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Application of chitosan as an adsorbent to treat rice mill wastewater—Mechanism, modelling and optimization

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ABSTRACT

The objectives of the present study is to investigate the chitosan as an adsorbent to treat rice mill wastewater under different process conditions such as agitation time (2–6 min), initial pH (2.5–6.5), chitosan dose (400–800 mg/l) and settling time (10–30 min) in order to study the removal efficiency of chemical oxygen demand (COD) and total suspended solids (TSS). The results showed that, all process variables have significant effect on the removal efficiencies. The optimum process conditions were determined (agitation time of 4 min, initial pH of 4.5, chitosan dose of 600 mg/l and settling time of 20 min) and showed high removal efficiencies (COD: 98% and TSS: 95%). FT-IR spectrophotometry was used to analyze and confirm the adsorption process. From the experimental data, Box–Behnken design (BBD) was used to develop the second order polynomial models with high coefficient of determination values (COD: 0.991 and TSS: 0.989).

1. Introduction

In recent years, clean environment is necessary for smooth living and better health of human beings (Pooja Vaishnav & Anupam Bala, 2012). For that fact, in last few decades, considerable attention has been paid to treat the industrial wastewaters, which are usually discharged on land or into different natural water sources such as rivers and canals. Higher growth rate of industrialization will increase the adverse impact on ecosystem drastically due to the discharge of untreated industrial wastewater, which could degrade the environment (Gálvez, Gomez, Hontoria, & Gonzalez-Lopez, 2003). Among, various kinds of wastewaters, rice mill wastewater are recognized as one of the toxic wastewater, which contains high amount of organic and inorganic pollutants (Rajesh, Bandyopadhyay, & Das, 1999). Therefore, there is a critical need to treat the rice mill wastewater before discharged into the environment. An extensive literature survey showed that, treatment methods such as UASB reactor, microbial fuel cells (MFC), electrocoagulation and biological treatment (Manaswini, Partha, Tanaji, & Ghangrekar, 2010; Manogari, Daniel, & Krastanov, 2008; Pooja Vaishnav & Anupam Bala, 2012; Rajesh et al., 1999) techniques have been employed to treat the rice mill wastewater. But, these treatment processes requires longer time, higher extensive setup and high energy consumption. So, these techniques are not economically applicable in small community systems. Moreover, in these techniques addition of chemicals generates a considerable quantity of secondary pollutants such as solid sludge, which also pose serious environmental problems (Kalyuzhnyi, Gladchenko, Starostina, Shcherbakov, & Versprille, 2005). Therefore, the research interest in use of alternative treatment processes for treatment of rice mill wastewater without producing secondary pollutants has nowadays intensified.

Chitosan is a linear copolymer of D-glucosamine and N-acetyl-D-glucosamine produced by the deacetylation of chitin. Chitosan have individual properties among various biopolymers especially the presence of primary amino groups ($-NH_2$) and higher nitrogen content (Synowiecki & Al-Khateeb, 2003). The potential application of chitosan has been widely recognized in various fields such as biomedical engineering, pharmacy, dentistry, ophthalmology, biotechnology, chemistry, cosmetics, textile, oenology, agriculture and photography (Rinaudo, 2006). Chitosan is also widely applied in wastewater treatment process, because it is environmentally friendly, non-toxic, bio-degradable, renewable and has the ability remove the pollutants with outstanding pollutant-binding capacities (Grégorio & Pierre-Marie, 2008; Ravi Kumar, 2000). Chitosan has been used as an adsorbent to treat various wastewaters such as food processing industrial wastewaters, brewery wastewater, pulp and paper mill wastewater, olive oil wastewater, effluent containing metal ions and phenol derivatives (Amit & Mika, 2009).

However, from the extensive literature analysis, there is no research reports are available on the treatment of rice mill

