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Statistical Optimization of Process Parameters for Dye Biosorption onto Biomass

Hussain Majeed Flayeh*, Yaseen R. Hasan
* Instructor College of Engineering-University of Baghdad
Instructor Ministry of Higher Education & Scientific Research
hussmf200211@yahoo.com

Abstract

In the present study, sorption of reactive azo dye, Blue H_3 R from aqueous solution on an efficient, economically biomass (sewage sludge) was investigate. Batch experiments have been carried out to find the effect of various parameters such as initial dye concentration, adsorbent dosage, pH , and temperature on the sorption of dye using biomass. A total of 30 sorption experimental runs were carried out employing the detailed conditions designed by response surface methodology based on the Box-Wilson design is used to optimize the bisorption of dye using Central Composite Design (CCD) model. The analysis of variance (ANOVA) depicted that the quadratic model was suitable for all the responses. The CCD model was designed for parameters using Design Expert software (Version 9.0). The optimum conditions for the sorption of dye were found to be as follows: Initial dye concentration 70 mg/l, biomass concentration 17.5 g/l, pH 3, and temperature 35℃. At these optimized conditions the maximum dye removal efficiency was found to be 91%. A coefficient of determination (R2) value of 0.8919 shows the fitness of RSM in this work.

Keywords: optimization; reactive azo dye, sewage sludge, Response surface Methodology, ANOVA, Design expert® 9 software.

Introduction

The effluents from textile, leather, food processing, dyeing, cosmetic, paper, and dye manufacturing industries are important sources of dye pollution [1]. Treatment of dyed effluents presents several problems mainly due to toxicity and recalcitrance of dyestuffs. The wastewater generated by textile industry is rated as one of the most polluting among all industrial effluents. The toxic wastes from industries affect visibility, photosynthesis and also aquatic life [2]. The textile industry utilizes about 10000 different dyes and pigments. The worldwide annual production of dyes is over $7*10^5$ tons [1, 4, and 8].Dyes are synthetic aromatic water-soluble dispersible organic colorants, having potential application in various industries. The dyestuff usage has been increased day by day because of tremendous increase of industrialization and man's urge for color [17]. Dyes tinctorial value is high: less than 1ppm of the dye produces obvious coloration [6].

It is quite difficult to treat the effluents by the conventional biological and physic-chemical processes, e.g. light, wash, heat, and oxidizing agents, used in regular treatment[9]. That is because of complexicity of the dyes aromatic molecular structures[16]. Adsorption is the most effective physical process in the treating these dye waste waters. Today activated carbon is commonly used for adsorption in many treatment plants[9]. But the producing costs for activated carbon is very high, there is a need of an alternative material that is more cost efficient[9]. A low costs adsorbent is defined as one which is abundant in nature or one that is produces as a byproduct in another industry[16].

Some existing technologies (oxidative destruction via UV/ozone treatment, photocatalytic degradation, electrochemical reduction etc.) may have a certain efficiency in the removal of dyes, but their initial and operational cost are so great, that they constitute an inhibition to dyeing and finishing industries[14, 4, and19]. On the other hand, low cost technologies do not allow a desired degree of color removal or have certain disadvantages. Therefore, in order to achieve the desired degree of treatment, it is necessary to integrate biological, chemical and physical processes.

At present colored wastewater is treated by physical, chemical and biological methods. Biological methods which include Biosorption process employing biopolymers (such as sawdust, wood chips, chitin/chitosan, starch, cyclodextrin and cross linked chitosan / cyclodextrin) and nonviable microbial (fungi, algae and bacteria) biomass has emerged as one of the powerful and attractive option since it is inexpensive, effective and simple to operate. Biosorption involves a combination of active and

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passive transport mechanisms starting with the diffusion of the adsorbed component to the surface of the microbial cell. A number of biomaterials have been used as biosorbents [5, 26]. Biospriton of malachite gree from aqueous solutions onto aerobic granolues:kinetics and equilibrium studies [27].

Some dyes, especially azo dyes, are known to be biorefractory pollutants even with carefully selected microorganism and under favorable conditions. Azo dyes are characterized by the presence of one or more azo bonds (−N=N-) and account for 60% to 70% of all textile dyes used. It is estimated that approximately 8 × 10^5 tons (t) of dyes are produced annually worldwide, and about 50% of them are azo dyes [15, 29].

Since little is known on the biosorption of dyes to microbial biomass, adsorptive properties of the microorganism for dyes should be investigated. Biological wastewater treatment produces a biological sludge (biosolid) including of inert materials and microorganisms. Return activated sludge (RAS) or waste activated sludge (WAS) can be used for biosorption of dye-contaminated industrial effluents. RAS consists of a variety of living organisms. However, waste activated sludge (WAS) consists of the non-living microorganisms.

