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Response Surface Methodology Approach for Analysis of Phytoremediation Process of Pb(II) from Aqueous Solution Using *Echinodorus palaefolius*

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Abstract. The purpose of this study was to observe the phytoremediation process for removal of Pb(II) from aqueous solution using *Echinodorus palaefolius* (Jasmine water). The process was determined by the root, stem and leaves ability to reduce the Pb(II). The experiment was done in three factors, as a function of weight of plant (2–10 g/ 100 mL), initial concentration of Pb(II) (10–30 mg/l) and contact time (30–120 minutes) into the final concentration of Pb(II) removal as the response. The effect of three factors in Pb(II) phytoremediation was explained using response surface methodology (RSM). The results revealed that the phytoremediation process can successfully remediate Pb(II) from aqueous solution with the Pb(II) adsorbed ranging between 0.231 mg/L – 2.431 mg/L. The highest phytoadsorption of Pb(II) using *Echinodorus palaeofolius* occurred in roots. Optimizing the condition using 2FI model predicts weight of plant 2 gr, initial concentration of Pb(II) 30 mg/L and contact time 120 minutes. The initial concentration and contact time are significant factors influencing the phytoremediation process.
Keywords: *Echinodorus palaefolius*, Phytoremediation, Response Surface Methodology, Pb(II).

1. Introduction

Heavy metals contained in wastewater deteriorate water quality. Regarding their toxicities, the most problematic heavy metals are Hg, Cd, Pb, As, Cu, Zn, Sn, and Cr [2, 3, 4, 5, and 11]. The mobilization of heavy metals through industrialization and disturbance of natural biogeochemical cycles release these elements into the environment. Lead (Pb(II)) for instance, its excessive concentration may lead to a serious environmental and health problem. The Pb(II) concentration beyond threshold limit has adverse effects to ecosystem balance, aquatic biota and human health. In human health problem, they interfere with the normal functioning of living systems such as neurological system, reproduction system, thought and behaviour changes, high blood pressure and anemia.

Since 1990, phytoremediation technology has been emerged as a recent technology to remediate heavy metal pollution. The green plants has potentially remove significant amount of heavy metal pollutant [3, 8, 9, 12]. Some of them are water hyacinth (*Eichhornia crassipes*), water caltrop (*Trapa natans*), water lettuce (*Pistia stratiotes*) and jasmine water (*Echinodorus palaefolius*). These plants can be sites for adsorbing and bioremediation the heavy metal pollutants from wastewater. The

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process of bioremediation will be discontinued as the amount of toxicant sorbet species is equal to its portion remaining in the solution [2, 8, 9]. Phytoremediation is low cost, effective method and environmental friendly compared to other remediation options [1, 2, 3]

Regarding previous studies, *Echinodorus palaefolius* can remove organic pollutant from grey water and tofu wastewater [6, 7]. The results show some physical variables influencing the phytoremediation process include pH, temperature, and contact time [3, 6, 7]. The current study focus on the capability of *Echinodorus palaefolius* in reducing Pb(II). To enhance the studies, the statistical experiment design is needed. It is also used to analyze and to optimize experiments are termed response surface designs. The effects of factors (dependent variables) including weight of plant, initial concentration of Pb(II) and contact time as well as its interaction were investigated.

2. Material and Method

2.1. Preparation of Aqueous Solution

The aqueous solution of Pb (II) was prepared by diluting the stock solutions of Pb(II) of 1000 mg/L concentration $PbSO_4$ (pure analytical grade) into appropriate concentration (10 mg/L, 20 mg/L and 30 mg/L).

2.2. Preparation of Plants

Echinodorus palaefolius (Jasmine water) used to as test plant for this experiment belongs to the family Alismatacea. For this experiment, these plants were collected from nearby wetlands located in Jambi city. These plants were grown up in some pots to keep their freshly. Then, the plant section of roots, stems and leaves were cut out measuring 1.0 – 1.5 mm. The healthy *Echinodorus palaefolius* were accurately weighted before and then used for the phytoremediation experiment.