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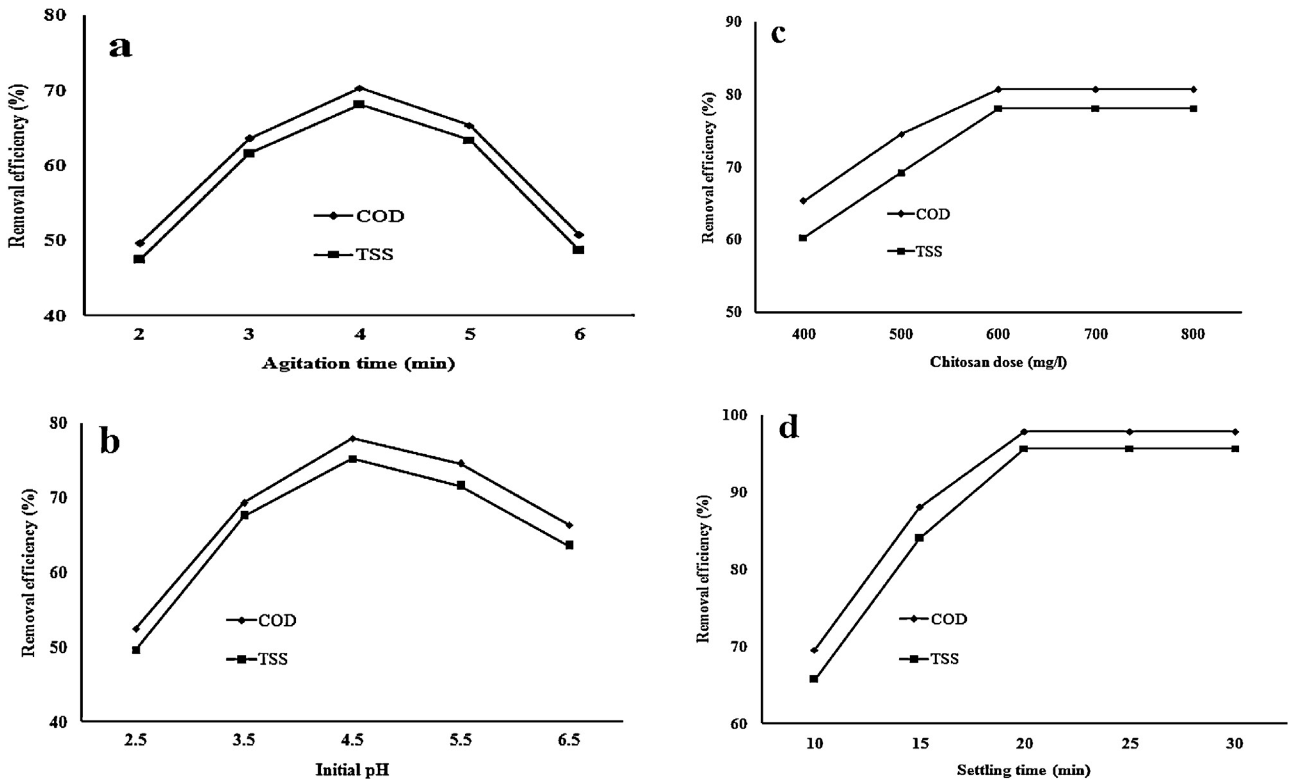

Fig. 1. Effect of process variables on the percentage removal of COD and TSS.

wastewater using chitosan as an adsorbent. Hence the primary objective of the present study has been made to investigate and optimize the individual and interactive effect of process variables such as agitation time, initial pH, chitosan dose and settling time on the percentage removal of chemical oxygen demand (COD) and total suspended solids (TSS) from rice mill wastewater. The mechanism of treatment process was analyzed and confirmed by using Fourier transform infrared (FT-IR) spectroscopy. Moreover, response surface methodology (RSM) coupled with Box–Behnken response surface design (BBD) was used to develop mathematical models in order to predict the response values under the experimental range used in this study. Response surface methodology is a collection of statistical techniques commonly used to understand the performance of complex systems and modelling the various biochemical process. This technique also be used to evaluate the relative significance of several affecting factors even in the presence of complex interactions between the independent variables (Prakash Maran, Sivakumar, Sridhar, & Thirugnanasambandham, 2013).

2. Materials and methods

2.1. Raw wastewater and chemicals

The rice mill wastewater used in this study was collected from rice processing industry near Erode, TamilNadu, India and were stored at 4 °C prior to the experiments in order to avoid changes in the physico-chemical properties of wastewater. The initial conditions of rice mill wastewater was determined and found to be: initial pH of 4.98, COD of 2200 mg/l and TSS of 768 mg/l respectively, which indicated the higher content of organic and inorganic pollutants. Hydrochloric acid (HCl) was purchased from Sigma Chemicals, Mumbai. Sodium hydroxide (NaOH) was supplied by Merck Chemicals, Chennai. All the chemicals used in this study were of analytical grade. Chitosan (RM9358-100G) was purchased from HiMedia Laboratories, Mumbai.

2.2. Experimental method

A conventional batch studies were carried out under different working conditions such as agitation time (2–6 min), initial pH (2.5–6.5) and chitosan dose (400–800 mg/l) in 250 ml conical flask containing 100 ml of composite rice mill wastewater. Incubator shaker equipment (SLM-1NC-OS-250) was used to agitate the sample containing conical flask and were allowed to settle at different settling time (10–30 min). Then the sample was centrifuged at 6500 rpm for 15 min (Remi R-24 Centrifuge, India) and the supernatant was filtered through a 0.45 μm filter papers and the filtrate was collected for the analysis of chemical oxygen demand (COD) and total suspended solids (TSS). All the experiments were