The present study use a biological sludge (biosolid) for the removal of reactive azo dye in simulated wastewater. Another part of this study involved the use of response surface methodology (RSM) and finding an applicable approximating function for predicting and determining the further response, and studying the optimum working state. The factors (variables) of initial dye concentration, biosorbent dosage, temperature, and pH were investigated [23]. RSM is a kind of mathematical and statistical technique for designing experiments, building models, evaluating the relative significance of several independent variables, and determining the optimum conditions for desirable responses [28, 3, and 6]. The two most common designs extensively used in RSM are the central composite design (CCD) and the Box-Wilson design (BWD). The CCD is ideal for sequential experimentation and allows a reasonable amount of information for testing lack of fit while not involving an unusually large number of design points [28-20].

Materials and methods

Material

The sewage sludge (biosolid) used in this study was taken from Al-Rostomia,a sewage treatment plant drying bed in Baghdad Iraq. The collected biomass was thoroughly washed with tap wateruntil all the dirt was removed finely the biomass was washed with de-ionized water until the entire colour of the material was removed. The result wet biomass was dried at 70°C for 6 h. The physical, chemical characteristic (dead biomass) were measured and listed in Table 1. Anaerobic and facultative anaerobic microorganisms (Aeromonas species, E-coli, Pseudomonas aerginrsa, Clostridium, Staphylococcus sp and Salmonella sp, Rhizopusarrhizus, Saccharomyces erevisiae) were found in biomass from the drying bed using API Instrument (Biomerieux, France).

Table 1 physical chemical and biological characteristic of DAB

properties	value	properties	value
Particle diameter, mm	0.775	Volatile Suspended, mg/L	78126
Surface area, m^2/g (*)	94.53	pH	5.5-6.3
Actual density, kg/m^3	1741.6	CEC, meq/100g (***)	51.2
Bulk density, kg/m^3 (**)	610		
Particle porosity	0.584	Heavy metals mg/l	0.02
Total Suspended Solid, mg/L	153950	Microorganism species Total Colony-forming unit(CFU)/ml	$4.1*10^6$

(*) Surface area analyzer, BET method, Quantachrome.com.(USA)
(**) Apparent density instrument, Autotap, Quantachrome.(USA),
(***) CEC Cat ion Exchange Capacity,

The reactive azo dye (Blue H_3R, Solid/powder, Wave length 585nm, solubility 90g/l, Mwt. 763.5, India, the chemical structure shown in Fig. 1, was obtained from Al-Hilla textile factory south of Baghdad. Simulated stock solution was prepared by dissolving 10 g of dye in one litter of distilled water then diluted to the desired solution concentration.

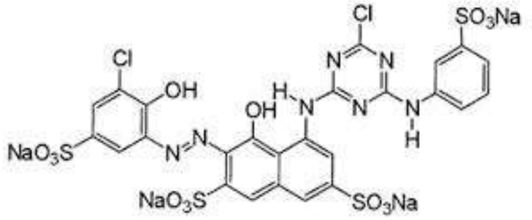


Fig. Chemical stricture of reactive azo dye, Blue H3R

The concentration of the dye solution in the experimental were determined at the maximum wave length $\lambda = 585\text{nm}$ using a spectrophotometer (Model SP-3000 plus, Optima Co., 2003, Japan), using OptiVie 3.2 software Windows-2000/XP (survey scan at wavelengths 200-1100 nm). The residual dye concentration in the reaction mixture was analyzed by centrifuging (Model: J2-21, BECKMAN) at 5000 rpm, before measuring the absorbance of the supernatant of the sample. Calibration curve were prepared by measuring the absorbance of different known concentration of dye solution at λ_{max} (plotted between absorbance and concentration of the dye solution). These analyses were carried out in duplicate. The percentage removal of dye from solution was calculated as follows:

$$\%\ dye\ removal = \left(\frac{C_i - C_f}{C_i}\right) \times 100 \qquad (1)$$

where C_i is the initial concentration of dye in the solution and C_f is the final concentration of dye in the solution, and the removal was taken as a response (Y) of the experimental design.

Experimental Design and Data Analysis

In this study, RSM was used for the optimization of various process parameters to study the dye removal efficiency combined with the factorial experimental design of CCD. Multifactor response surface methodology RSM is a useful method for studying the effect of several variables influencing the responses by varying them simultaneously and carrying out a limited number of experiments. The CCD is an effective design that is ideal for sequential experimentation and allows a reasonable amount of information for testing the lack of fit while not involving an unusually large number of design points. It was first announced by Box Wilson in 1951, and is well suited for fitting a quadratic surface, which usually works well for the process optimization [23-21].

In the present study, a CCD was employed for determining the optimum conditions for dye removal. The experimental results were analyzed using Design-Expert Version: 9.01.0, and the regression model was proposed. initial dye concentration (x1), adsorbent dosage (x2), pH (x3), and temperature (x4) were chosen as four independent variables in the biosorption process. Accordingly, the CCD matrixes of 30 experiments covering the full design of four process factors (parameters) chose to study, names and levels are shown in the following table.

