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Study of the efficiency of bio-filter and activated sludge (BF/AS) combined process in phenol removal from aqueous solution: determination of removing model according to response surface methodology (RSM)

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ABSTRACT

Since most industrial wastewaters contain phenol compounds, achieving eco-friendly and economic processes for filtration of such wastewaters is one of the concerns of environmental researchers. Thus, present study aims to determine the efficiency of bio-filter and activated sludge (BF/AS) combined process in phenol removal from aqueous solution. In this experimental study, BF/AS reactor in a pilot scale with continuous flow and useful volume of 24 L, made of Plexiglas was utilized. Effect of dissolved oxygen concentration, hydraulic residence time, mixed liquor suspended solids (MLSS) and glucose concentration on phenol removing efficiency were evaluated. In order to determine the phenol removal model and optimum removal conditions using central composite design in response surface methodology environment, the reactor was studied in separate runs according to software design. Accuracy of results was evaluated in MLSS concentrations of 2,250 mg/L, glucose of 250 mg/L and dissolved oxygen of 3 mg/L and quadratic model (R^2 = 0.99) well fitted to the results. With regard to high yield of BF/AS biological system in decreasing phenol content from wastewater, the system under study could be used widely in refining the toxic wastewaters of industries given the appropriate operation conditions.

Keywords: BF/AS; Biodegradation; Phenol; RSM; Wastewater

1. Introduction

Phenol is among aromatic hydrocarbons which is flammable and widely present in water, oil and some of the organic solvents in dissolved form and has sweet, medicinal or pitch-like odor [1]. Phenol in wastewater is resulted from various industries including petrochemical, oil refining, resin production, pharmacy, dry synthesis units and coking industry [2,3]. According to reports, phenol concentration in these industries varied from 300 to 400 mg/L [4]. Presence of phenol in water sources leads to declining water quality, organisms and also inhibition of usual activities in biological systems. Also, the phenol enters into food chain and threatens public health [5]. Phenol is acidic and water soluble, has high corrosive property and causes nervous poisoning and digestive problems [6,7]. Also, due to creating mutagen, teratogen and carcinogenesis effects, phenol was recognized as priority pollutant by United States Environmental Protection Agency, so among 129 priority pollutants, phenol was ranked the 11th and has 1 m/L discharge limit in refined wastewaters [8,9]. World Health Organization (WHO), defined 0.001 mg/L phenol concentration as a limit for potable water [10]. Thus,

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removing toxic phenol from wastewater prior to discharging to the environment is essential. There are various methods for phenol removing from aqueous solutions including chemical degradation [11], electrochemical degradation [12], biological degradation [13] and absorption [14].

Biological degradation is among the eco-friendly methods and was preferred to phenol removal from aqueous solutions due to inexpensive and simple technology and high efficiency [14]. Recently, uses of combined biological systems attract great attention [15]. One of these methods is use of bio-filter/activated sludge (BF/AS) system. In recent years, bio-filters as a promising biological process in dangerous pollutants removal from wastewaters have received great attention. In bio-filtration, microorganisms were fixed on a porous substrate in order to remove pollutants present in wastewater. In fact bio-filter technique is a combination of water clarification filters and physical and biological purification processes through filtration of suspended materials which could be aerobic or anaerobic depending on the refining purpose. It should be noted that microorganism's growth in bio-filter system is in both fixed on bio-film and suspended in aqueous phase around the substrate particles. In order to assure a high level of connectivity areas and supplying additional nutrients, filter substrate was composed of relatively neutral material [16]. Some of advantages of submerged biological systems include high density, little space, ability to compatibility with varying wastewater flows and need to small hydraulic residence time. Due to abovementioned advantages, activated sludge systems were replaced by bio-filters or used in combination with utilizing in modern sewage treatment plants in large urban areas [17]. Present work aims at dealing with the potential of combination of two conventional biotechnology processes (AS/BF) for phenol removal from aqueous solution in terms of abatement efficiency. So, this work in the first step aims to evaluate the effectiveness of integrated biological wastewater treatment (BF/AS) in the presence of phenol and, in the second step, to evaluate the effect of different levels of concentration of glucose on removal efficiency of phenol, and finally, the effects of dissolved oxygen concentration, hydraulic residence time, MLSS and glucose concentration on phenol removing efficiency.