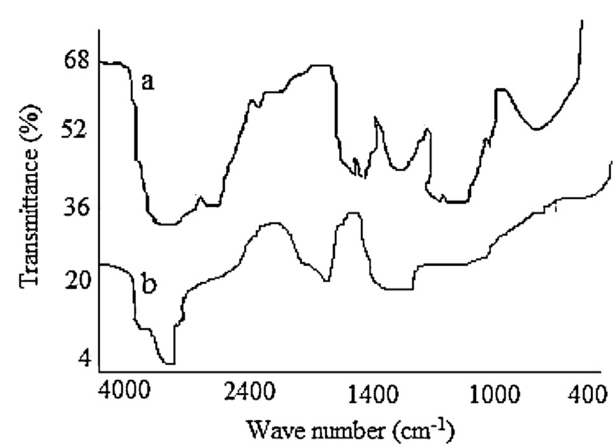

Fig. 2. FT-IR spectra of raw chitosan (a) and after treatment process (b).

Table 1
Ranges of independent variables and their levels.

Variable (unit)	Levels		
	−1	0	1
Agitation time (min, A)	2	4	6
Initial pH (B)	2.5	4.5	6.5
Chitosan dose (mg/l, C)	400	600	800
Settling time (min, D)	10	20	30

performed in triplicates and the results were presented as the mean value of the triplicates.

2.3. Analytical method

The pH of the sample was measured with the help of pH meter (Elico LI120, India), and pH adjustment was done using 0.1 N HCl or 0.1 N NaOH (Sridhar, Sivakumar, Prince Immanuel, & Prakash Maran, 2011). The COD and TSS analysis were carried out by according to the procedures described by American Public Health Association (APHA). The removal efficiency (RE) of COD and TSS was calculated by using the following equation (Sridhar et al., 2011):

$$RE = \left(\frac{c_0 - c_e}{c_0}\right) \times 100 \tag{1}$$

where c_0 and c_e is the initial and final concentrations of COD and TSS respectively.

2.4. Fourier transform infrared (FT-IR) spectroscopy analysis

FT-IR spectra of chitosan (before and after adsorption process) was recorded in the range of 4000–400 cm^{-1} with the help of potassium bromide (KBr) pellets using a FT-IR spectrometer (Instrument model RX_1, India).

3. Results and discussion

3.1. Effect of agitation time

The agitation time is one of the important parameter, which influences the performance of the adsorption process significantly. To examine its effect, the experiments were carried out at different agitation time (2–6 min), while keeping the other working parameters (initial pH of 3.5, chitosan dose of 500 mg/l and settling time of 15 min) constant and the results were exhibited in Fig. 1a. The mechanical agitation caused more fibrillation and increases the exposed specific surface area for adsorption process by creating collisions between the chitosan and organic particles present in the effluent (Dzul Erosa, Saucedo Medina, Navarro Mendoza, Avila Rodriguez, & Guibal, 2001), which could increase the removal efficiency of COD and TSS, while increasing the agitation time from 2 to 4 min. Further increase in agitation time could destabilize interaction (bond) between chitosan and organic particles and decreased the removal efficiency. So, agitation time of 4 min is considered as the optimum time for the higher percentage removal of COD (70.25%) and TSS (68.02%).

3.2. Effect of initial pH

The surface charge of the chitosan effectively varies with varying the pH of the wastewater. So, the experiments were performed at different initial pH (2.5–6.5) while keeping the agitation time of 4 min, chitosan dosage of 500 mg/l, settling time of 15 min as constant and the observation is depicted in Fig. 1b. From Fig. 1b, it was observed that the percentage of COD and TSS removal increased with increasing initial pH up to 4.5. This is mainly due to the formation $—NH_3^+$ ions in acidic conditions, which could adsorb the negatively charged organic matters present in the wastewater significantly (Guzman, Saucedo, Revilla, Navarro, & Guibal, 2003) and increased the removal efficiency of COD (77.98%) and

Table 2
BBD and their experimental results.

Run	Agitation time (A)	Initial pH (B)	Chitosan dose (C)	Settling time (D)	COD removal (%)	TSS removal (%)
1	4.00	2.5	800	20	76.54	74.38
2	4.00	2.5	600	10	36.54	34.28
3	2.00	2.5	600	20	59.54	57.62
4	2.00	6.5	600	20	60.54	58.62
5	2.00	4.5	600	10	30.54	28.34
6	6.00	6.5	600	20	55.64	53.02
7	4.00	6.5	800	20	65.54	63.56
8	4.00	4.5	400	10	44.32	42.34
9	6.00	4.5	800	20	70.54	68.48
10	4.00	4.5	600	20	95.84	93.86
11	4.00	6.5	400	20	80.54	78.56
12	4.00	2.5	600	30	73.24	71.26
13	4.00	4.5	800	10	45.54	43.28
14	4.00	4.5	600	20	95.84	93.86
15	6.00	4.5	600	30	68.54	66.54
16	4.00	4.5	600	20	95.84	93.86
17	6.00	4.5	400	20	70.54	68.34
18	4.00	4.5	800	30	89.84	87.86
19	4.00	4.5	600	20	95.84	93.86
20	4.00	6.5	600	10	38.54	36.48
21	2.00	4.5	400	20	68.78	66.38
22	6.00	2.5	600	20	45.64	43.28
23	6.00	4.5	600	10	40.54	38.56
24	4.00	2.5	400	20	58.54	56.38
25	4.00	6.5	600	30	82.64	80.66
26	4.00	4.5	400	30	95.98	94.28
27	2.00	4.5	600	30	84.54	82.54
28	4.00	4.5	600	20	95.84	93.86
29	2.00	4.5	800	20	75.34	73.24

Table 3
Multi regression analysis of responses.

