia Orbiculata* L. was correlated well with the Langmuir and Temkin as compared to Freundlich, Dubinin–Radushkevich isotherm equations under the concentration range studied. Examination of the Freundlich, Dubinin–Radushkevich isotherm data shows that these isotherms were not modeled as well across the concentration range studied.

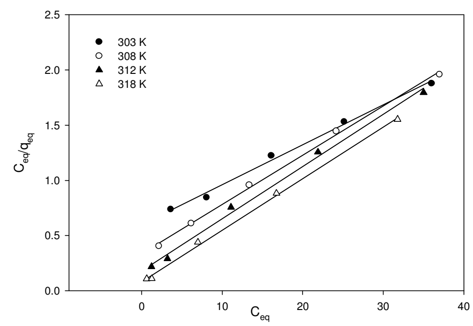

Figure.6. Langmuir biosorption isotherm for zinc at different temperatures and 0.1g/ml of biosorbent concentration

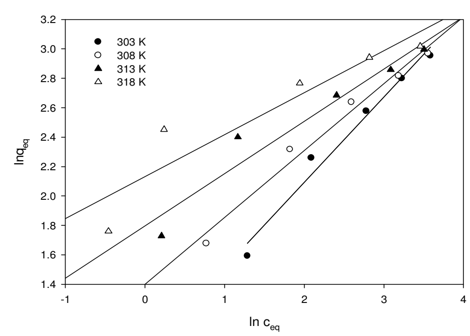

Figure.7. Freundlich biosorption isotherm for zinc at different temperatures and 0.1g/ml of biosorbent concentration.

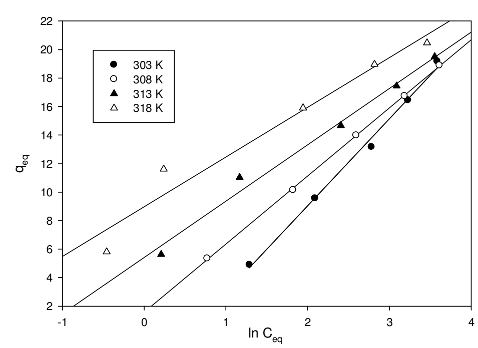

Figure.8. Temkin biosorption isotherm for zinc at different temperatures and 0.1g/ml of biosorbent concentration

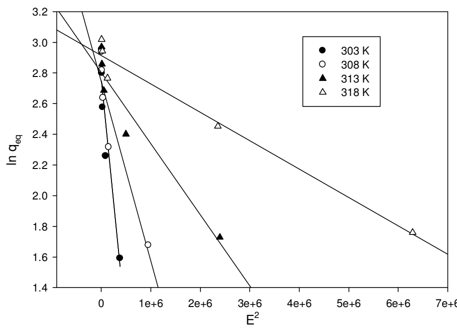

Figure.9. Dubinin- Radushkevich biosorption isotherm for zin at different temperatures and 0.1g/ml of biosorbent concentration

Table.1. The values of parameters and correlation coefficients for each isothermal model for zinc:

Temp(K)	Langmuir			
	q_m,mg/g	b,L/mg	R^2	
303	27.66399	0.060386	0.99434078	
308	23.37796	0.122355	0.99327211	
313	21.70886	0.248543	0.99168221	
318	21.38628	0.573465	0.99831975	
Temp(K)	**Freundlich**			
	n	K_f, mg/g	R^2	
303	1.721026	2.535027	0.9785366	
308	2.198428	4.052025	0.9812287	
313	2.810769	6.026645	0.9397608	
318	3.500519	8.424316	0.8678837	
Temp(K)	**Tempkin**			
	b	A_T,L/g	R^2	
303	412.18	0.59370	0.9945616	
308	514.88	1.29419	0.9954781	
313	636.67	3.63994	0.9858182	
318	758.81	13.1665	0.9568688	
Temp(K)	**Dubinin Radushkevich**			
	K,mol^2/KJ^2	Q_D,mg/g	R^2	E,KJ/mol
303	0.008	15.8239	0.903	0.078
308	0.002	15.7130	0.874	0.130
313	0.004	16.6350	0.919	0.108
318	0.004	18.4443	0.972	0.104