All independent variables were coded to four levels as Xi according to equation 1.

$$\mathbf{X_i} = \frac{(x_i - x_0)}{\Delta x} \qquad (1)$$

Where Xi is independent variable Xi = , i = 1,2,3,……k, x_i is the real value of an independent variable, x_0 is the real value of the independent variable at the centre point, and Δx is the step change. According to the obtained experimental data, the levels of the four main parameters investigated in this study are presented in Table 2.

A polynomial (Equation 2) was developed to estimate the behavior of the percentage removal of dye.

$$Y = \beta_0 + \sum_{i=1}^{k} \beta_i x_i + \sum_{i=1}^{k} \beta_{ii} x_i^2 + \sum_{i=1}^{k-1} \sum_{i=2}^{k} \beta_{ij} x_i x_j + e_i \qquad (2)$$

Where Y is the response, β0 was the intercept term, β_i, $\beta_{ii,}$ β_{ij} are the first-order, quadratic, and interaction effects, respectively; i and j are the index numbers for factor; and e_i is the residual error [16, 10].

Results and discussion

A statistical approach using a CCD was used for efficiency removal of dye and for determining the interaction between these factors. For the response surface methodology involving CCD, a total of 30 experiments was conducted for four factors Table 2, at five levels Table 3 with six replicates at center point. Table 4 provides a list of independent variables and coded factor levels. The total number of design points N of a rotatable design is determined from $N = 2^k + 2k + n_0 = n_j + n_\propto + n_0$ is given by the expression: ($2^k = 2^4 = 16$; star points) + 2 k (2 × 4 = 8; axial points) + 6 (center points; 6 replications).

A central composite design in made rotatable by the choice of α (value for readability) depends on the number of variables K(i.e. for four variables, K=4, $\alpha = \pm\sqrt{K} = 2$).

An RSM is appropriate when the optimal region for running the process has been identified. The design used for the optimization and observed responses for 30 experiments are given in Table 4. Table(4) Central composite design matrix of coded and real values along with the experimental values for percentage biosorption of dyes.

In Equation 3, Y is the percentage removal of dye; and x1, x2, x3, and x4 are the corresponding coded variables of initial dye concentration (x1), adsorbent dosage (x2), pH (x3), and temperature (x4), respectively.

The final model according to the RSM results, polynomial regression modeling was performed on the responses of the corresponding coded values of the four different prameters, and the results were evaluated. The predicted response (Y) for the percentage dye removal efficiency of samples treated was obtained using Equation 4:

$$Y = -50.03958 + 1.62764 * \text{In. Dye Conc.} + 4.67167 * \text{Biomass con.} - 24.22188 * \text{pH} + +4.07937 * \text{Temp.} + +0.015278 * \text{In. Dye Conc.} * \text{Biomass con.} - 0.067708 * \text{In. Dye Conc.} * \text{pH} - 0.011458 * \text{In. Dye Conc.} * \text{Temp.} - -0.46250 * \text{Biomass con.} * \text{pH} + +0.0625 * \text{Biomass con.} * \text{Temp.} + +0.14688 * \text{pH} * \text{Temp.} - -8.34722E-003 * \text{In. Dye Conc.}^2 - 0.095778 * \text{Biomass con.}^2 + +2.21563 * \text{pH}^2 - 0.071250 * \text{Temp.}^2 + e_i$$

Analysis of variance

Analysis of variance (ANOVA) values for the quadratic regression model obtained from CCD employed in the optimization of dye removal are listed in Table 5.

On the basis of the experimental values, statistical testing was carried out using Fisher's test for ANOVA.

The statistical significance of the second-order equation revealed

that the regression is statistically significant ($P < 0.0001$); however, the lack of fit is not statistically significant at 99% confidence level. Table 3 depicts the significance of the regression coefficients and ANOVA for the regression model, respectively. The results indicate that the response equation proved to be suitable for the CCD experiment [24,10].

The model's F value of 3.75 in these tables implies that the model is significant for the removal of the dye. If the model has a very high degree of adequacy for predicting the experimental results, the computed F value should be greater than the tabulated F value at a level of significance α. Thus, the calculated F value ($F_{model} = 3.75$) was compared with the tabulated F value ($F_{0.05,df,(n-df+1)}$) at a significance level of 0.05, when the df for the model was 14 and n = 30. It can be observed that the tabular F value ($F_{0.05,14,17} = 2.31, 2.38$ at $F_{0.05,14,17}, F_{0.05,14,17}$, respectively) is clearly less than the calculated F value of 3.75. The model's P values less than 0.05 indicate that the model terms are significant, whereas values greater than 0.1 are not significant. The fit of the models were controlled by the coefficient of determination R^2. Based on the ANOVA results, the models report high R^2 value of 89.19% for dye removal using biosorbent. Also, an acceptable agreement with the adjusted determination coefficient is necessary.