Source	Sum of squares	Degree of freedom	Mean square	F-Value	Prob > F	Remarks
Sequential model sum of squares for COD removal						
Mean	137,616.73	1.00	137,616.73			
Linear	5739.44	4.00	1434.86	5.61	0.0025	
2FI	499.52	6.00	83.25	0.27	0.9459	
Quadratic	5531.43	4.00	1382.86	174.67	<0.0001	Suggested
Cubic	87.89	8.00	10.99	2.87	0.1078	Aliased
Residual	22.95	6.00	3.83			
Total	149,497.96	29.00	5155.10			
Sequential model sum of squares for TSS removal						
Mean	129,455.73	1.00	129,455.73			
Linear	5791.77	4.00	1447.94	5.62	0.0025	
2FI	501.04	6.00	83.51	0.26	0.9465	
Quadratic	5566.43	4.00	1391.61	160.50	<0.0001	Suggested
Cubic	99.22	8.00	12.40	3.36	0.0787	Aliased
Residual	22.16	6.00	3.69			
Total	141,436.37	29.00	4877.12			

Source	Std. Dev.	R^2	Adjusted R^2	Predicted R^2	PRESS	Remarks
Model summary statistics for COD removal						
Linear	15.9971	0.4831	0.3969	0.3411	7828.91	
2FI	17.7048	0.5251	0.2613	0.1104	10,570.00	
Quadratic	2.8137	0.9907	0.9813	0.9462	638.72	Suggested
Cubic	1.9560	0.9981	0.9910	0.7218	3305.49	Aliased
Model summary statistics for TSS removal						
Linear	16.0583	0.4834	0.3973	0.3414	7890.16	
2FI	17.7761	0.5252	0.2615	0.1099	10,664.40	
Quadratic	2.9445	0.9899	0.9797	0.9416	699.47	Suggested
Cubic	1.9220	0.9981	0.9914	0.7336	3191.71	Aliased

TSS (75.24%) up to the pH of 4.5. After that, solubility of chitosan was decreased, while increasing the pH towards the basic condition and leads to decrease in removal efficiencies of COD and TSS. From the results, initial pH of 4.5 and agitation time of 4 min were determined as an optimum conditions for further experiments.

3.3. Effect of chitosan dose

The chitosan dose is a key factor to determine the percentage removal of COD and TSS in rice mill wastewater. In order to evaluate its effect, experiments were performed at various ranges of chitosan dose (400–800 mg/l), while other parameters such as agitation time (4 min), initial pH (4.5) and settling time (15 min) were kept at constant and the results are illustrated in Fig. 1c. The percentage removal of COD and TSS was increased with increasing chitosan dose up to 600 mg/l, further increase in chitosan dose (600–800 mg/l) did not show any significant effect. Higher dosage of chitosan (up to 600 mg/l) can effectively react with the organic and inorganic pollutants present in the wastewater, due to the availability of more of number reaction sites, which can increase the removal efficiency of COD (80.68%) and TSS (78.08%). Beyond this dosage, the adsorption process attains an equilibrium stage with the charged molecules of the wastewater (Riaño, Molinuevo, & Garcia-Gonzalez, 2012) and did not improve the efficiency of adsorption process. Hence, initial pH of 4.5 agitation time of 4 min and chitosan dose of 600 mg/l were chosen as optimal conditions for upcoming experiments.

Table 4
ANOVA table for responses.