5. Kinetics of adsorption

The prediction of adsorption rate gives important information for designing batch adsorption systems. Information on the kinetics of solute uptake is required for selecting optimum operating conditions for full-scale batch process. The adsorption rate within the first 10 min was observed to be very high and there after the reaction proceeds at a slower rate till equilibrium and finally a steady state was obtained after equilibrium. The saturation time was found to be 15 min based on the initial metal concentration. The kinetics of the adsorption data was analyzed using two kinetic models, pseudo-first- and pseudo-second-order kinetic model. These models correlate solute uptake, which are important in predicting the reactor volume. These models are explained as follows:

5.1 The pseudo-first-order equation

The pseudo-first-order equation of Lagergren (S.Lagergren et al, 1898) is generally expressed as follows:

$$\frac{dq_t}{dt} = k_1(q_e - q_t) \qquad (13)$$

where q_e and q_t are the sorption capacities at equilibrium and at time *t*, respectively (mg/g) and k_1 is the rate constant of pseudo first-order sorption (min−1). After integration and applying boundary conditions, q_t =0 to q_t = q_t at *t* = 0 to *t* = *t*; the integrated form of Eq. (13) becomes:

$$\log(q_e - q_t) = \log(q_e) - \frac{k_1}{2.303}t \qquad (14)$$

The pseudo-first-order rate constant k_1 can be obtained from the slope of plot between log (q_e −*q*) versus time, t. The calculated k_1 values and their corresponding linear regression correlation coefficient values are shown in Table 2. These linear regression correlation coefficient values (R_1^2) shows that this model cannot be applied to predict the adsorption kinetic model.

5.2 The pseudo-second-order equation

If the rate of sorption is a second-order mechanism, the pseudo-second-order chemisorption kinetic rate equation is expressed as (Y.S Ho et al., 1998):

$$\frac{dq_t}{dt} = k(q_e - q_t)^2 \qquad (15)$$

Where q_e and q_t are the sorption capacity at equilibrium and at time *t*, respectively (mg/g) and *k* is the rate constant of pseudo-second-order sorption (g/(mg min)). For the boundary conditions q_t =0 to q_t = q_t at *t* = 0 to *t* = *t*; the integrated form of Eq. (15) becomes:

$$\frac{t}{q_t} = \frac{1}{kq_e^2} + \frac{1}{q_e}t \qquad (16)$$

The second-order rate constants were used to calculate the initial sorption rate, given by the following equation:

$$h = k.q_e^2 \qquad (17)$$

where *t* is the contact time (min), q_e (mg/g) and q_1 (mg/g) are the amount of the solute adsorbed at equilibrium and at any time, *t*. Eq. (16) does not have the problem of assigning as effective q_e. If pseudo-second-order kinetics is applicable, the plot of t/q_t against *t* of Eq. (16) should give a linear relationship, from which q_e and *k can* be determined from the slope and intercept of the plot (Fig. 10) and there is no need to know any parameter beforehand.

The pseudo-second-order rate constant k_2, the calculated q_e value and the corresponding linear regression correlation coefficient value R_2^2 are given in Table 2. At an initial zinc concentrations of 20, 40, 60, 80, 100 mg/L, the linear regression correlation coefficient R_2^2 value was higher. The higher R_2^2 value confirms that the adsorption data were well represented by pseudo-second order kinetic model.

The values of initial sorption (h) that represents the rate of initial adsorption Eq. (17), is increased with increasing metal concentrations onto *Grewia Orbiculata* L. (Table 2).

Table.2. Kinetic constants for zinc onto *Grewia Orbiculata* L. :

Inicial Conc,mg/l	Pseudo second order			
	q_e, mg/g	K_2	R_2^2	h, mg/g.min
20	5.1369	0.1025	0.9982	2.70628
40	10.087	0.0442	0.9977	4.50102
60	13.804	0.0358	0.9979	6.82716
80	17.358	0.0249	0.9975	7.51121
100	20.852	0.0129	0.9939	5.63994
Inicial Conc,mg/l	**Pseudo second order**			
	q_e, mg/g	K_2	R_2^2	h, mg/g.min
20	5.1369	0.1025	0.9982	2.70628
40	10.087	0.0442	0.9977	4.50102
60	13.804	0.0358	0.9979	6.82716
80	17.358	0.0249	0.9975	7.51121
100	20.8528	0.01297	0.99396	5.63994