2.3. Phytoremediation Experiments

Experiments were conducted at room temperature in some test tubes, with the amount of part of *Echinodorus palaefolius* (roots, stems and leaves) were added to 100 mL solution in certain initial concentration, B and C designated contact time. The phytoremediation process was done in stagnant condition. Then, these roots, stems and leaves sample were dried in the oven until constant weight. The dried samples were crushed and diluted with 10 ml of 6 M HNO_3. Finally, the diluted samples were filtered to be analysed by atomic absorption spectrophotometer. All the experiments were conducted in duplicates and their mean values of Pb(II) final concentration, C_f are reported here.

2.4. Response Surface Methodology (RSM)

A total 18 experimental runs were designed as in Table 1 and performed to evaluate the phytoremediation of Pb(II) by roots, stems and leaves of *Echinodorus palaefolius* separately. It was designed according to Box-Behnken statistical design (BBD), one of RSM experimental design under DESIGN EXPERT 11.0.0 software (Stat-Ease Inc., Minneapolis,MN, USA).

Three factors viz., A: weight of plant (2, 6, 10 gr); B: initial concentration of Pb(II) (10, 20, and 30 mg/L) and C: contact time (30, 75 and 120 minutes) were selected as the independent variables (factors). While, R1, R2 and R3 for final concentration of Pb(II) (mg/L) from the phytoremediation process were taken as dependent variables to study the response of independent variables, respectively. The main effects and interaction of the factors were plotted in response surface and determined by fitting a polynomial equation (Eq. (1)) as well as by interpretation of Analysis of variance (ANOVA). A variable was considered significant if the calculated probability value (p) was smaller than the significance level (0.1).

$$Y = \beta_0 + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k} \beta_{ii} X_{ii}^2 + \sum_{i<j=2}^{k} \sum \beta_{ij} X_i X_j + \varepsilon \quad (1)$$

Here, *Y* is the dependent variable (response variable) to be modeled, *i* and *j* take value from 1 to the number of independent variables, β is constant regression coefficients of the model, *X is* the independent variables (factors), k is the number of factors studied and ϵ is the error.

3. Results and Discussion

3.1. RSM approach for optimization of phytoremediation of Pb(II)

Table 1. shows the *Echinodorus palaefolius* can reduce Pb(II) which the final concentrations range between 0.231 and 2.431 mg/L. The final concentration of Pb(II) represents amount of Pb(II) phytoadsorbed by part of *Echinodorus palaefolius*. The highest final concentration of Pb(II) adsorbed by roots, stems and leaves is 2.413 mg/L, 0.571 mg/L and 0.294 mg/L respectively. According to the results, the highest phytoremediation process occurs in the roots. Fibrous and fine root shaped is supposed to be the key factor in Pb(II) phytoadsorption process [3,4]. Phytoadsorption and phytoabsorption occurs simultaneously in the roots. The Pb(II) then translocate into upper part of plant by roots.

Table 1. Experimental design and results

No	A	B	C	Final concentration of Pb(II) (Cf)*		
	Weight of plant (gr)	Initial concentration (mg/L)	Contact time (minute)	R1 (mg/L)	R2 (mg/L)	R3 (mg/L)
1	10	10	30	1.324	0.425	0.231
2	10	10	30	1.325	0.429	0.236
3	10	30	75	1.342	0.431	0.268
4	10	30	75	1.340	0.432	0.271
5	6	20	120	1.370	0.369	0.261
6	6	20	120	1.372	0.371	0.267
7	6	20	75	1.412	0.432	0.262
8	6	20	75	1.412	0.431	0.259
9	10	30	120	2.077	0.435	0.278
10	10	30	120	2.078	0.438	0.283
11	10	10	120	2.097	0.452	0.264
12	10	10	120	2.098	0.457	0.265
13	6	10	120	2.122	0.467	0.373
14	6	10	120	2.124	0.469	0.378
15	6	20	120	2.124	0.433	0.291
16	6	20	120	2.125	0.435	0.294
17	2	30	120	2.401	0.57	0.285
18	2	30	120	2.413	0.571	0.286

