

Research note

Application of Response Surface Methodology for the Optimization of Cu and Zn Removals by Sorption on Pre-treated Oil Palm Frond (OPF)

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Abstract

The increase in the usage of heavy metals in various industrial processes results in increasing heavy metal wastes that need further treatment. Cu and Zn remain hazardous even at low concentration and it is a clear necessity that they be removed from the effluent. The performance of oil palm frond (OPF) pre-treated with NaOH as a sorbent to remove Cu and Zn from aqueous solution was optimized in this study. 2 g of OPF was treated for 300 min in a 250 ml of 1.0 M NaOH solution to improve its sorption ability. Response Surface Methodology (RSM) based on the Three-Variable Composite Face Centered Design was employed as an experimental model to evaluate the effects of initial Zn and Cu concentration (5-100 mg/l), pH of the solution (2-9) and biomass loading (0.5-2.0 g) on the sorption process carried out at 25°C. The solution pH, initial metal concentration and biomass loading were used as the main process variables while the sorption performance was based on the removal efficiency. The coefficient of determination, R^2 , was found to be 0.96 and 0.97 for Cu and Zn removals, respectively. The initial concentration of 89 mg/l, biomass loading of 1.7 g and initial pH of 4.5 were found to be the optimum conditions for the maximum Cu removal of 91.0%, corresponding to sorption capacity of 11.9 mg/g. The optimum conditions for the highest Zn removal of 80.5 % or sorption capacity of 9.0 mg/g were found to be at an initial concentration of 76 mg/l, biomass loading of 1.7 and initial pH of 5.5.

Keywords: Zinc; Copper; Sorption; Response Surface Methodology; Low-Cost Sorbents, Oil Palm Frond

Introduction

Some industrial processes result in the release of heavy metals into natural water systems. This leads to an increasing concern about the effect of toxic metals as environmental contaminants [1]. Due to its special

physical and chemical properties, water is consumed in many industries and has consequently been degraded with different pollutants, depending on the industrial process involved [2]. Copper compounds which are used widely in wire, other

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electrical conductors and coins can create muscle pain, hemolytic and heart failure. Zinc compounds which are used as an alloy, galvanized metal, fluorescence components, paint pigments, sunscreens, fast-setting dental cements, deodorants, embalming and fireproofing lumber can cause mucous membrane damage, diarrhea, and dizziness [3]. The effluent discharge standards for Malaysian inland waters for both Cu and Zn are 1.0 mg/l [4]. Therefore, it seems imperative for Cu and Zn to be removed from the effluent prior to discharge into the aquatic environment.

There are several conventional methods for heavy metal removal from wastewater. The most widely used methods are membrane filtration, chemical precipitation, ion exchange, reverse osmosis and solvent extraction, liquid extraction, electro-dialysis, settling as settleable metal hydroxides, activated carbon adsorption, evaporation, biological methods, electrochemical treatment and the membrane separation process [5-8].

Most of these methods are often ineffective or uneconomical when the heavy metal concentration is in the range of 10-100 mg/l [9]. Therefore, finding a low cost technique for the removal of heavy metals, especially if they be present at low concentrations seems a worthwhile effort [2].

Different low-cost materials are reported to have the capacity for heavy metal sorption. Removal of Cr (III), Cu (II) and Zn with carrot residues [10], Cu (II) removal using wheat husk [7], sorption of Pb on peat [11] and rice husk for uptake of Zn [12] and Pb [5] are some examples of low-cost removals for heavy metals.

Removals up to 90 % have been reported using all those sorbent materials. The abundant supply of biomasses from the Malaysian oil palm industry justifies the use of those materials for this application.

Response surface methodology (RSM) is a mathematical/statistical based technique

which is useful for analyzing the effects of several independent variables on the response, [13]. In most RSM applications, the relationship between the response and the independent variables is unknown. Therefore, the first step in RSM is to approximate the function (f). Usually, this process employs a low-order polynomial in some region of the independent variables. If the response is well-modeled by a linear function of the independent variables, then the approximating function is a first-order model.

If there is curvature in the system or in the region of the optimum, then a polynomial of higher degree must be used to approximate the response. This is to analyze and locate the optimum, i.e. the set of independent variables wherein the partial derivatives of the model respond with respect to the individual independent variables that equal to zero. The eventual objective of RSM is to determine the optimum operating conditions for the system, or to determine the region, which satisfies the operating specifications. Almost all RSM problems utilize one or both of these approximating polynomials [14-16].