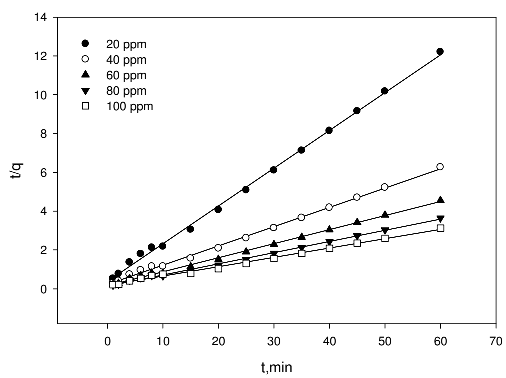

Figure.10. Pseudo second order biosorption of zinc by Grewia Orbiculata L. for different concentrations of metal and 0.1g/ml of biosorbent concentration

6. Thermodynamic Parameters

In environmental engineering practice, both energy and entropy factors must be considered in order to determine what processes will occur spontaneously. Gibb's free energy change, Δ*G*°, is the fundamental criterion of spontaneity. Reactions occur spontaneously at a given temperature if Δ*G*° is a negative value. The thermodynamic parameters of Δ*G*°, enthalpy change, Δ*H*°, and entropy change, Δ*S*°, for the adsorption processes are calculated using the following equations (A.E,Martel et al., 1977; J.M.Murray et al., 1979):

$\Delta G° = -RT \ln K_a$ (18)

and

$\ln K_d = \Delta S^0/R - \Delta H^0/RT$ (19)

Where *R* is universal gas constant (8.314 J/mol K) and *T* is the absolute temperature in K.

A plot ln $K_d(q_e/C_e)$ versus temperature 1/*T*, was found to be linear (Fig. 11). The values of Δ*H*° and Δ*S*° were respectively determined from the slope and intercept of the plot. Δ*H*° and Δ*S*° for the sorption process were calculated to be 101.4683 KJ/mol and 0.337119 KJ/mol K, respectively. The negative values (Table 3) of Δ*G*° confirm the feasibility of the process and the spontaneity of sorption with a high preference for zinc to adsorb onto *Grewia Orbiculata L.* The value of Δ*H*° and Δ*S*° were positive, indicating that the process is spontaneous at high temperatures, where exothermicity plays a small role in the sorption process (Zawani et al., 2009; Jing he et al., 2010).

Table. 3. Thermodynamic Parameters:

Temperature (K)	Thermodynamic Parameters		
	ΔG^0, KJ/mol	ΔH^0, KJ/mol	ΔS^0, KJ/mol
303	-0.76903	101.468	0.337119
308	-2.32269		
313	-3.9542		
318	-5.86523		

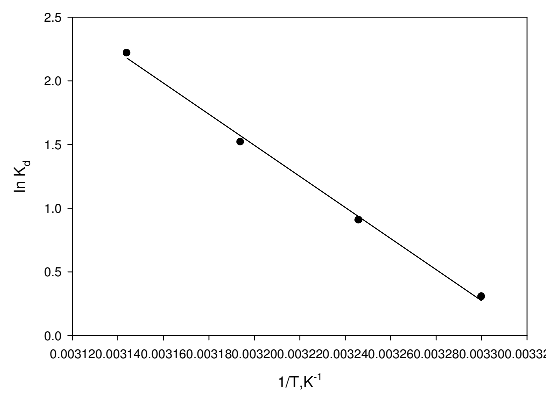

Figure.11. A plot of lnK_d vs.1/T for estimation of thermodynamic parameters

7. Optimization of the Selected Parameters Using CCD

The experiments with different pH values of 2–10, different zinc concentrations of 20–100 mg/L, different biosorbent dosages of 0.1–0.5 g/L and different temperatures of 303–318 K were coupled to each other and varied simultaneously to cover the combination of parameters in the CCD. The levels and ranges of the chosen independent parameters used in the experiments for the removal of zinc were given in Table 4. A 2^4 – factorial CCD design, with eight axial points (α= √4) and six replications at the center points (*n*o=6) leading to a total number of 30 experiments (Table 5) was employed for the optimization of the parameters. The calculated regression equation for the optimization of medium constituents showed that percentage removal of zinc (Y) was function of the temperature (X_1), pH (X_2), biosorbent dosage (X_3) and initial concentration (X_4). Multiple regression analysis of the experimental data resulted in the following equation for the biosorption of zinc:

$$Y = -212.740 + 12.065X_1 + 1.747X_2 + 67.310X_3 + 0.481X_4 - 0.171X_1^2 - 1.954X_2^2 + 34.332X_3^2 - 0.003X_4^2 + 0.574X_1X_2 - 2.085X_1X_3 - 0.005X_1X_4 + 0.949X_2X_3 + 0.007X_2X_4 - 0.118X_3X_4 \quad (20)$$