*Final concentration of Pb(II) phytodsorbed by roots (R1), stems (R2) and leaves (R3)

The results of the 2FI models are presented in table 2. Regarding these F-values imply the models are significant. It also supported by p-values which are less than 0.1 indicating the individual terms in these models are significant on the effects. The Lack of fit of F-values implies all models are significant. The values of R-squared and adjusted R-squared for all models are close to 1 (table 3). Accordingly, it implies that strong relationship between these factors (weight of plant, initial concentration of Pb(II) and contact time) and these responses. The optimum conditions were predicted as weight of plant 2 gr, initial concentration of Pb(II) 30 mg/L and contact time of 120 min.

Table 2. Analysis of variance for model

	R1		R2		R3	
	F-value	**p-value**	**F-value**	**p-value**	**F-value**	**p-value**
Model	3.719	0.029	2.323	0.100	4.870	0.012
A- Weight of plant	0.208	0.657	0.0170	0.899	0.004	0.953
B- Initial Concentration	0.003	0.961	3.411	0.092	4.110	0.068
C- Contact time	6.184	0.030	0.303	0.593	5.840	0.034
AB	1.494	0.247	5.063	0.046	9.160	0.012
AC	0.094	0.764	0.285	0.604	3.220	0.100
BC	0.462	0.511	0.480	0.503	0.483	0.502
Residual						
Lack of Fit	7.612	0.020	38.833	<0.000	67.390	<0.000

Table 3. Correlation value of models

Source	**r^2**	**r^2 Adjusted**	**r^2 Predicted**
R1	0.670	0.490	0.493
R2	0.559	0.318	0.217
R3	0.727	0.578	0.498

Analysis of variance was conducted to evaluate the intensity of the effects of process conditions. The contact time and initial concentration are significant factors for the phytoremediation process due to probability values (p-values) for roots, stems and leaves calculated less than 0.1. The interaction is indicated significantly by interaction between weight of plant and initial concentration of Pb(II) as well as weight of plant and contact time. The resulting regression models equations for Pb(II) final concentration of roots (R1), stems (R2) and leaves (R3) after phytoremediation process are given in equation 2, 3 and 4, respectively.

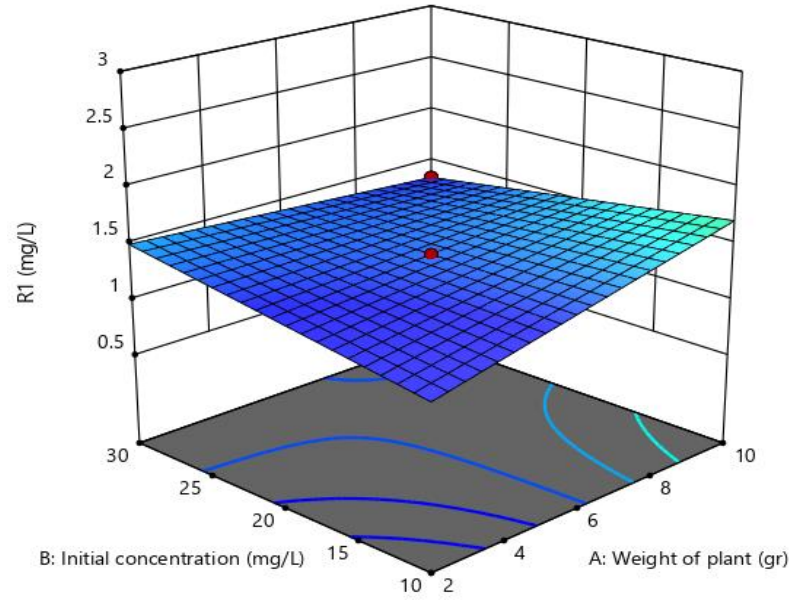
$$R1 = 1.412 + 0.114A + 0.009B + 0.584C - 0.194AB - 0.085AC + 0.113BC \quad (2)$$