RSM has an important application in the process design and optimization as well as the improvement of existing design. This methodology is more practical compared to the approaches mentioned above as it arises from experimental methodology which includes interactive effects among the variables and, eventually, it depicts the overall effects of the parameters on the process [17].

The design of the adsorption column is often carried out based on the kinetic and mass transfer coefficients determined by isotherm models or the general practice of determining the optimal operating conditions while keeping the others at a constant level or one-variable-at-a-time technique. Therefore, the steady state models are basically able to predict the parameters which have been considered in mass balance relations but are unable to estimate other interrelated effluent quality parameters (responses) [18].

The present study focuses on the optimization of Cu and Zn from aqueous solution using oil palm frond (OPF) pre-treated with NaOH. The alkaline treatment was found to be the most effective procedure for improving the sorption capacity among all the methods chosen for treating the oil palm derived biomasses. Oil palm frond as a new low-cost sorbent material has been optimized for the best sorption capacity in this study. The optimization of initial concentration, biomass loading and pH were deliberated via the Central Composite Face-Centered RSM experimental design. Optimization was carried on in order to define the most suitable conditions for the sorption process. The interactions between factors influencing the sorption process of Cu and Zn were identified, modeled and a description given of the behavior of the factors on the removal of the heavy metals was established.

Material and Methods

Stock Solutions

Stock solution for Zn was prepared by dissolving zinc nitrate hexahydrate ($\text{Zn}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, BDH) into distilled water while for Cu, copper nitrate trihydrate ($\text{Cu}(\text{NO}_3)_2 \cdot 3\text{H}_2\text{O}$, Merck) was used [19]. The initial solution was made at a concentration of 100 mg/l and diluted to the desired concentrations using distilled water if required. The nitrate salts were of analytical grade.

Biomass Preparation

Oil palm frond (OPF) was obtained from the palm oil trees in Universiti Sains Malaysia. The biomass was ground with a blender (model Epicson Eb-321) for 1 min, washed thoroughly with distilled water and then, dried in an oven at 70°C for 12 h to reach a constant weight [1]. For the preparation of alkaline treated sorbent material, 2 g of biomass was treated for 45 min with a 1.0 M solution of NaOH at a room temperature of

25°C and then thoroughly washed with distilled water until a constant pH of the washing water was reached. It was then dried for 12 h at 70°C in an oven until constant weight was obtained.

Measurements

The Zn concentration was measured with a HANNA zinc meter (model HI93731), while that of Cu was measured using a HANNA copper meter (model HI93702). The solution pH was measured using a WTW pH meter (model pH330i 2A2O-1012).

Batch Sorption Studies

In a typical experiment, the mixture for sorption study consisted of 0.5-2.0 g of OPF biomass in 250 ml of known concentration of heavy metal solution. The mixture was agitated at 150 rpm for 10 min under a normal room temperature of 25°C. It was then filtered through Whatman filter paper (110mm cat no. 1001 110), and the filtrate obtained was analyzed for heavy metal concentration.

The effect of pH was investigated by varying the pH of the solution in the range of 3-6 by the addition of 0.1 M of HCl or NaOH. The study on the effect of the initial concentration of Cu and Zn on the removal efficiency was carried out with initial concentrations in the range of 5-100 mg/l. The effect of biomass loading on the sorption process was also performed with variable biomass loadings of 0.5-2.0 g.

The removal efficiency of Cu and Zn from the solution was calculated using the following relationship:

$$\text{Removal}\% = \frac{C_i - C_f}{C_i} \times 100 \quad (1)$$

Where C_i is the initial concentration of Zn and Cu in the lead solution (mg/l) and C_f is the final concentration of Zn and Cu in the

solution (mg/l). The pH after contact time was also considered as a response to the heavy metal removals.