2. Materials and methods

Present study was an experimental type and was conducted in a pilot scale on synthetic wastewater containing phenol.

2.1. Chemicals and reagents

Phenol and all chemicals and reagents required for the experiments were purchased from Merck Co. (Deutschland) with purity degree of >99%. The water from urban potable water network was used for preparation of synthetic wastewater.

2.2. Pilot characteristics

Aerobic bio-reactor consists of three units: (1) feed tank, (2) bio-filter reactor and (3) the settling tank. Bio-filter reactor was made from Plexiglas with useful volume of 24 L (Fig. 1). Inside the bio-filter reactor was equipped with diffuser aerators, and reticular plastic discs were fixed on substrate as biofilm holder. Hydraulic flow type in the system under study was continuous and upward. In order to inject solution into the system, two two-way diaphragm pumps with discharge capacity of 0.1–2 mL/s where utilized. A settling basin was made of Plexiglas with 60 × 30 × 20 cm dimensions (height × length × width).

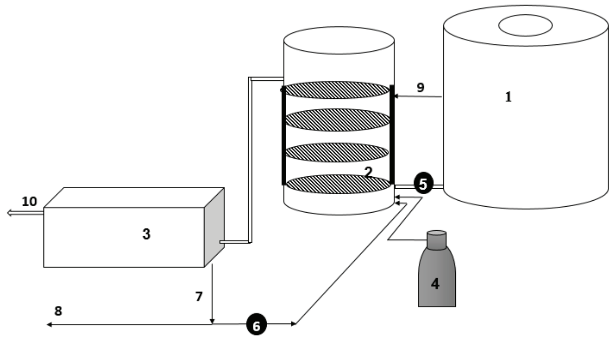

Fig. 1. Schematic of studied BF/AS combined system.

2.3. Adaptation

Initial source of utilized microorganisms was from urban sludge treatment plant which initially proliferated by addition of dry milk and then gradually phenol concentration increased and milk reduced during 3 months complete adaptation with phenol.

2.4. Effect of separate operating parameters in phenol removal efficiency

The system under study was operated in laboratory environment with 18°C–28°C. The effects of dissolved oxygen concentration (0.5–3.7 mg/L), hydraulic residence time (5.5–14 h), MLSS (1,500–3,000 mg/L) and glucose concentration (0–500 mg/L) on phenol removing efficiency at 500 mg/L concentration were evaluated. In order to measure the phenol concentration volatilizing from the top of the bio-reactor through aeration, a suction pump was used, and total gasses and vapors from reactors passed through the impinge containing NaOH 0.1 N solution. In all steps, phenol removing efficiency until reaching <5% change was measured to assure that microorganisms were completely adapted to these new conditions. For data reliability, sampling was conducted in three replications and the results were presented as average. It should be noted that sampling, samples maintenance and experiments were conducted according to water and wastewater standard methods [18]. Finally, data were analyzed using Excel software.

2.5. Experimental design and response surface methodology analysis for determination of interaction effects of operated parameters in phenol removal

In present study, in order to determine phenol removing model and optimum operation conditions, central composite design (CCD) test in the environment (response surface methodology [RSM] analysis) was used in designing Expert Software. Independent variables in this stage included dissolved oxygen concentration (*C*; 1, 2 and 3 mg/L), MLSS (*A*; 1,500, 2,250 and 3,000 mg/L) and glucose concentration (*B*; 0, 250 and 500 mg/L) which were presented in three levels including minimum (–1), average (0) and maximum (+1). According to interaction effects of factors in three levels with three factors, total number of experiments in RSM designing method was 20 of which five were used to test the accuracy of repeated experiments (dissolved oxygen concentration: 2 mg/L; MLSS: 2,250 mg/L; glucose concentration: 250 mg/L). In this stage, bio-reactor was operated for 20 d by designing the RSM experiments.