$$R2 = 0.432 + 0.005A + 0.46B + 0.018C - 0.051AB - 0.021AC - 0.016BC \quad (3)$$

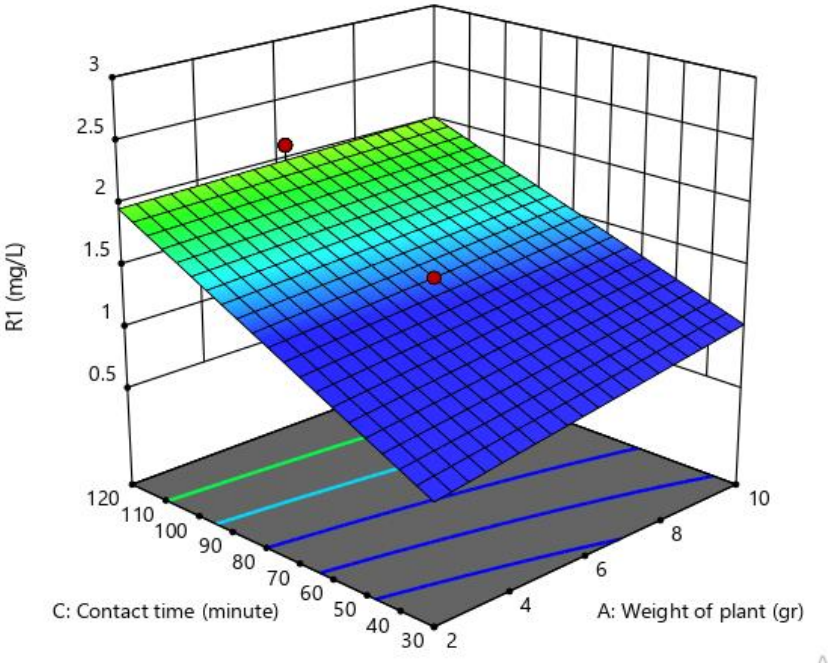
$$R3 = 0.261 - 0.001A - 0.030B + 0.047C + 0.040AB - 0.041AC - 0.010BC \quad (4)$$

3.2 Effect of factors on phytoremediation of Pb(II)

The combined effects of process factors were determined to analyse and optimize the process design and constructed in 3D plot as shown in figure 1 - 3. The interaction between initial concentration and weight of plant for roots, stems and leaves in figure 1(a), figure 2(a) and figure 3(a) respectively. These shows the higher phytoadsorption of Pb(II) was attained with the decrease initial concentration. The higher initial concentration of Pb(II) increases the toxicity of plant which might decrease the phytoadsorption process [10]. The weight of plant also influence indirectly, as seen in these figures, the final concentration becomes higher with the increase in weight of plant.



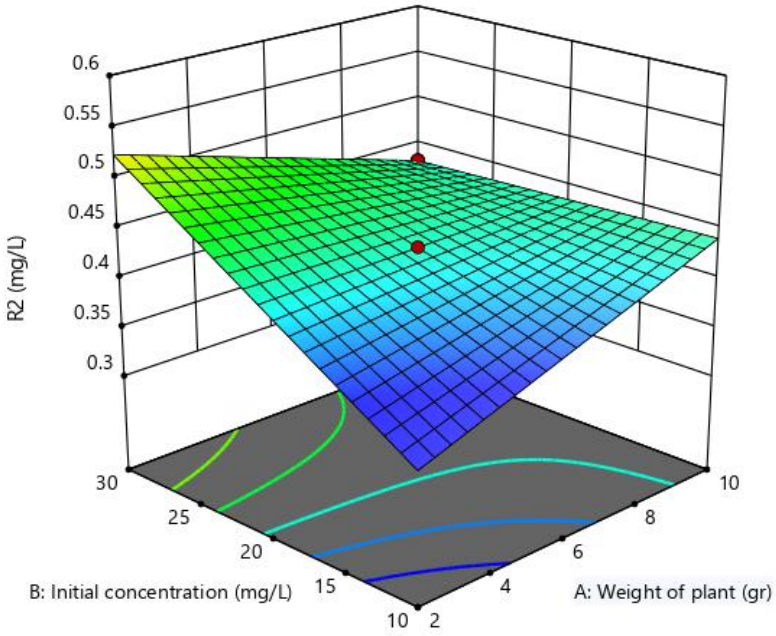
(a)