Experimental Design for Optimization

The optimization of Cu and Zn removals was carried out by three chosen independent process variables using 2^3 factorial experiments design with six star points ($\alpha=1$) and five replicates at center points, according to the Central Composite Face-Centered Design (CCFD). The ranges and the designed levels of the variables investigated in this study are given in Table 1. The amount of metal removal and pH after contact time were taken as the responses of the design experiments. The quadratic equation model for predicting the optimal point was expressed by Equation. 2:

$$Y = \beta_0 + \sum_{i=1}^k \beta_i x_i + \sum_{i=1}^k \beta_{ii} x_i^2 + \sum_{i=1}^k \sum_{j=i+1}^k \beta_{ij} x_i x_j + \varepsilon \quad (2)$$

where:

Y is the response (dependent variables),

β_0 is constant coefficient,
 β_i, β_{ii} and β_{ij} are coefficient for the linear, quadratic and interaction effect,
 x_i and x_j are factors (independent variables) while
 ε is the error

The Design Expert (Version 6.0.7, StatEase, Inc., USA) was used for regression and graphical analyses of the data obtained. The variability in dependent variables was explained by the multiple coefficients of determination, R^2 , while the model equation was used to predict the optimum values [14].

Result and Discussion

Experimental Design and Analysis of Variance (ANOVA)

The results at each point based on experimental design for Cu and Zn are shown in Table 2 and Table 3, respectively. A quadratic regression model was made by using coded values from the estimation of data. The model for removal of Cu is given by Equation 3 and the model for predicting the pH after contact time is demonstrated in Equation 4.

Table 1. Coded and actual values of variables of the experimental design

Factor	Coded levels of variables			
	-1.00	0	1.00	
pH	A	4.5	6	3
Initial concentration (mg/l)	B	52.5	100	5
Biomass loading (g)	C	1.25	2	0.5

Table 2. Experimental design and results for Cu removal and pH after contact time

Run	Coded values			Cu Removal, %		pH after Contact time	
	A	B	C	Observed	Predicted	Observed	Predicted
1	-1	-1	-1	0.00	3.30	3.21	3.13
2	1	1	1	87.60	91.95	6.52	6.74
3	0	0	0	66.48	68.42	7.13	7.16
4	0	1	0	77.50	74.44	6.02	6.26
5	1	-1	1	22.00	23.43	10.58	10.95
6	1	-1	-1	4.00	3.30	10.19	9.34
7	0	0	-1	53.52	51.29	6.14	6.35
8	0	0	0	72.95	68.42	6.50	7.16
9	0	0	1	72.29	85.54	9.73	7.96
10	-1	0	0	42.29	46.02	4.82	4.26
11	-1	1	1	77.70	71.00	5.35	5.38
12	0	0	0	73.71	68.42	6.85	7.16
13	0	0	0	74.86	68.42	7.50	7.16
14	-1	1	-1	16.40	22.62	3.42	3.77
15	1	0	0	60.76	56.50	7.28	8.04
16	-1	-1	1	30.00	23.43	4.49	4.75
17	1	1	-1	44.40	43.57	5.63	5.13
18	0	-1	0	28.00	30.52	7.42	8.05
19	0	0	0	64.57	68.42	7.12	7.16

Table 3. Experimental design and results for Zn removal and pH after contact time

Run	Coded values			Zn Removal, %		pH after contact time	
	A	B	C	Observed	Predicted	Observed	Predicted
1	0	0	0	60.00	64.26	8.01	7.96
2	1	-1	1	60.00	63.08	10.60	10.82
3	0	1	0	65.00	64.16	6.60	6.99
4	-1	-1	1	50.00	50.82	6.45	6.20
5	0	0	0	67.62	64.26	8.13	7.96
6	0	0	1	69.07	69.46	9.96	8.96
7	0	-1	0	56.00	51.76	8.48	8.94
8	0	0	0	63.81	64.26	8.05	7.96
9	0	0	0	61.90	64.26	7.21	7.96
10	-1	1	1	64.00	63.22	5.45	5.67
11	0	0	-1	37.14	31.67	7.04	6.97
12	-1	0	0	56.19	54.26	4.03	4.57
13	1	1	1	79.00	75.48	7.30	7.45
14	-1	-1	-1	2.00	5.29	3.50	3.47
15	1	-1	-1	36.00	33.05	9.67	9.57
16	1	1	-1	40.00	45.45	6.20	6.20
17	0	0	0	60.00	64.26	8.16	7.96
18	-1	1	-1	18.00	17.69	3.40	2.93
19	1	0	0	75.24	74.27	8.79	8.51

$$\text{Removal (\%)} = +68.42 + 5.24A + 21.96B + 17.13C - 17.16 A^2 - 15.93 B^2 + 5.24AB + 7.06BC \quad (3)$$

$$\text{pH after contact time} = +7.16 + 1.89A - 0.90B + 0.81C - 1.01A^2 - 1.21AB \quad (4)$$