This indicates that the regression model provides a good explanation of the relationship between the independent variables and the response. The diagnostic plots given in Figures 2, 3, and 4 were used for estimating the adequacy of the regression model.

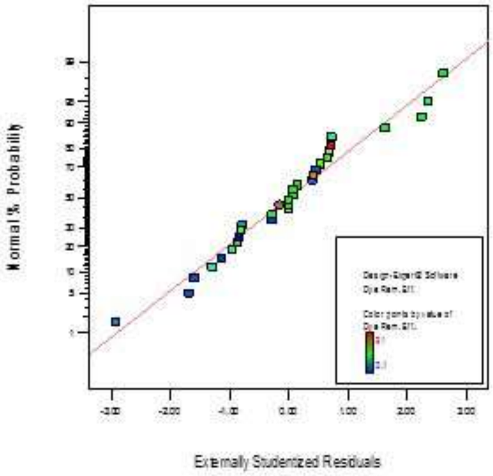


Figure 2 The studentized and normal percentage probability plot of dye removal

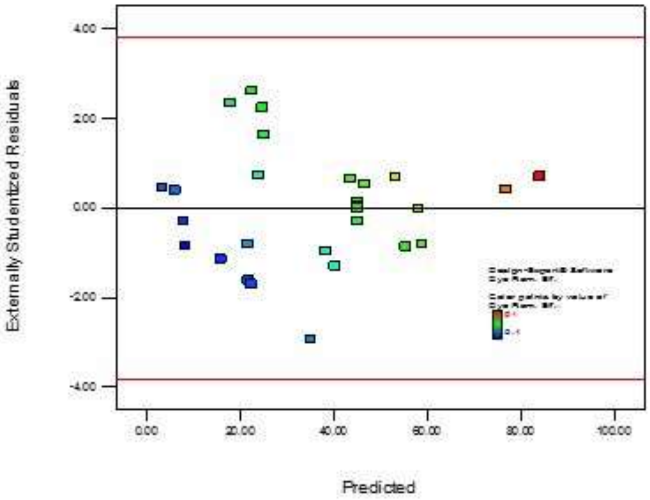


Figure 3 The predicted Dye removal% and studentized residual plot.

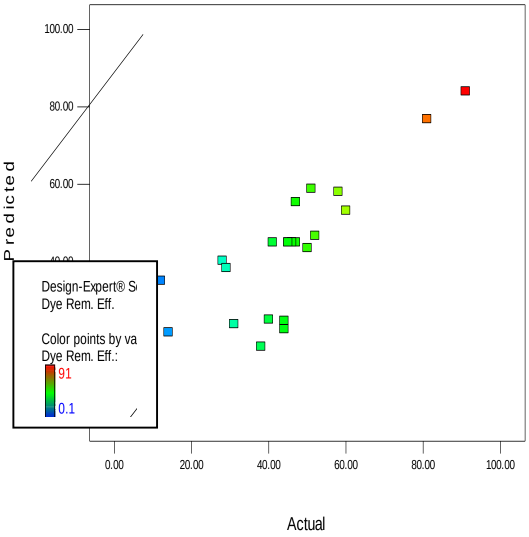
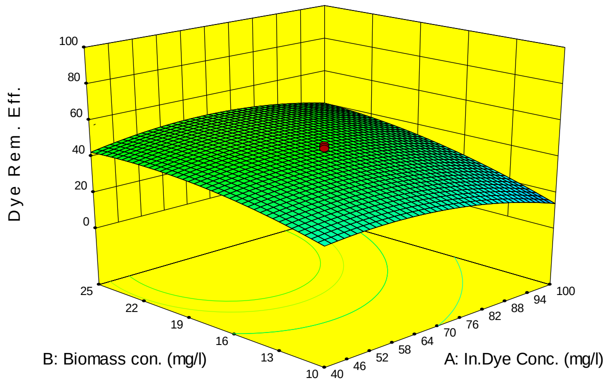
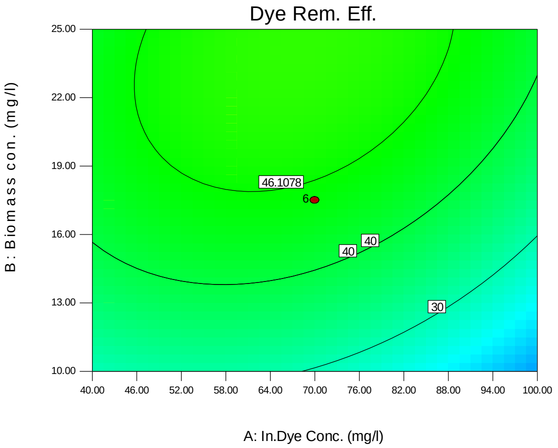