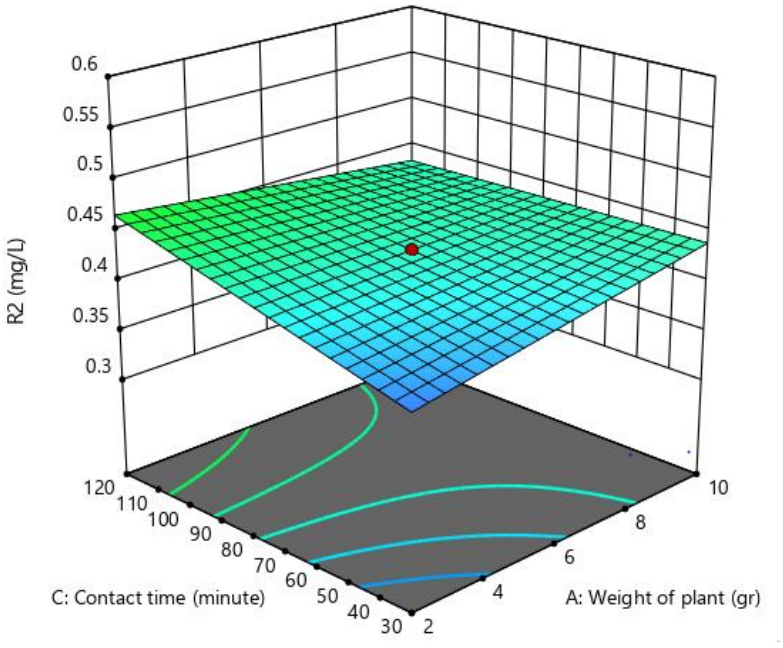
(b)

Figure 1. (a) Response surface 3D plots of roots indicating interaction effects of weight of plant and initial concentration; (b) Response surface 3D plots indicating interaction effects of weight of plant and contact time.

Figure 1(b), Figure 2(b) and Figure 3(b) show the 3D response surface plot of the interaction effect between weight of plant and contact time for roots, stems and leaves respectively. It is seen that a longer time for contacting between roots, stems and leaves resulting the higher Pb(II) adsorbed. It is similar to previous report that the contact time influencing the phytoremediation process [6, 7]. The Pb(II) phytoadsorption may occur in the part of *Echinodorus palaefolius* by the increasing time. Then it is accumulated into the part of *Echinodorus palaefolius*, mainly in bottom part (roots). The maximum phytoremediation was achieved at 120 minutes contact time.