For Zn, the models for removal and pH after contact time as responses of the design by using coded values are given in Equation 5 and Equation 6, respectively.

$$\text{Removal (\%)} = +64.26 + 10.00A + 6.20B + 18.89C - 6.30B^2 - 13.70C^2 - 3.87AC \quad (5)$$

$$\text{pH after contact time} = +7.96 + 1.97A - 0.98B + 0.99C - 1.42A^2 - 0.71AB - 0.37AC \quad (6)$$

For Zn removal, the highly significant result at 99% of confidence level for both Zn removal and pH after contact time was obtained. The coefficients of determination ($R^2=0.9737$ and $R^2= 0.9632$ for Zn removal and pH after contact time, respectively) were also reasonably good.

The significance of each coefficient in the Equations 3 to 6 was determined by Student *t*-test and *p*-values [14].

The ANOVA for Cu removal and the predicted pH after the desired contact time model used to estimate the responses as a function of pH, initial concentration and biomass loading is shown in Table 4 and the ANOVA for Zn removal and pH after the

desired contact time model for estimating the responses as the function of the same factors are presented in Table 5.

The regression of 99% was quite significant as a confidence level for both Cu removal and the pH after contact time. The coefficient of determination ($R^2 = 0.9625$ for Cu removal and $R^2 = 0.9111$ for pH after contact time) were reasonably good which suggested 96.3% for removal and 91.1% for pH after contact times were explained by the variation in the variables. The remaining (5.75 % and 8.9 % for removal and final pH, respectively) were therefore, explicated by the residues.

Table 4. Analysis of variance for the quadratic model for Cu removal and pH after contact time

	Source	Sum of Squares	DF ^a	Mean Square	F Value	Prob. > F
Removal, %	Model	12740.90	7	1820.13	40.29	< 0.0001
	Residual	496.88	11	45.173		
	Lack of Fit	410.23	7	58.603	2.70	0.1765
	Pure Error	86.64	4	21.663		
	Total	13237.77	18			
$R^2 = 0.9625$, $CV^b = 13.18\%$						
pH after contact time	Model	66.84	5	13.37	26.66	< 0.0001
	Residual	6.52	13	0.50		
	Lack of Fit	5.97	9	0.66	4.81	0.0725
	Pure Error	0.55	4	0.14		
	Total	73.36	18			

$R^2=0.9111$, $CV= 10.69\%$

^a DF= Degree of freedom

^b CV= Coefficient of variation

The small value of “Prob.>F” (less than 0.05) indicated the significance of the model terms. The non-significant value of lack of fit for all

the models (more than 0.05) clearly indicated that the quadratic models were valid for the present study [1].

Table 5. Analysis of variance for the quadratic model for Zn removal and pH after contact time

	Source	Sum of Squares	DF	Mean Square	F Value	Prob > F
Removal, %	Model	6624.83	6	1104.14	73.90	< 0.0001
	Residual	179.28	12	14.94		
	Lack of Fit	138.63	8	17.33	1.71	0.3182
	Pure Error	40.65	4	10.16		
	Total	6804.12	18			
R ² = 0.9737, CV= 13.18%						
pH after contact time	Model	73.02	6	12.17	52.40	< 0.0001
	Residual	2.79	12	0.23		
	Lack of Fit	2.16	8	0.27	1.71	0.3171
	Pure Error	0.63	4	0.16		
	Total	75.81	18			
R ² = 0.9632, CV= 13.18%						

Overall Comparison Based on the Model Terms

In the model for removal of Cu (Equation 3) a modified quadratic model was established. In this model, the two-level interaction between terms A (pH) and C (biomass loading) and the second order effect of term C were insignificant, while the other terms were significantly introduced. This shows that biomass loading had the least effect on the sorption model of Cu while the initial concentration played the most important role in this process.

The model derived for Zn indicated insignificant values for the two-level interaction between term B (initial concentration) and term C (biomass loading), two-level interaction between terms A and B and second order for term A, while the other model terms are significant.