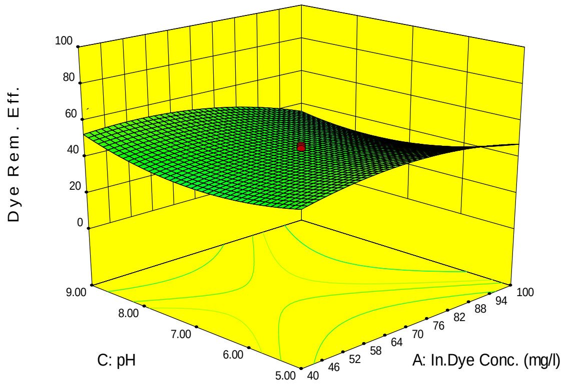
Figure 4 The actual and predicted Dye removal Percentage.

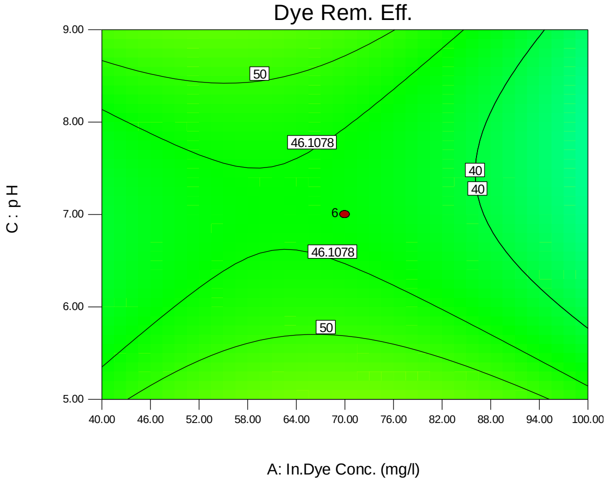
The normal percentage probability and studentized residual plot are shown in Figure 2. The data points indicate that neither response transformation was required nor there was any apparent problem with normality. Figure 3 depicts the studentized residual and predicted dye removal percentage of dye biosorption. The actual and the predicted dye removal percentage values are given in Figure 4. It can be observed that there are tendencies in the linear regression fit, and the model adequately explains the experimental range studied. The actual dye removal percentage value is the measured result for a specific run, and the predicted value is evaluated from the independent variables in the CCD model [29, 12].

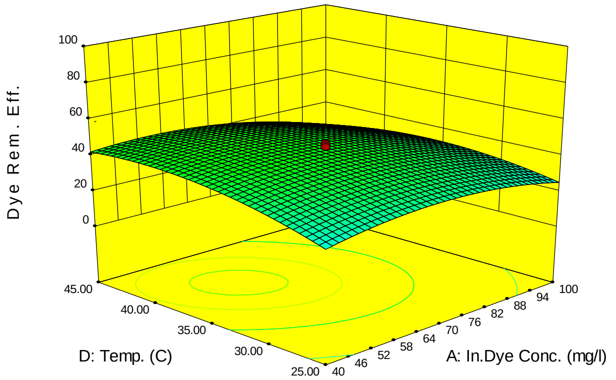
Interactive effect of processes of independent variables to understand the impact of each variable, three dimensional (3D) plots were made for the estimated responses, which were the bases of the model polynomial function for analysis to investigate the interactive effect of the two factors on the dye removal percentage within the experimental ranges given in Figures below. The inferences so attained are discussed below [11].