3. Results and discussion

3.1. Impact of operation variables on BF/AS bio-reactor efficiency

In a part of the study, in each stage, only the effects of one parameter on phenol removal efficiency was studied by constant taking of other parameters which are presented as follows.

3.1.1. Effect of hydraulic residence time at different glucose concentrations on system efficiency

In present study, phenol bio-degradation, glucose consumption and cell growth were evaluated in different glucose concentrations from 0 to 500 mg/L at different residence times (Figs. 2–4). Results showed higher degradability of glucose than phenol. Phenol removing efficiency at 0, 50, 125, 250 and 500 mg/L glucose concentrations was obtained as 79.5%, 89.6%, 43.6%, 38.7% and 35%, respectively. By increasing glucose concentration, phenol removal efficiency decreased. For example, by increasing glucose at 250 mg/L concentration at the same hydraulic residence time (6 h), efficiency decreased by about 4%. This decreasing trend reached 2.85% at 8 h residence time. By addition of glucose by initial concentration of 250 mg/L, the required time for complete removal (100%) increased about 6 h, that is, from 8 to 14, while in order to reach 98% system efficiency, it increased about 1.5 h. This means that reaching the final stages of complete removal of phenol is progressed slowly in a way that 1% increase is obtained in removal efficiency without glucose in 1 h, while 1% increase in removal efficiency in the presence of 250 mg/L glucose requires about 5 h. This result is in consistent with the results of Dargahi et al. [19,20] and Shokoohi et al. [21].

Usually glucose is consumed easily while phenol is strongly toxic and stable, so glucose consumption rate is higher than that of phenol [22]. Therefore, current biological system requires 14 h for complete removal of phenol. Since glucose is bio-degradable and phenol is degraded slowly, so probably phenol is removed later. According to Fig. 1, phenol degradation at glucose concentration up to 50 mg/L is increased. In this concentration, phenol degrading microorganisms are dominant in the system. On the other hand, coliforms consume only glucose and are unable to degrade phenol [23]. Our results are in contrast with those of Pishgar et al. [22]. It was found that glucose concentration in the range of 25–1,000 mg/L and 500–3,000 mg/L

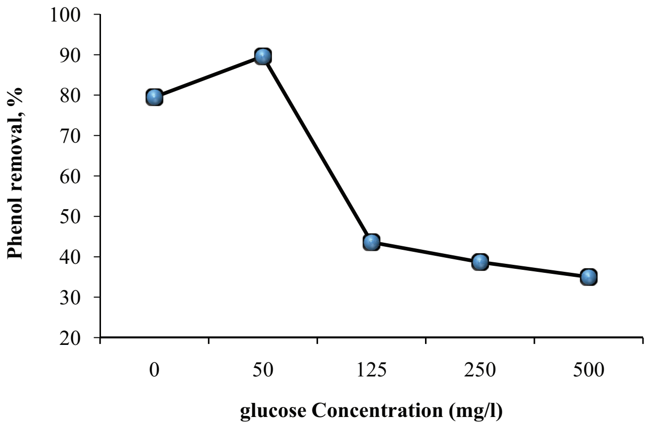

Fig. 2. Phenol removal efficiency at various glucose concentrations for phenol removal from aqueous solutions (initial phenol concentration of 500 mg/L).

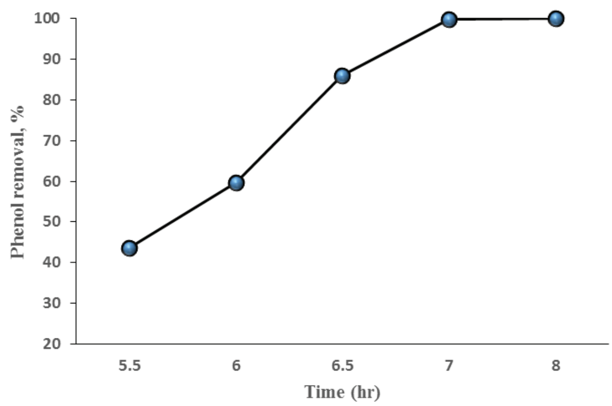

Fig. 3. Phenol removal efficiency at various residence times for phenol removal from aqueous solutions (initial phenol concentration of 500 mg/L).