A comparison between the sorption model of Cu and Zn indicated that as the insignificance of second order models in Zn sorption process are of higher extent than that of Cu sorption process. Thus, it is predicted that the curvature of the trend with the Cu model

should be more noticeable compared to the Zn adsorption model.

By evaluating the models for predicting the pH after the contact time, it was estimated that term A was the most important variable in affecting the value of pH. In both models for Cu and Zn, the two-level interaction between terms A and B is considered significant. For Zn sorption, the two-level interaction between term A and term C show significant value, while in the Cu sorption model, this interaction seems to be insignificant.

Graph-based Overall Discussion

Figure 1 shows the response surface 3D plot for the effect of interaction between pH and the initial concentration on Cu removal while the biomass loading term was at 1.25g. It is noted in this figure that the removal increased via the increase in the initial heavy metal concentration. The removal would initially increase and then decrease at initial concentrations near to 100 mg/l. The increasing pH would affect the removal up to

a pH of around 4.5, which was the optimum predicted. Further increase in the pH would be detrimental to the removal efficiency.

By analysing Figure 2 which is the corresponding plot for Zn, it is obvious that the effect of initial concentration and pH on Zn removal was similar to that for Cu removal. However, the maximum point for pH occurred at around 5.5 and the initial concentration curve reached its maximum point at around 77 mg/l.

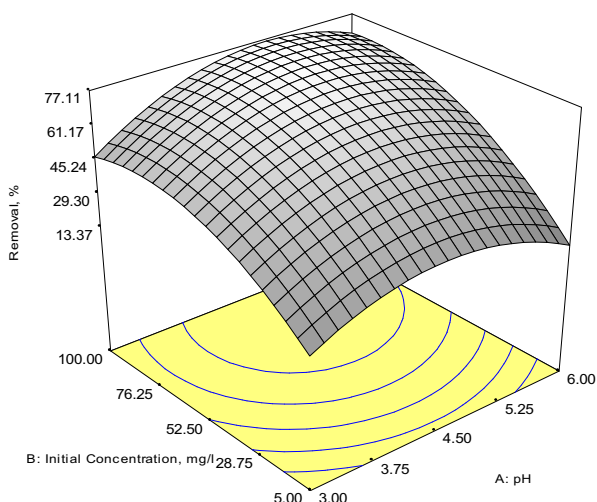


Figure 1. Response surface 3D plot indicating the effect of interaction between initial concentration and pH on Cu removal while holding the biomass loading at 1.25g

The maximum predicted removal was obtained at an initial concentration of 89 mg/l and pH 4.5 to achieve a Cu removal of 89.4%. Also the maximum removal for Zn of 77.3% was predicted at 76 mg/l and pH 5.5. Therefore, a combination of high initial concentration and moderate pH of the solution resulted in the maximum uptake of the metals. To avoid colour change in the solution that would complicate the analysis, all the experiments were conducted at pH values less than 6.0.

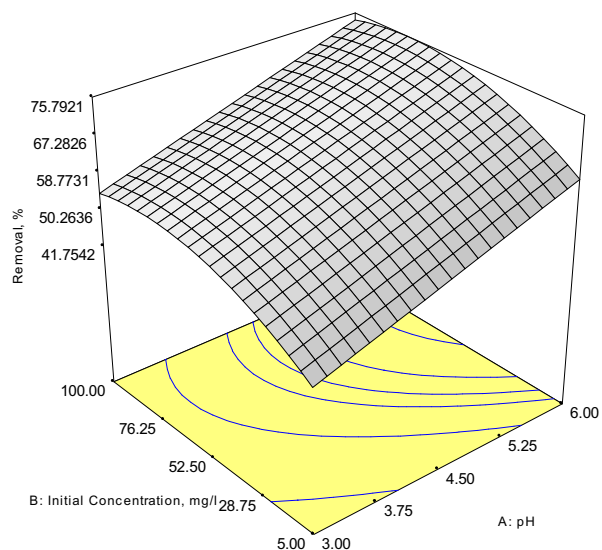


Figure 2. Response surface 3D plot indicating the effect of interaction between initial concentration and pH on Zn removal while holding the biomass loading 1.25g

It was also noted that the effect of interaction between initial concentration and biomass loading was significant as demonstrated in the 3D plot in Figure 3 and Figure 4 for Cu and Zn removal, respectively.