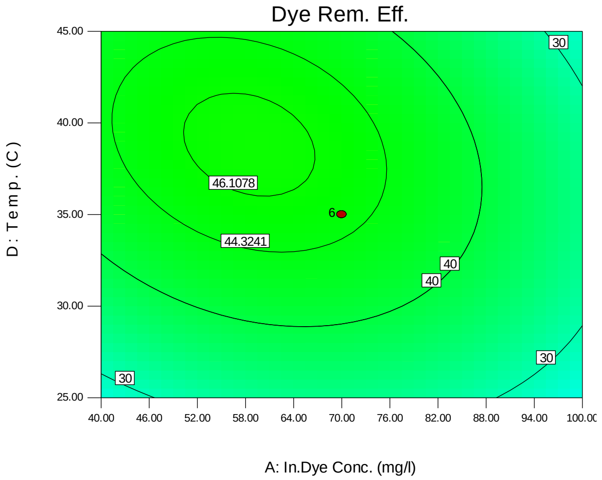


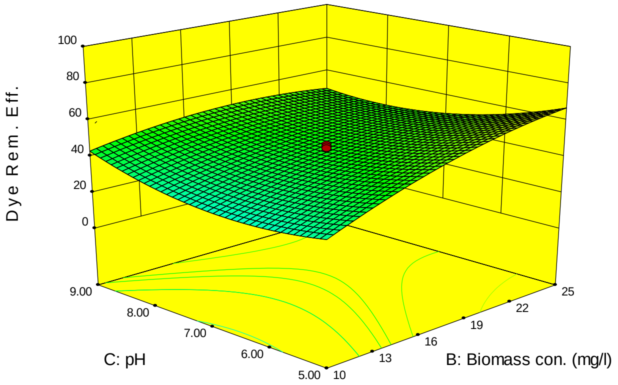


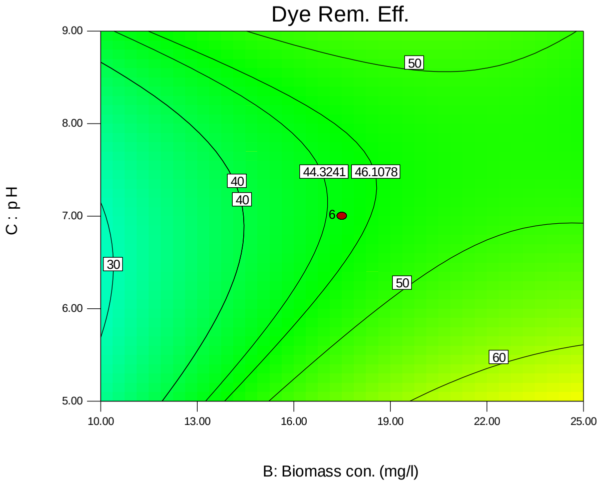

Dye Rem. Eff.
100
80
60
40
20
0
C: pH
9.00
8.00
7.00
6.00
5.00
A: In.Dye Conc. (mg/l)
40
46
52
58
64
70
76
82
88
94
100

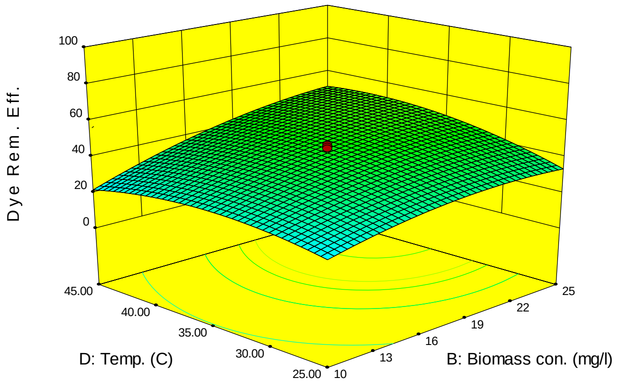

Dye Rem. Eff.
C: pH
9.00
8.00
7.00
6.00
5.00
50
46.1078
40
40
6
46.1078
50
40.00
46.00
52.00
58.00
64.00
70.00
76.00
82.00
88.00
94.00
100.00
A: In.Dye Conc. (mg/l)

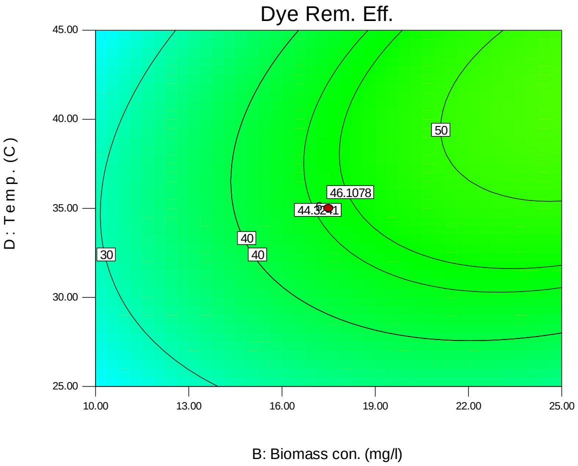

Dye Rem. Eff.
100
80
60
40
20
0
D: Temp. (C)
45.00
40.00
35.00
30.00
25.00
A: In.Dye Conc. (mg/l)
40
46
52
58
64
70
76
82
88
94
100

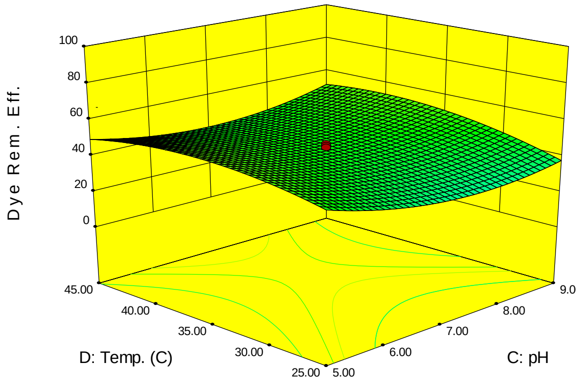

Dye Rem. Eff.
D: Temp. (C)
45.00
40.00
35.00
30.00
25.00
30
46.1078
6
44.3241
40
40
30
30
40.00
46.00
52.00
58.00
64.00
70.00
76.00
82.00
88.00
94.00
100.00
A: In.Dye Conc. (mg/l)