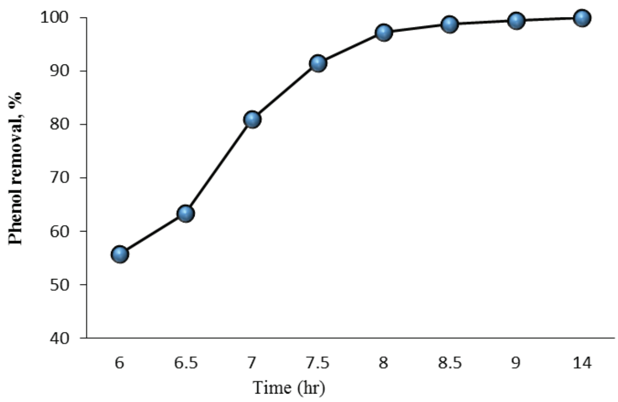

Fig. 4. Phenol removal efficiency at various residence times for phenol removal from aqueous solutions (initial phenol concentration of 500 mg/L, and glucose increasing concentration of 250 mg/L).

had positive impact on phenol removal efficiency. Similar results indicated low concentrations of glucose, and an increase in phenol bio-degradation rate was also reported by Hoodaji et al. [24]. In present study, the resulted trend up to 6.5 h hydraulic residence time was in consistent with Pishgar et al. [22] findings and afterward we observed negative impact of glucose on bio-reactor efficiency. So, it can be said that in order to reach higher removal efficiency, keeping the glucose content low in wastewaters containing phenol is essential. Chen and Hao [25] showed that high amounts of glucose strongly inhibited aerobic removal of toxic materials. In studying the BF/AS performance under steady-state conditions at various hydraulic residence times, it was recognized that obtaining 100% phenol removal at 8 h residence time is possible and by increasing hydraulic residence time, removal efficiency increased so that 43.6% removal efficiency at 5.5 h reached 99.8% at 7 h and increased to 100% at 8 h (Table 1).

3.1.2. Effect of MLSS concentration on system efficiency

Results in Fig. 5 show that phenol removal efficiency has direct relation with MLSS concentration. According to results, the microbial mixture has the potential of removing phenol from wastewater. Maximum phenol removal (99.8%) was obtained in 3,000 mg/L bio-mass. Ryu et al. [26] showed that increasing bio-mass up to 3,000 mg/L had restrictive impact on bio-filter efficiency, which is in contrast to our results. Studying the effect of MLSS concentration on system efficiency shows that in BF/AS system, phenol removal efficiency is between 48.57% and 99.8%. It is clear that by increasing MLSS content, activity and reproduction rate of biological bulk increased and biological system could tolerate shock resulted from increasing load at final stages, so the system efficiency increased [27] and outgoing phenol concentration decreased by increasing bio-mass. Marrot et al. [15] found that only activated sludge process was unable to afford high phenol contents. So phenol concentration of 350 mg/L had inhibitory impact on system representing toxic traits of phenol. This issue justifies the use of bio-filter in combination with activated sludge in present study. The highest phenol removal (99.8%) was observed at concentration of 3,000 mg/L MLSS. Increasing bio-reactor efficiency by increasing initial concentration of sludge possibly was due to more absorbance sites for phenols in cytoplasm and cell wall or more absorbing sites in extracellular polymers. Consequently, protection of biological mass for effective phenol removal is essential while increasing much more amounts of sludge content could be uneconomic [28]. Also, in a similar study, Jung et al. [29] showed that by reducing the amount of MLSS, phenol removal performance was reduced. The study by Kılıç and Dönmez [30] showed that with an increase in bio-mass concentration, phenol degradation also increased.