The coefficients of the regression model were calculated and listed in Table 6. They contain one block term, four linear, four quadratic and six interaction terms. The significance of each coefficient was determined by student's t-test and p-values and listed in Table 6. The larger the magnitude of the t-value and smaller the p-value, the more significant was the corresponding coefficient. This implies that the linear, quadratic and interaction effects of temperature, pH, biosorption dosage and initial concentration of zinc are highly significant as is evident from their respective p-values in (Table 6). The parity plot (Figure 12) showed a satisfactory correlation between the experimental and predicted values of percentage removal of zinc indicating good agreement of model data with the experimental data. The results of the second order response surface model, fitting in the form of ANOVA were shown in Table 7. The Fisher variance ratio, the F-value ($= S_r^2 / S_e^2$), is a statistically valid measure to test the significance and adequancy of the model. The greater the F-value above unity, it is more certain that the factors adequately explain the variation in the data about its mean, and the estimated factor effects are real. The ANOVA of the regression model demonstrated that the model was highly significant, as is evident from the Fisher's F-test (Fmodel = 718.5) and a very low probability value (Pmodel > F=0.000000). More ever, the computed F-value ($F_{0.05(14.15)} = S_r^2/Se^2 = 718.5$) was greater than the tabular F-value ($F_{0.05(14.15)\ tabulars} = 2.46$) at the 1% level, indicating that the treatment differences were significant. The correlation coefficient (R^2) provides a measure of the models variability in the observed response values. The closer the R^2 value to 1, the stronger the model is and it predicts the response better. In this present study, the value of the correlation coefficient (R^2 = 0.9985) indicated that 99.85 % of the variability in the response could be explained by the model. In addition, the value of the adjusted correlation coefficient (Adj R^2 = 0.9971) was also very high to advocate for a high significance of the model. The response surface plots of percentage biosorption of zinc versus the interactive effect of pH, initial zinc concentration, biosorbent dosage and temperature were shown in the Figure 13.

Each response plot represents a number of combinations of two test parameters with the other parameter maintained at zero levels. The maximum percentage biosorption of zinc is indicated by the surface confined in the smallest curve (circular or elliptical) of the response plot. The optimal set of conditions for maximum percentage biosorption of zinc is pH = 5.86, initial concentration of zinc in aqueous solution = 53.07 mg/L, biosorbent dosage = 0.277 g/L and temperature = 39.42^{0}C. The extent of biosorption of zinc at these optimum conditions was 75.865%.

Table.4. Experimental range and levels of the independent parameters:

Independent Parameters	Range and Level				
	-2	**-1**	**0**	**+1**	**+2**
Temperature (X_1), K	30	35	40	45	50
pH (X_2)	4	5	6	7	8
Adsorbent Dosage(X_3), g/L	0.1	0.2	0.3	0.4	0.5
Initial Concentration (X_4), mg/L	20	40	60	80	100

Table 5: CCD matrix showing coded and real values along with the experimental values for percentage biosorption of zinc.

Run no.	Coded values				Real values				% Biosorption of zinc	
	x_1	x_2	x_3	x_4	X_1	X_2	X_3	X_4	Experimental	Predicted
1	-1	-1	-1	-1	35	5	.2	40	69.2	69.5016
2	-1	-1	-1	1	35	5	.2	80	58.38	58.3920
3	-1	-1	1	-1	35	5	.4	40	66.13	66.4404
4	-1	-1	1	1	35	5	.4	80	59.66	59.5283
5	-1	1	-1	-1	35	7	.2	40	62.22	62.4854
6	-1	1	-1	1	35	7	.2	80	60.12	60.1933
7	-1	1	1	-1	35	7	.4	40	59.01	59.0616
8	-1	1	1	1	35	7	.4	80	60.23	60.96708
9	1	-1	-1	-1	45	5	.2	40	65.92	65.58542
10	1	-1	-1	1	45	5	.2	80	60.21	60.40833
11	1	-1	1	-1	45	5	.4	40	63.2	63.37667
12	1	-1	1	1	45	5	.4	80	62.26	62.39708
13	1	1	-1	-1	45	7	.2	40	59.29	59.67167
14	1	1	-1	1	45	7	.2	80	63.22	63.31208
15	1	1	1	-1	45	7	.4	40	56.7	57.100