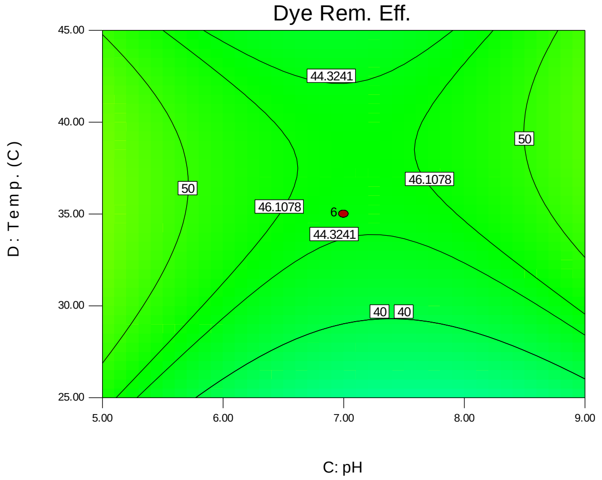

Dye Rem. Eff.
100
80
60
40
20
0
C: pH
9.00
8.00
7.00
6.00
5.00
B: Biomass con. (mg/l)
10
13
16
19
22
25


Dye Rem. Eff.
C: pH
9.00
8.00
7.00
6.00
5.00
50
40
40
44.3241
46.1078
6
30
50
60
10.00
13.00
16.00
19.00
22.00
25.00
B: Biomass con. (mg/l)









Interactive effect of initial dye concentration and biomass concentration. To investigate the integrated effect of initial dye concentration and biomass concentration, RSM was used and the result was given in the form of 3D plots, and the 2D contour plots. As indicated in Figure 4, the initial dye concentration and biomass concentration have considerable influence on the dye removal efficiency achieved. Indicates that the decreasing initial dye concentration and increasing biomass concentration partially increased removal efficiency of dye, and the dye removal efficiency decreases with increasing initial dye concentration due to the saturation and quick exhaustion of the binding sites on the biosorbent reached as the number of dye molecules per unit volume increased. At low initial concentration, dye molecules are bioadsorbed on a specific binding site, however, when the concentration increases, there exist reduction in immediate solute biosorption due to the lack of available binding sites [24]. The dye removal efficiency increase with increasing biomass dosage resulted in an increase in the amount of biosorption dye.

Interactive effect of initial dye concentration and pH. To investigate the integrated effect of initial dye concentration and pH, RSM was used and the result was given in the form of 3D plots, and the 2D contour plots. As indicated in Figures , the decreasing pH (<7) and increasing pH(>7) increase H+ and OH- ions respectively which adsorbed quit strongly and therefore the biosorption of other ions is affected by the pH of the solution. Change of pH affects the adsorptive process through dissociation of functional groups on the biosorbent surface active site. This subsequently leads to a shift in reaction kinetic and equilibrium characteristics of biosorption process.

As indicated in above Figures, The dye removal efficiency is found to increase with rising temperature from (25-35 ℃) there are increased, and decreased with rising temperature from from (35-45 ℃). The temperature has two major effects on the biosoebtion process. Increasing the temperature is known to increase the rate of diffusion of the adsorbed molecules across the external boundary layer and the internal pores of the adsorbent particles, owing to the decrease in the viscosity of the solution. In addition, changing temperature will change the equilibrium capacity of the adsorbent for a particular adsorbate [22].

The optimum condition of all parameters effecting the percentage removal efficiency of dye was predicted by using prediction profiler of the software. The maximum removal efficiency was predicted to be 91% which was obtained at a initial dye concentration of 70 mg/l, biomass concentration of 17.5 g/l, pH of 3, and temperature of 35°C. The optimum condition was repeated three times and dye removal efficiencies of 92.2, 90.8, and 91.1 were resulted. The average of 91.4% dye removal efficiency was found close to the model prediction of 91%.