3.1.3. Effect of DO on system efficiency

Since the system under study is aerobic, results showed that dissolved oxygen concentration in BF/AS system is sufficient for phenol degradation. By increasing dissolved oxygen concentration from 0.7 to 3.5 mg/L, phenol removal efficiency increased by 54%, so achieving 100% removal efficiency is feasible. By increasing dissolved oxygen concentration from 0.7 to 1.3 mg/L, the curve slope increased more, so by increasing 0.6 mg/L DO, removal efficiency increased by 29%. Afterward, we observed smoother trend in phenol removal; therefore, by increasing 2.2 mg/L initial DO concentration, initial phenol content decreased about 25%. According to Fig. 6, bio-reactor efficiency at 2–2.6 mg/L was moderate and by increasing DO concentration to 3 mg/L, considerable change was observed. Dissolved oxygen concentration of >3 mg/L showed no change in phenol decreasing rate. According to results, absence of oxygen could be a limiting factor in phenol degradation. Increasing phenol degradation rate by increasing DO could be attributed to increasing permeation of oxygen into inner layers of biological clots and creating oxygen-rich medium for more and more activity of microorganisms and consequently more phenol removal [31]. By fivefold increasing dissolved oxygen concentration required for biological activity, phenol removal rate increased by 2.1 times.

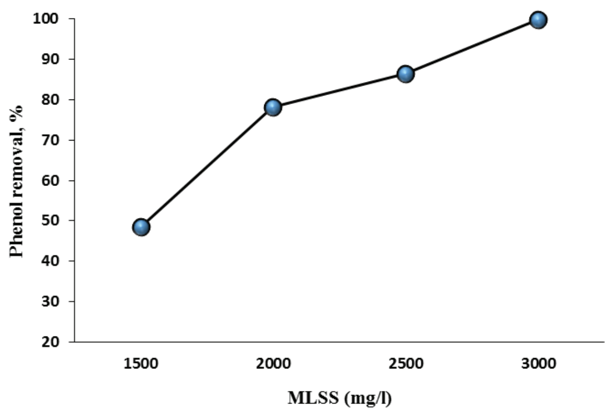

Fig. 5. Phenol removal efficiency at various concentrations of MLSS (initial increasing phenol concentration of 500 mg/L).

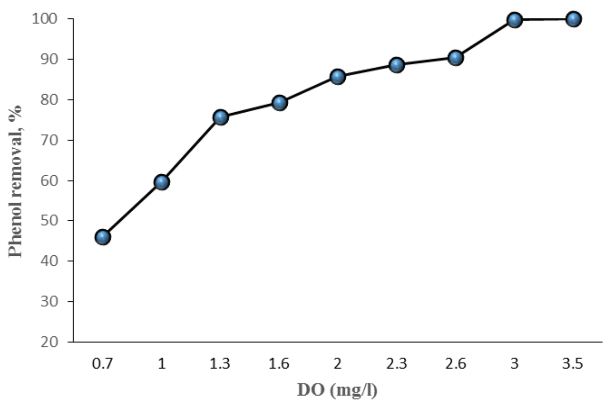

Fig. 6. Phenol removal efficiency at various dissolved oxygen concentrations for phenol removal from aqueous solutions (initial increasing phenol concentration of 500 mg/L).

3.1.4. Effect of organic load on system efficiency

In reactor studied, favorable yield was achieved at 5.23 kg/m^3 d organic load, so in this organic load 100% phenol removing efficiency was possible. Tolerance of organic load at different reactors varies for different wastewaters. Moussavi et al. [32] studied the phenol removal at 100–2,000 mg/L used in granular sequencing batch reactor and found that at 1,000 mg/L phenol concentration and organic load of 1.41 kg/m^3 d, removal amount was 99%. Yari et al. [33] upflow anaerobic sludge blanket in refining drink industries wastewater and found that by increasing reaction time from 1 to 80 d, organic load changed from 1 to 2.8 kg/m^3 d and at all stages, the system efficiency was 78%. Najafpour et al. [34] used rotating biological contactors for treatment of oil refining wastewater and showed that by increasing organic load from 0 to 0.25 kg/m^3 D, COD removal efficiency decreased from 80% to 30%. According to results of reactor operation, it can be concluded that studied reactor at high organic load (5.23 kg/m^3 d) had favorable yield.