Source	COD removal				TSS removal			
	Sum of squares	Mean square	F-Value	p-Value	Sum of squares	Mean square	F-Value	p-Value
Model	11,770.39	840.74	106.19	0.0001	11,859.25	847.09	97.70	0.0001
A	64.59	64.59	8.16	0.0127	67.78	67.78	7.82	0.0143
B	93.05	93.05	11.75	0.0041	94.72	94.72	10.92	0.0052
C	1.79	1.79	0.23	0.6422	1.69	1.69	0.20	0.6652
D	5577.87	5577.87	704.53	0.0001	5625.39	5625.39	648.81	0.0001
AB	20.25	20.25	2.56	0.1321	19.10	19.10	2.20	0.1600
AC	10.74	10.74	1.36	0.2635	11.27	11.27	1.30	0.2733
AD	169.05	169.05	21.35	0.0004	171.92	171.92	19.83	0.0005
BC	272.25	272.25	34.39	0.0001	272.25	272.25	31.40	0.0001
BD	13.69	13.69	1.73	0.2097	12.96	12.96	1.49	0.2417
CD	13.54	13.54	1.71	0.2120	13.54	13.54	1.56	0.2319
A^2	2555.76	2555.76	322.81	0.0001	2591.06	2591.06	298.84	0.0001
B^2	2468.17	2468.17	311.75	0.0001	2490.79	2490.79	287.28	0.0001
C^2	228.61	228.61	28.88	0.0001	231.44	231.44	26.69	0.0001
D^2	2553.68	2553.68	322.55	0.0001	2544.46	2544.46	293.47	0.0001
Residual	110.84	7.92			121.38	8.67		
Lack of fit	110.84	11.08			121.38	12.14		
Pure error	0.00	0.00			0.00	0.00		

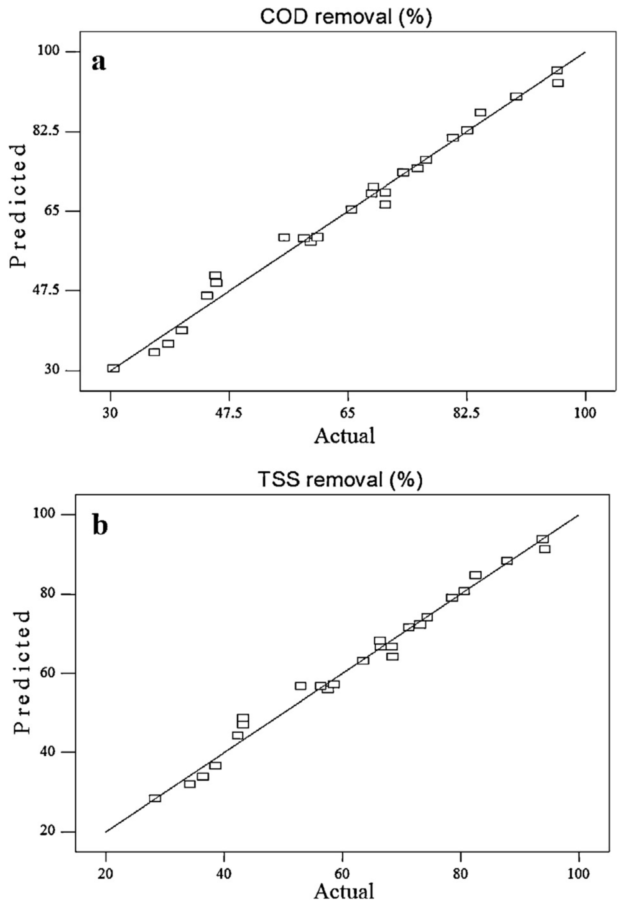

Fig. 3. Adequacy of model tested for responses.

3.4. Effect of settling time

In the adsorption process, the settling time is an important factor which can influence the overall removal efficiencies of COD and TSS in wastewater treatment. To determine the effective settling time, experiments were done at different settling time of 10–30 min while other parameters such as agitation time (4 min), initial pH (4.5), and chitosan dose (600 mg/l) were kept as constant. The results indicate that, the percentage removal of COD and TSS was increased with increasing settling time up to 20 min and then did not show any significant effect. Aggregation of flocs (formed between chitosan and colloidal particle in the wastewater) increases up to 20 min, in which most of the flocs are settle down, thus removal efficiencies increased during adsorption process (Abu Hassan, Pei Li, & Zainon Noor, 2009). Finally, the percentage removal efficiency of COD and TSS under optimum settling time (20 min) were found to be 97.84 and 95.34% respectively.

3.5. FT-IR analysis

FT-IR spectra of chitosan (before and after adsorption process) was recorded in the range of 4000–400 cm^{-1} (Fig. 2) in order to study the mechanism of adsorption process. From Fig. 2a, it was found that the broad band at 3402 cm^{-1} and the peak at 1371 cm^{-1} indicating the presence of $-NH_2$ and O–H groups on the raw chitosan (Anirudhan & Radhakrishnan, 2009). Transmittance shifted from 3402 cm^{-1} (Fig. 2a) to 3118 cm^{-1} (Fig. 2b) and the formation of new peak at 1655 cm^{-1} (Fig. 2b) clearly indicates the interaction of chitosan with the organic pollutants present in the wastewater. This is explained by the fact that the presence of $-NH_2$ and O–H groups in the raw chitosan can act as the reaction sites for the adsorption process (Pawlak & Mucha, 2003).