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Table 2 Names and levels of process factors (parameters)

Factor	Units	Low Level (-1)	High Level (+1)
A-Initial Dye Conc.	mg/l	40	100
B-Biomass Conc.	g/l	10	25
C-pH		5	9
D-Temperature	℃	25	45

Table 3 Experimental range and levels of the independent parameters:

Independent Parameters	Range and level				
	-∝	-1	0	+1	+∝
x1, Influent conc.(mg/l)	50	162.5	275	387.5	500
x2, Biomass conc.(g/l)	1	3.25	5.5	7.75	10
x3, pH	2	4	6	8	10
x4, Temp.℃	30	33.75	37.5	41.25	45

Table 4 Central composite design experiments and experimental results

Std	Run	Experiment design Coded values				Experiment plan				Response 1
						Factor 1	Dye Rem. Eff.	Factor 3	Factor 4	
		x1	x2	x3	x4	A:In.Dye Conc.	B:Biomass con.	C:pH	D:Temp.	Dye Rem. Eff.
27	1	0	0	0	0	70.00	17.50	7.00	35.00	41
30	2	0	0	0	0	70.00	17.50	7.00	35.00	45
25	3	0	0	0	0	70.00	17.50	7.00	35.00	46
11	4	1	-1	1	-1	40.00	25.00	5.00	45.00	51
20	5	0	2	0	0	70.00	32.50	7.00	35.00	50
6	6	-1	1	-1	1	100.00	10.00	9.00	25.00	38
29	7	0	0	0	0	70.00	17.50	7.00	35.00	46
23	8	0	0	0	-2	70.00	17.50	7.00	15.00	0.1
4	9	-1	-1	1	1	100.00	25.00	5.00	25.00	60
3	10	-1	-1	1	-1	40.00	25.00	5.00	25.00	28
10	11	1	-1	-1	1	100.00	10.00	5.00	45.00	5

24	12	0	0	0	2	70.00	17.50	7.00	55.00	44
19	13	0	-2	0	0	70.00	2.50	7.00	35.00	8
16	14	1	1	1	1	100.00	25.00	9.00	45.00	29
7	15	-1	1	1	-1	40.00	25.00	9.00	25.00	40
14	16	1	1	-1	1	100.00	10.00	9.00	45.00	5
13	17	1	1	-1	-1	40.00	10.00	9.00	45.00	52
2	18	-1	-1	-1	1	100.00	10.00	5.00	25.00	7
28	19	0	0	0	0	70.00	17.50	7.00	35.00	47
5	20	-1	1	-1	-1	40.00	10.00	9.00	25.00	12
9	21	1	-1	-1	-1	40.00	10.00	5.00	45.00	7
21	22	0	0	-2	0	70.00	17.50	3.00	35.00	91
1	23	-1	-1	-1	-1	40.00	10.00	5.00	25.00	44
18	24	2	0	0	0	130.00	17.50	7.00	35.00	10
22	25	0	0	2	0	70.00	17.50	11.00	35.00	81
17	26	-2	0	0	0	10.00	17.50	7.00	35.00	31
12	27	1	-1	1	1	100.00	25.00	5.00	45.00	58
26	28	0	0	0	0	70.00	17.50	7.00	35.00	45
15	29	1	1	1	-1	40.00	25.00	9.00	45.00	47
8	30	-1	1	1	1	100.00	25.00	9.00	25.00	14

Table 5 ANOVA regression model for Dye Removal

ANOVA for Response Surface Quadratic model						
Analysis of variance table [Partial sum of squares - Type III]						
	Sum of		**Mean**	**F**	**p-value**	
Source	**Squares**	**df**	**Square**	**Value**	**Prob > F**	
Model	11756.72	14	839.77	3.75	0.0079	significant
A-A	*477.04*	*1*	*477.04*	*2.13*	*0.1650*	
B-B	*2420.04*	*1*	*2420.04*	*10.81*	*0.0050*	
C-C	*77.04*	*1*	*77.04*	*0.34*	*0.5662*	
D-D	*406.73*	*1*	*406.73*	*1.82*	*0.1977*	
AB	*189.06*	*1*	*189.06*	*0.84*	*0.3726*	
AC	*264.06*	*1*	*264.06*	*1.18*	*0.2946*	
AD	*189.06*	*1*	*189.06*	*0.84*	*0.3726*	
BC	*770.06*	*1*	*770.06*	*3.44*	*0.0834*	
BD	*351.56*	*1*	*351.56*	*1.57*	*0.2293*	
CD	*138.06*	*1*	*138.06*	*0.62*	*0.4445*	
A^2	*1548.00*	*1*	*1548.00*	*6.91*	*0.0189*	
B^2	*796.12*	*1*	*796.12*	*3.56*	*0.0788*	
C^2	*2154.35*	*1*	*2154.35*	*9.62*	*0.0073*	
D^2	*1392.43*	*1*	*1392.43*	*6.22*	*0.0248*	
Residual	3357.95	15	223.86			
Lack of Fit	*3335.95*	*10*	*333.59*	*75.82*	*< 0.0001*	*significant*
Pure Error	*22.00*	*5*	*4.40*			

Table 6 Design expert® 9 software Design and Model Summary

Design Summary			
File Version	9.0.1.0		
Study Type	Response Surface	**Runs**	30
Design Type	Central Composite	**Blocks**	No Blocks
Design Model	Quadratic	**Build Time (ms)**	8.00
Model Summary Statistics			
Std.Dev.	15.28	**R-Squared**	0.8919