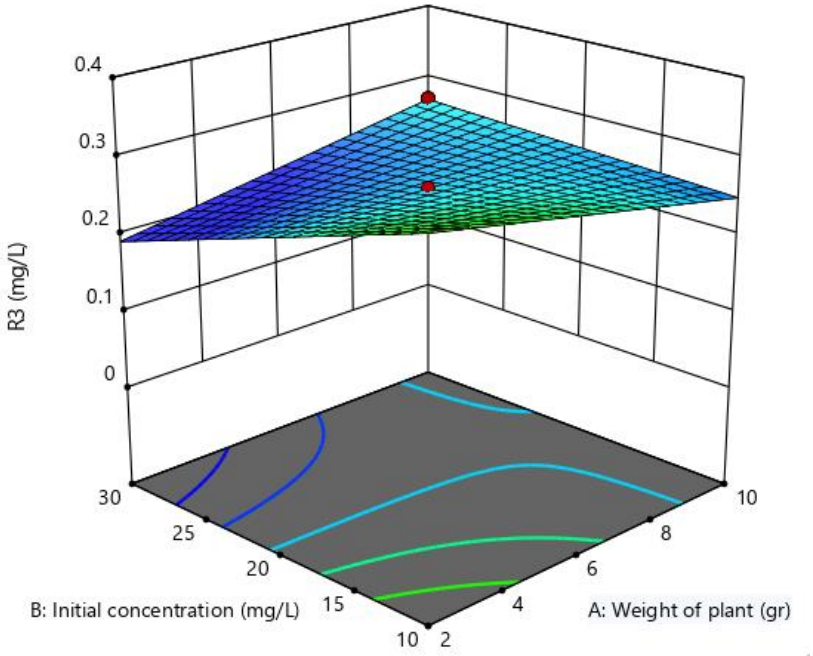


(a)

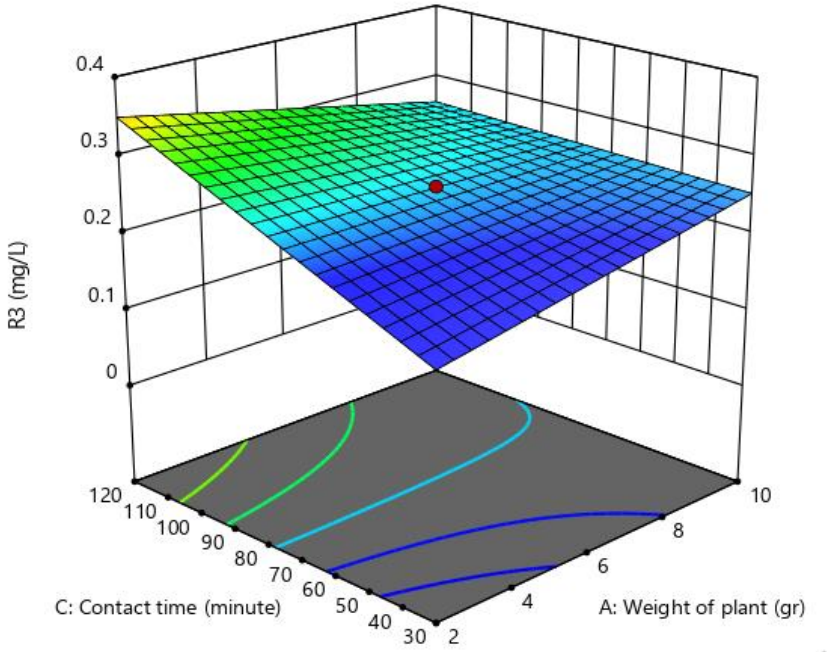


(b)

Figure 2. (a) Response surface 3D plots of stems indicating interaction effects of weight of plant and initial concentration; (b) Response surface 3D plots indicating interaction effects of weight of plant and contact time.



(a)



(b)

Figure 3. (a) Response surface 3D plots of leaves indicating interaction effects of weight of plant and initial concentration; (b) Response surface 3D plots indicating interaction effects of weight of plant and contact time.

4. Conclusion

The highest phytoadsorption of Pb(II) using *Echinodorus Palaeofolius* occurred in roots. The highest final concentration of Pb(II) adsorbed by roots, stems and leaves is 2.413 mg/L, 0.571 mg/L and 0.294 mg/L respectively. According to 2FI model, optimum condition is predicted at weight of plant 2 gr,

initial concentration of Pb(II) 30 mg/L and contact time 120 minutes. The initial concentration and contact time are significant factors influencing the phytoremediation process.

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