3.6. Process modelling and statistical analysis

In this study, Box–Behnken response surface experimental design (BBD) with four factors at three levels was employed to investigate the interactive effect of process variables (agitation time (2–6 min), initial pH (2.5–6.5), chitosan dose (400–800 mg/l) and settling time (10–30 min)) on the removal efficiencies of COD and TSS. In addition, this experimental design was used to develop mathematical models in order to predict the responses. Process variables (coded and un-coded) and their ranges were shown in Table 1. A total number of 29 experiments (including five centre points) were established based on the method described by Prakash Maran, Sivakumar, Sridhar, and Thirugnanasambandham (2013) and the results are stated in Table 2. The results were analyzed by multiple regression analysis, Pareto analysis of variance (ANOVA), *F*-test, coefficient of determination (R^2), adjusted coefficient of determination (adj-R^2) and predicted coefficient of determination (pre-R^2). All the statistical analyses were done with the help of Stat ease Design Expert 8.0.7.1 statistical software package (Stat-Ease Inc., Minneapolis, USA). After analysis of the experimental data, second order polynomial model (SOPM) was developed and general form of SOPM was given in elsewhere (Prakash Maran, Manikandan, Thirugnanasambandham, Vigna Nivetha, & Dinesh, 2013). The developed SOPM were used for the construction of contour response surface plots to examine the relationships between independent and dependent variables.

3.6.1. Model development and analysis

The experimental data was analyzed by two different tests namely the sequential model sum of squares and model summary statistics in order to obtain suitable regression models to predict the responses and the results are listed in Table 3. From Table 3, it was observed that, quadratic model shows high R^2, adjusted-R^2, predicted-R^2, *F*-value and low *p*-value, when compared with other models (linear and interactive (2FI)). Cubic model was found to be aliased. Therefore the quadratic model is selected to portray the effects of process variables on the responses.

From the experimental data, two empirical models were developed to understand the interactive correlation between the responses and process variables (Prakash Maran, Sivakumar, Sridhar, & Prince Immanuel, 2013) and final developed mathematical models obtained in terms of coded factors are given below

$$\begin{aligned}\text{COD removal}\quad(\%) = {} & 95.84 - 2.32A + 2.78B + 0.39C + 21.56D \\ & + 2.25AB - 1.64AC - 6.50AD - 8.25BC \\ & + 1.85BD - 1.84CD - 19.85A^2 - 19.51B^2 \\ & - 5.94C^2 - 19.84D^2 \end{aligned} \tag{2}$$

$$\begin{aligned}\text{TSS removal}\quad(\%) = {} & 93.86 - 2.38A + 2.81B + 0.38C + 21.65D \\ & + 2.18AB - 1.68AC - 6.56AD - 8.25BC \\ & + 1.80BD - 1.84CD - 19.99A^2 - 19.60B^2 \\ & - 5.97C^2 - 19.81D^2 \end{aligned} \tag{3}$$

Moreover, Pareto analysis of variance (ANOVA) was used to analyze the experimental data and significance of the developed model equations were evaluated by their corresponding *F*-value and *p*-values (Table 4). The higher model *F*-values and lower *p*-values