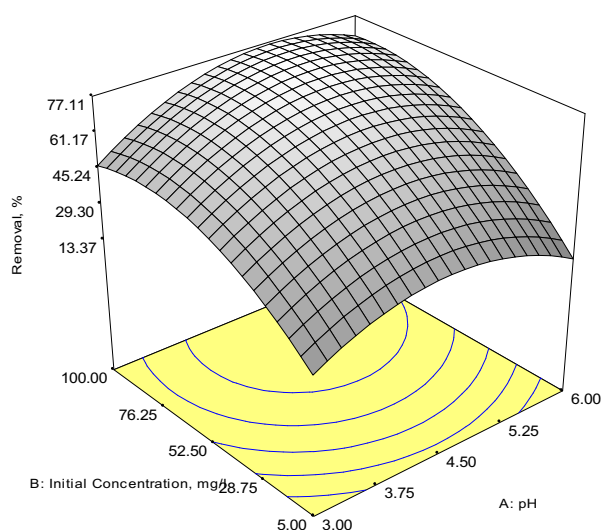


Figure 3. Response surface 3D plot indicating the effect of interaction between initial concentration and biomass loading on Cu removal while holding the pH of 4.5

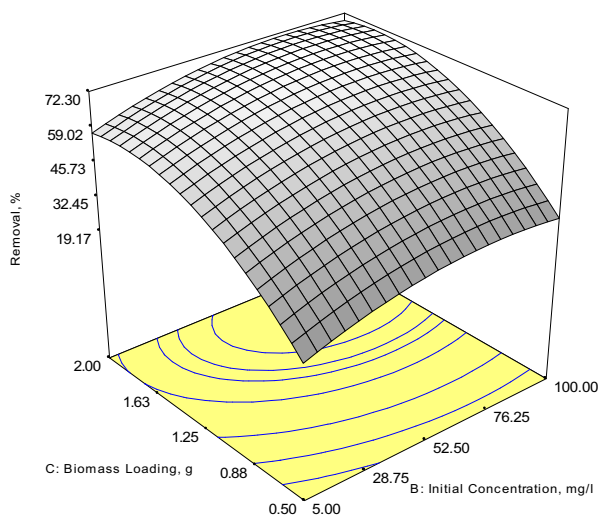


Figure 4. Response surface 3D plot indicating the effect of interaction between initial concentration and biomass loading on Zn removal while holding the pH of 4.5

The optimum point for the biomass loading for both Cu and Zn removals were determined to be with 1.7g of pre-treated biomass. The interaction between pH and biomass loading while the initial concentration was set at 52.5 mg/l as the center point of the design are demonstrated in Figure 5 and Figure 6 for Cu and Zn, respectively. Relatively higher initial concentration and very low biomass loading favor Cu and Zn removal. This can be explained in terms of the more Cu^{2+} and Zn^{2+} ions existing at higher concentrations of the respective heavy metal solutions.

Figure 5 and 6 suggest that the amount of removal is directly related to the amount of biomass and the pH. The increase in biomass loading would favor the removal efficiency. This was attributed to the greater number of metal binding sites on the biomass which resulted in higher heavy metal sorption. It was anticipated that high biomass loading could create a 'screen effect' of the dense outer layer of cells, protecting the binding sites from metal [20]. This effect can be observed in Figure 6 which shows a reduction in removal after a certain amount of biomass loading.

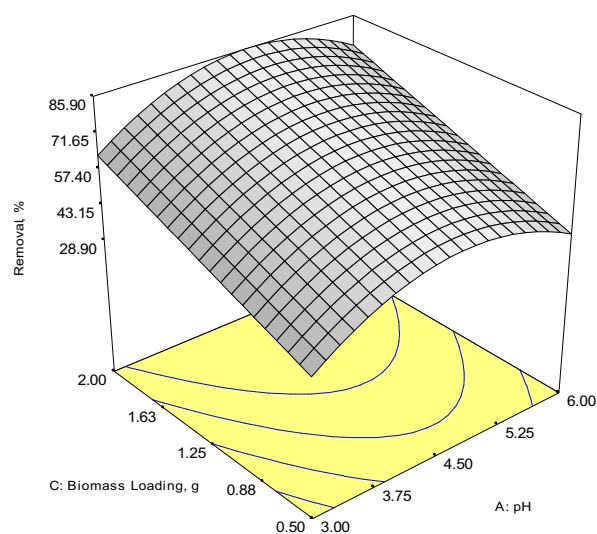


Figure 5. Response surface 3D plot indicating the effect of interaction between pH and biomass loading on Cu removal while holding the initial concentration at its respective level of 52.5 mg/l