									1	42
16	1	1	1	1	45	7	.4	80	64.99	64.93833
17	-2	0	0	0	30	6	.3	60	65.98	65.49625
18	2	0	0	0	50	6	.3	60	65.72	65.55125
19	0	-2	0	0	40	4	.3	60	49.12	49.11125
20	0	2	0	0	40	8	.3	60	45.28	44.63625
21	0	0	-2	0	40	6	.1	60	69.98	69.81125
22	0	0	2	0	40	6	.5	60	68.86	68.37625
23	0	0	0	-2	40	6	.3	20	68.93	68.48458
24	0	0	0	2	40	6	.3	100	65.42	65.21291
25	0	0	0	0	40	6	.3	60	75.61	75.61
26	0	0	0	0	40	6	.3	60	75.61	75.61
27	0	0	0	0	40	6	.3	60	75.61	75.61
28	0	0	0	0	40	6	.3	60	75.61	75.61
29	0	0	0	0	40	6	.3	60	75.61	75.61
30	0	0	0	0	40	6	.3	60	75.61	75.61

X_1= Temperature, X_2= pH, X_3= Biosorbent dosage, X_4= Initial Concentration

Table.6. Coefficients, t-statistics and significance probability of the model

Term	Coefficient	Value	Standard error of coefficient	t-value	p-value
Constant	b_0	-2672	9.18784	-29.08	0.0000[a]
X_1	b_1	6.72	0.29259	22.97	0.0000[a]
X_1^2	b_{11}	-0.10	0.00313	-32.23	0.0000[a]
X_2	b_2	76.5	1.32233	57.88	0.0000[a]
X_2^2	b_{22}	-7.18	0.07823	-91.83	0.0000[a]
X_3	b_3	51.0	11.7083	4.361	0.0005[a]
X_3^2	b_{33}	-162.	7.82299	-20.82	0.0000[a]
X_4	b_4	-0.79	0.05854	-13.59	0.0000[a]
X_4^2	b_{44}	-0.00	0.00020	-27.99	0.0000[a]
X_1*X_2	b_{12}	0.05	0.02049	2.690	0.0167[a]
X_1*X_3	b_{13}	0.42	0.20485	2.080	0.0550
X_1*X_4	b_{14}	0.01	0.00102	14.47	0.0000[a]
X_2*X_3	b_{23}	-0.90	1.02427	-0.884	0.3902
X_2*X_4	b_{24}	0.11	0.00512	21.52	0.0000[a]
X_3*X_4	b_{34}	0.52	0.05121	10.24	0.0000[a]

X_1= Temperature, X_2= pH, X_3= Biosorbent dosage, X_4= Initial Concentration.
[a]Significant ($p\leq0.05$)

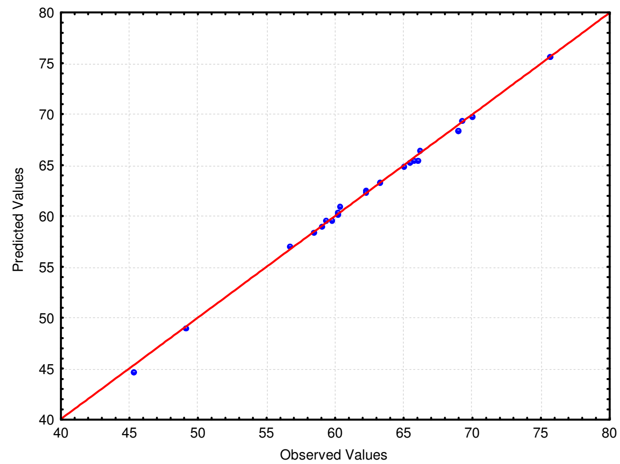

Figure.12. Parity plot showing the distribution of observed vs. predicted values of percentage biosorption of zinc with *Grewia Orbiculata* L.

Table.7. ANOVA for the entire quadratic model.