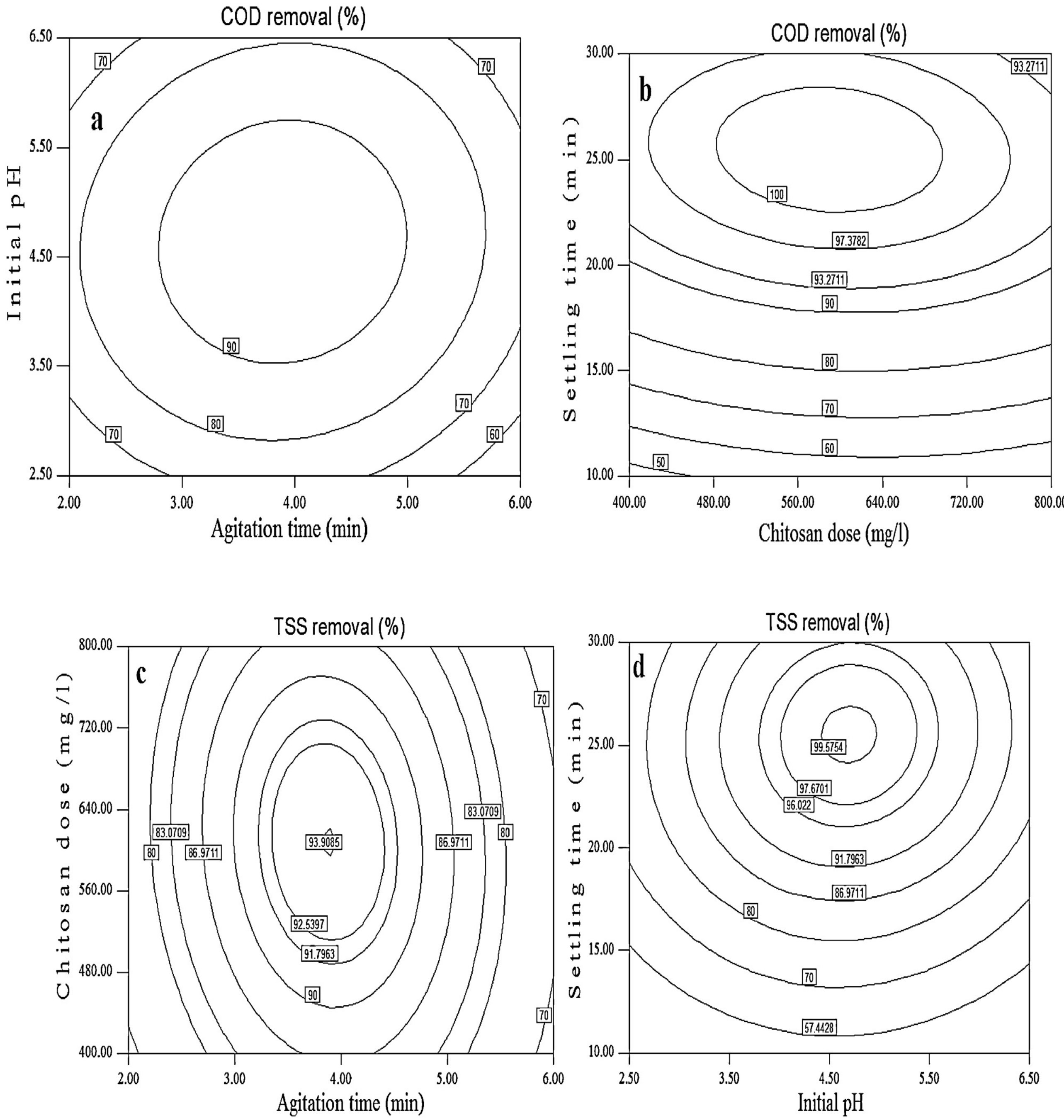

Fig. 4. Response surface contour plots for responses.

of the both responses (COD removal and TSS removal) demonstrated that, the developed model was highly significant. From these results, it was concluded that, the developed mathematical models has the ability to describe the adsorption process very robustly (Prakash Maran, Sivakumar, Sridhar, & Prince Immanuel, 2013).

3.6.2. Adequacy of developed models

Generally, it is very essential to check the capability of the developed mathematical model to represent and describe the present treatment process conditions. In this perspective, construction of diagnostic plot such as predicted versus actual plot (Fig. 3) was used to evaluate the model suitability and find out the relationship between predicted and experimental values (Sridhar, Sivakumar, Prince Immanuel, and Prakash Maran (2012)). The data points lies very close to the diagonal line and it depicts a good relationship between experimental and predicted data (Prakash Maran, Sivakumar, Thirugnanasambandham, & Sridhar, 2013).

3.6.3. Response surface analysis of process variables on responses

From the developed models, response surface contour plots were constructed to evaluate the interactive effects of the independent variables on the responses and it was given in Fig. 4. From Fig. 4, it was observed that all the combined process variables show significant effect on the percentage removal of COD and TSS in adsorption process as well as the trends obtained in the contour plots are closely agreed with Fig. 1.

4. Conclusions

In this present study, removal efficiencies of chemical oxygen demand (COD) and total suspended solids (TSS) from rice mill wastewater was examined under different process conditions such as agitation time (2–6 min), initial pH (2.5–6.5), chitosan dose (400–800 mg/l) and settling time (10–30 min). Under the optimum conditions such as agitation time of 4 min, initial pH of 4.5, chitosan dose of 600 mg/l and settling time of 20 min shows the removal efficiency of >95% for COD and TSS respectively. Process mechanism was identified by using FT-IR spectrophotometry. Process modelling of the present study was done by response surface methodology (RSM) coupled Box–Behnken response surface design (BBD) with four factors at three levels. Second order polynomial models were developed from the experimental data in order to predict the responses with high correlation coefficient values. This results exhibited that, chitosan is found to be a suitable adsorbent to treat rice mill wastewater in terms of high removal efficiency of pollutants as well as protection of ecosystem by eliminating the creation of secondary pollutants.

References

Abu Hassan, M. A., Pei Li, T., & Zainon Noor, Z. (2009). Coagulation and flocculation treatment of wastewater in textile industry using chitosan. *Journal of Chemical and Natural Resources Engineering*, *4*, 43–53.

Amit, B., & Mika, S. (2009). Applications of chitin and chitosan derivatives for the detoxification of water and wastewater—A short review. *Advances in Colloid and Interface Science*, *152*, 26–38.

Anirudhan, T. S., & Radhakrishnan, P. G. (2009). Kinetics, thermodynamics and surface heterogeneity assessment of uranium (VI) adsorption onto cation exchange resin derived from a lignocellulosic residue. *Applied Surface Science*, *255*, 4983–4991.