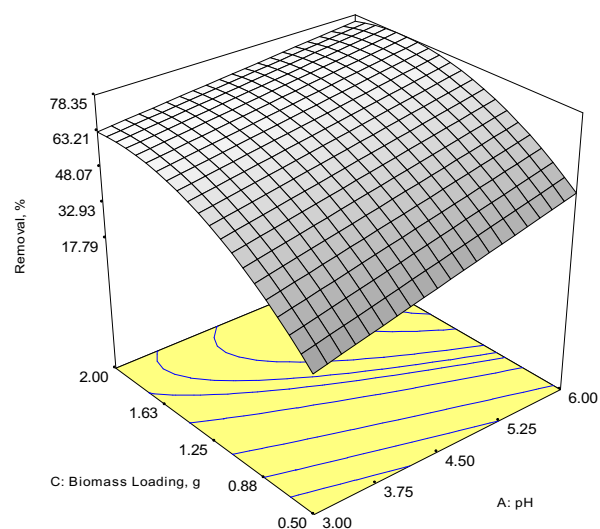


Figure 6. Response surface 3D plot indicating the effect of interaction between pH and biomass loading on Zn removal while holding the initial concentration at its respective level of 52.5 mg/l

A set of solutions that were given by the software in order to determine the optimum conditions of the Cu and Zn removal process

are summarized in Table 6. These solutions were varied by performing the sorption process under the predicted conditions. Maximum removal of 91.0 % for Cu, corresponding to 11.9 mg/g of sorption capacity was achieved at pH 4.5 when an initial concentration of 89 mg/l when 1.7 g of biomass was used. The solution given for Zn removal led to a removal of 80.5 %, corresponding to 9.0 mg/g of metal sorption capacity when 76 mg/l of initial concentration with 1.7 g of biomass loading at pH 5.5 were used.

The sorption uptake values obtained in the optimized process were significantly higher than reported values in the literature. According to Basci et al. [7], wheat husk had

the sorption capacity of 6.82 mg Cu(II)/ g at pH 5 with the initial concentration of 100 mg/l and a corresponding removal efficiency of 82%. Alhakawati and Banks [21] reported that immobilised *Ascophyllum nodosum* in hydrophilic polyurethane foam could reach the sorption capacity of 2.35 mg Cu (II) / g at pH value of 5 with 2 mg/l of initial concentration. Meanwhile, Zn has been reported to be removed from wastewater with an efficiency rate of 75% when the initial concentration was 20 mg/l and the optimum pH of 5 using carrot residues as the sorbent material. The amount of biomass in use has been reported to be 10 g/l [10].

Table 6. Optimum conditions found by the Design Expert software for the Cu and Zn sorption processes

Metal	Run	A	B	C	Removal, %			S.D.*
					Obtained	Predicted	Error	
Cu	1	4.5	89	1.7	91.0	89.75	1.25	± 0.88
	2	4.5	97	1.8	90.5	94.94	-4.44	± 3.14
	3	4.5	85	1.5	81.0	83.65	-2.65	± 1.87
	4	4.5	97	1.7	90.0	88.54	1.46	± 1.03
Zn	1	5.5	76	1.7	80.5	77.31	3.2	± 2.26

* S.D. = Standard Deviation

Conclusion

The optimization of the removal of Cu and Zn from aqueous solution by NaOH pre-treated OPF was conducted in batch experiments. The response surface methodology was employed for optimization and analysis of the adsorption process. The existence of interactions among the factors and the results were deliberated. The optimum conditions for Cu removal was observed to be at an initial concentration of

89 mg/l with 1.7 g of biomass loading at pH 4.5 which led to a removal of 91.0 % or a corresponding sorption capacity of 1.9 mg/g. Optimum conditions for the Zn sorption process were found to be at an initial concentration of 76 mg/l, a biomass loading of 1.7 g and a pH of 5.5. The removal efficiency for Zn was found to be at 80.5 %, corresponding to a sorption capacity of 9.0 mg/g at the optimum conditions.