Source of variation	Sum of squares (SS)	Degrees of freedom (D.F)	Mean squares (MS)	F-value	Probe>F
Model	1689.9	14	120.70	718.5	0.000
Error	2.518	15	0.168		
Total	1692.4	29			

R^2= 0.9985; Adjusted R^2= 0.9971

$F_{0.05(14,15)} = Sr^2 / Se^2 = 718.5 > F_{0.05(14,15)}$Tabular = 2.46

Pmodel > F = 0.000000

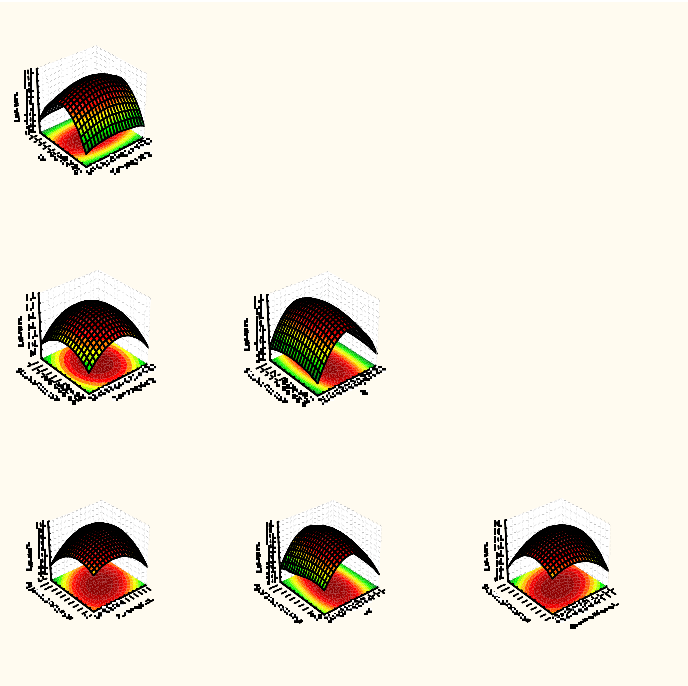

Figure.13. Response surface plots of the effects of different parameters on percentage biosorption of zinc by *Gerwia Orbiculata* L

8. Conclusion

The present study involves the use of statistical experimental design to optimize process conditions for maximal biosorption of zinc from aqueous solution. These parameters were optimized using CCD involving RSM. The significant interactions between the parameters were observed from the response plots. The maximum biosorption of zinc (75.865 %) onto *Grewia Orbiculata*L. powder was observed when the processing parameters were set as follows: pH = 5.86, initial concentration of chromium in aqueous solution = 53.07 mg/L, biosorbent dosage = 0.277g/L and temperature = 39.42^{0}C. This methodology could therefore be effectively used to study the importance of individual, cumulative and interactive effects of the test parameters in biosorption and other processes. The effect of process parameters like pH, metal ion concentration, adsorbent dosage and temperature on process equilibrium was studied. The uptake of zinc ions by *Grewia Orbiculata* L. was increased by increasing the metal ion concentration, adsorbent dosage and decreased by increasing the temperature. The uptake was also increased by increasing pH up to 6. The adsorption isotherms could be well fitted by the Langmuir and Temkin models. The biosorption process could be best described by the Pseudo second-order equation. The negative values of $\Delta G°$ and the positive value of $\Delta H°$ indicate the spontaneous nature of adsorption with a high preference of zinc metal on *Grewia Orbiculata* L., and that the adsorption reaction is endothermic, respectively. The positive value of $\Delta S°$ suggests increasing randomness at the solid/ liquid interface during the adsorption of zinc metal on *Grewia Orbiculata* L. in the aqueous solution.