APHA. (1995). *Standard methods for the examination of water and wastewater* (17th ed.,). WA, USA: American Public Health Association.

Dzul Erosa, M. S., Saucedo Medina, T. I., Navarro Mendoza, R., Avila Rodriguez, M., & Guibal, E. (2001). Cadmium sorption on chitosan sorbents: Kinetic and equilibrium studies. *Hydrometallurgy*, *61*, 157–167.

Gálvez, J. M., Gomez, M. A., Hontoria, E., & Gonzalez-Lopez, J. (2003). Influence of hydraulic loading and air flow rate on urban wastewater nitrogen removal with a submerged fixed-film reactor. *Journal of Hazardous Materials*, *101*, 219–229.

Grégorio, C., & Pierre-Marie, B. (2008). Application of chitosan, a natural aminopolysaccharide, for dye removal from aqueous solutions by adsorption processes using batch studies: A review of recent literature. *Progress in Polymer Science*, *33*, 399–447.

Guzman, J., Saucedo, I., Revilla, J., Navarro, R., & Guibal, E. (2003). Copper sorption by chitosan in the presence of citrate ions: Influence of metal speciation on sorption mechanism and uptake capacities. *International Journal of Biological Macromolecules*, *33*, 57–65.

Kalyuzhnyi, S., Gladchenko, M., Starostina, E., Shcherbakov, S., & Versprille, A. (2005). Combined biological and physico-chemical treatment of baker's yeast wastewater. *Water Science and Technology*, *52*, 175–181.

Manaswini, B., Partha, S. J., Tanaji, T. M., & Ghangrekar, M. M. (2010). Rice mill wastewater treatment in microbial fuel cells fabricated using proton exchange membrane and earthen pot at different pH. *Bioelectrochemistry*, *79*, 228–233.

Manogari, R., Daniel, D., & Krastanov, A. (2008). Biodegradation of rice mill effluent by immobilised *Pseudomonas* sp. cells. *Ecological Engineering and Environment Protection*, *1*, 30–35.

Pawlak, A., & Mucha, M. (2003). Thermogravimetric and FTIR studies of chitosan blends. *Thermochima Acta*, *396*, 153–166.

Pooja Vaishnav, S., & Anupam Bala, S. (2012). Treatment of rice mill effluent for pollution control by electrocoagulation. *Journal of Chemical, Biological and Physical Sciences*, *2*, 480–483.

Prakash Maran, J., Manikandan, S., Thirugnanasambandham, K., Vigna Nivetha, C., & Dinesh, R. (2013). Box–Behnken design based statistical modeling for ultrasound-assisted extraction of corn silk polysaccharide. *Carbohydrate Polymers*, *92*, 604–611.

Prakash Maran, J., Sivakumar, V., Sridhar, R., & Prince Immanuel, V. (2013). Development of model for mechanical properties of tapioca starch based edible films. *Industrial Crops and Products*, *42*, 159–168.

Prakash Maran, J., Sivakumar, V., Sridhar, R., & Thirugnanasambandham, K. (2013). Development of model for barrier and optical properties of tapioca starch based edible films. *Carbohydrate Polymers*, *92*, 1335–1347.

Prakash Maran, J., Sivakumar, V., Thirugnanasambandham, K., & Sridhar, R. (2013). Model development and process optimization for solvent extraction of polyphenols from red grapes using Box–Behnken design. *Preparative Biochemistry and Biotechnology*, http://dx.doi.org/10.1080/10826068.2013.791629

Rajesh, G., Bandyopadhyay, M., & Das, D. (1999). Some studies on UASB bioreactors for the stabilization of low strength industrial effluents. *Bioprocess Engineering*, *21*, 113–116.

Ravi Kumar, N. V. (2000). A review of chitin and chitosan applications. *Reactive and Functional Polymers*, *46*, 1–27.

Riaño, B., Molinuevo, B., & Garcia-Gonzalez, M. C. (2012). Optimization of chitosan flocculation for microalgal-bacterial biomass harvesting via response surface methodology. *Ecological Engineering*, *38*, 110–113.

Rinaudo, M. (2006). Chitin and chitosan: Properties and applications. *Progress in Polymer Science*, *31*, 603–632.

Sridhar, R., Sivakumar, V., Prince Immanuel, V., & Prakash Maran, J. (2011). Treatment of pulp and paper industry bleaching effluent by electrocoagulation process. *Journal of Hazardous Materials*, *186*, 1495–1502.

Sridhar, R., Sivakumar, V., Prince Immanuel, V., & Prakash Maran, J. (2012). Development of model for treatment of pulp and paper industry bleaching effluent using response surface methodology. *Environmental Progress & Sustainable Energy*, *31*, 558–565.

Synowiecki, J., & Al-Khateeb, N. A. (2003). Production, properties, and some new applications of chitin and its derivatives. *Critical Review of Food Science and Nutrition*, *43*, 145–171.