References

1. Zulkali, M.M.D., Ahmad, A.L. and Norulakmal, N.H. "Oryza sativa L. husk as heavy metal adsorbent: Optimization with lead as model solution," *Bioresource Technology*, **97**, 21-25 (2006).
2. Salamatinia, B., Kamaruddin, A.H. and Abdullah, A.Z., "A review on low-cost biosorbents for heavy metal removal from waste water", *Proceedings of the AEESEAP Conference, Kuala Lumpur*, (2005).
3. Brady, J. E., *General Chemistry, Principles & Structure* 5th edition", John Wiley & Sons, New York, (1990).
4. Environmental Quality Act and Regulations Malaysia, "Sewage and Industrial Effluents, 1979". Department of Environment Malaysia, (2001).
5. Zulkali, M.M.D., and Norulakmal, N.H., "Optimization of lead removal using rice husk by response surface methodology". *Proceedings of 17th SOMCHE, Penang*, 641-647 (2003).
6. Jang, A., Seo, Y. and Bishop, P.L., "The removal of heavy metals in urban runoff by sorption on mulch", *Environmental Pollution*, **133**, 117-127 (2004).
7. Basci, N., Kocadagistan, E. and Kocadagistan, B., "Biosorption of copper (II) from aqueous solutions by wheat shell." *Desalination*, **164**, 135-140 (2004).
8. Tarley, C.R.T. and Arruda, M.A.Z. "Biosorption of heavy metals using rice milling by-products. Characterization and application for removal of metals from aqueous effluents", *Chemosphere*, **54**, 987-995 (2004).
9. Holan, Z.R., Volesky, B., "Biosorption of Pb and Ni by biomass of marine algae", *Biotechnology and Bioengineering*, **43**, 1001-1009 (1994).
10. Nasernejad, B., Esslamzadeh, T., Bonakdarpour, B., Esmaailbygi M. and Zamani, A., "Camparison for biosorption modeling of heavy metals (Cr (III), Cu (II), Zn (II)) adsorption from wastewater by carrot residues", *Process Biochemistry*, **40**, 1319-1322 (2005).
11. Ho, Y. S. and Mckay, G., "The sorption of lead (II) ions on peat", *Water Research*, **33**, 578-584 (1999).
12. Mishra, S. P., Tiwari, D. and Dubey, R. S., "The uptake behavior of rice (jaya) husk in the removal of Zn (II) ions: A radiotracer study", *Appl. Radiat. Isot*, **48**, 877-882 (1997).
13. Box, G.E.P. and Draper, N.R., *Empirical Model-Building and Response Surfaces*. John Wiley & Sons, New York, (1987).
14. Montgomery, D.C., *Design and Analysis of Experiments*, 5th ed." John Wiley & Sons, New York, (2001).
15. Mason, R. I. Gunst, R. F. and Hess, J. L., *Statistical Design and Analysis of Experiments. Eighth Applications to Engineering and Science*, 2nd edition", John Wiley & Sons, New York, (2003).
16. Khuri, A. I. and Cornell, J. A., *Response Surfaces: Design and Analyses*; 2nd edition, Marcel Dekker, New York, (1996).
17. Bař, D. and Boyaci, İ. H., "Modeling and optimization I: usability of response surface methodology". *Journal of Food Engineering*, article in press, (2006).
18. Sötemann, S. W., Ristow, N. E., Wentzel M.C., Ekama, G. A., "A steady state model for anaerobic digestion of sewage sludge". *Water SA*, **31**, 511-527 (2005).

19. Kenkel, J., *Analytical Chemistry for Technicians*, 3rd edition”, CRC Press, London, (2003).
20. Sekhar, K.C., Subramaniam, S., Modak, J.M. and Natarajan, K.A., “Removal of metal ions using an industrial biomass with reference to environmental control”. *International Journal of Mineral Processing*, **53**, 107–120 (1998).
21. Alhakawati, M.S. and Banks, C.J., “Removal of copper from aqueous solution by *Ascophyllum nodosum* immobilized in hydrophilic polyurethane foam”. *Journal of Environmental Management*, **72**, 195-204 (2004).