References

1. A. E MARTELL AND R. M. SMITH, 1977. Critical Stability Constants (Inorganic Chemistry), vol. IV, Plenum, New York, (1977).
2. A. ESPOSITO, F. PAGNANELLI, F. VEGLIO, 2002. pH related equilibria models for biosorption in single metal systems, Chem. Eng. Sci. 57 (2002) 307–313.
3. A. IYER, K. MODY, B. JHA, (2004), Accumulation of hexavalent chromium by an exopolysaccharide producing marine Enterobacter cloaceae, Mar. Pollut. Bull. 49, 974–977, 2004.
4. BOX GEP, WILSON KB (1951) On the experimental attainment of optimum conditions, J R Stat Soc 13: 1-45, 1951.
5. B. VOLESKY, IN: B. VOLESKY (Ed.), 1990. Biosorption of Heavy Metals, CRC Press, Boca Raton, 1990, p. 3.
6. COCHRAN WG, COX GM (1957) Experimental Designs, second ed Wiley New York 346-354, 1957.
7. C. AHARONI, M. UNGARISH, (1977), Kinetics of activated chemisorption Part 2. The oriental models, J. Chem. Soc. Faraday Trans. 73, 456-464, 1977.
8. EPA, 1976. Application of Sewage Sludge to Cropland, Office of Water Prog Operations, US Environmental Agency, Washington, DC, USA, 1976.
9. F.A. ABU AL-RUB, M.H. EL-NAAS, F. BENYAHIA, (2004), I. Ashour, Biosorption of nickel on blank alginate beads, free and immobilized algal cells, Process. Biochem. 39, 1767–1773, 2004.
10. FARSHID G, HABIBOLLAH Y (2008) Application of response surface methodology for optimization of cadmium biosorption in an aqueous solution by Saccharomyces cerevisiae. Chem Eng J 145: 267-275, 2008.
11. G. OZDEMIR, N. CEYHAN, T. OZTURK, F. AKIRMAK, T. COSAR, (2004), Biosorption of chromium(VI), camium(II) and copper(II) by Pantoea sp. TEM 18, Chem. Eng. J. 102, 249–253, 2004.
12. H. CRIST RAY, K. OBERHOLSER, N. SHANK, M. NGUYEN, 1981. Environ. Sci. Technol.15 (1981) 1212.
13. H.M.F. FREUNDLICH, (1906), Over the adsorption in solution, J. Phys. Chem. 57, 385-470, 1906.
14. I. LANGMUIR, (1916), The adsorption gasses on plane surface of glass, mica and platinum, J. Am. Chem. soc. 40, 1361-1368, 1916.
15. J. M. MURRAY AND J. R. DILLARD, 1979. Geochim. Cosmochim. Acta 43 (1979) 781.
16. JING HE, SONG HONG, LIANG ZHAN, FUXING GAN, AND YUH-SHAN HO, 2010. "Equilibrium and thermodynamic parameters of adsorption of methylene blue onto rectorite" vol 19, no.11A, Fresenius Environmental Bulletin, 2010.
17. K.G. BHATTACHARYYA, A. SHARMA, (2004), Adsorption of Pb(II) from aqueous solution by Azadirachta indica (Neem) leaf powder, J. Hazard. Mater. B113, 97–109, 2004.

18. KUMAR R, SINGH R (2009) Response surface methodology approach for optimization of biosorption process for removal of Cr (VI), Ni (II) and Zn (II) ions by immobilized bacterial biomass, sp.Bacillus Brevis. Chem Eng J 146: 401-407, 2009.
19. L. NORTON, K. BASKARAN, S.T.MCKENZIE, 2004. “Biosorption of zinc from aqueous solutions using biosolids”, Adv. Environ. Res., 8, 2004, 629-635.
20. 10. PREETHA B, VIRUTHAGIRI T (2007) Application of response surface methodology for the biosorption of copper using Rhizopus arrhizus. J Hazard Mater 143: 506-510, 2007.
21. P.X. SHENG, Y.P. TING, J.P. CHEN, L. HONG, (2004), Sorption of lead, copper, cadmium, zinc, and nickel by marine algal biomass: characterization of biosorptive capacity and investigation of mechanisms, J. Colloid Interface Sci. 275, 131–141, 2004.
22. R. GONG,Y.D. DING, H. LIU, Q. CHEN, Z. LIU, (2005), Lead biosorption by intact and pretreated Spirulina maxima biomass, Chemosphere 58, 125–130, 2005.
23. S. LAGERGREN, S. KUNGLIGA SVENSKA VENTENSKAPSAKADEMIENS,1898. Handlingar, (1898), Band 24 (4) 1.
24. S.TUNALI, A. CABUK,T. AKAR, 2006. Removal of lead and copper ions from aqueous solutions by bacterial strain isolated from soil, Chem. Eng. J. 115 (2006) 203–211.
25. T.SANTHI, S.MANONMANI, T.SMITHA, 2010. “Kinetics and isotherm studies on cationic dyes adsorption onto annona squmosa seed activated carbon” International journal of engineering science and technology, 2(3), 2010, 287-295.
26. Y.S. HO, E. MCKAY, (1998), “The kinetics of sorption of basic dyes from aqueous solution by Sphagnum moss pea”t, Can. J. Chem. Eng. 76, 822–827, 1998.
27. ZAWANI Z, LUQMAN CHUAH A, THOMAS S Y CHOONG, 2009. “Equilibrium, Kinetics, Thermodynamic Studies: Adsorption of ramazol black 5 on the palm kernel shell activated carbon”, European Journal of scientific research, 37, 1, (2009), pp.63-71