3.2. Determining the interactions of operation parameters on BF/AS bio-reactor efficiency

CCD was selected to find interactions between process responses and variables. Laboratory results for response (phenol removal efficiency) were presented in Table 2 and Fig. 7. According to experimental setup of response level model indicating BF/AS efficiency relation with dependent variables, results were fitted to laboratory results. Table 3 presents results of analysis of variance (ANOVA) for reflected responses. Process response was second-order model. The terms in the models were obtained from removing non-significant variables and their interactions. Results of ANOVA determined models significance level. The *P* value obtained was 0.0001. Higher *P* value and lower *P* value represent model's significance [35]. *P* value < 0.0001 represents higher significance of the model. In other words, just 0.01 is probable that model significance might be resulted from error. Also, *P* value < 0.05 indicates that the terms of resulted models are significant. According to results, lack-of-fit for second-order model from the present study was not significant (P > 0.05). Non-significant lack-of-fit represents well predictability of the model.

Also, the resulted model fitness was confirmed by R^2 adjusted correlation coefficients and R^2 predicted among laboratory amounts and predicted by model so correlation coefficients of predicted R^2, adjusted R^2 and R^2 were very close to each other and close to 1. R^2 coefficient described as explained variable to total variation ratio and assessing model fitness degree which logically was high in all models ($R^2 > 0.99$). In other words, changes for phenol removal were well explained through dependent variables. Assessing the accuracy determines error rate in experiments. If the accuracy was more than 4, then model accuracy was reported as favorable [36]. So, high ratio of accuracy in present study (158.19) verifies accuracy of phenol removal model. Additionally, low amount of standard deviations (SDs; 0.94) and coefficient of variance (CV; 1.45) implies the considerable accuracy and high reliability of results [37]. It should be noted that coefficients of mentioned equations and also mathematical symbols (±) represent amount and direction of impact of independent parameters on BF/AS efficiency. Eq. (1) represents experimental relation of encoded variables in combined BF/AS system, is presented. According to Eq. (1), *B* (glucose concentration) and *C* (dissolved oxygen concentration) coefficients, respectively, had the highest impact on phenol removal efficiency. According to Eq. (1), minus sign of parameter *B* indicates that phenol removal was inversely related to the concentration of glucose. While dissolved oxygen and concentration of mixed liquor suspended

Table 2
Experimental conditions and results in RSM

Run	*A*: MLSS, mg/L	*B*: glucose concentration, mg/L	*C*: dissolved oxygen, mg/L	Response, %
1	3,000	0	3	100
2	2,250	0	2	80.6
3	1,500	250	2	54.4
4	2,250	250	2	66.1
5	2,250	250	2	67.2
6	2,250	250	2	65.4
7	2,250	250	2	65.2
8	1,500	500	1	26.6
9	2,250	250	3	75.3
10	1,500	0	3	81.6
11	2,250	250	2	67.3
12	2,250	500	2	50.1
13	3,000	0	1	76.2
14	1,500	0	1	57.4
15	3,000	500	1	47.5
16	2,250	250	1	53.3
17	1,500	500	3	50.2
18	3,000	250	2	77.4
19	2,250	250	2	66.3
20	3,000	500	3	71.2

Table 1
Phenol concentration change to residence time ratio

Residence time (T), h	Effluent phenol concentration (A) mg/L	Concentration difference (ΔA) mg/L	Residence time difference (ΔT) h	$-(\Delta A/\Delta T)$	Mean $-A$	Log$-(\Delta A/\Delta T)$	Mean Log (A)
1.5	450	–160	1.5	106.7	370	2.03	2.57
3	290	–145	1.5	90.6	217.5	1.96	0.29
4.6	145	–105	6.2	16.9	92.5	1.23	0.09
10.8	40	–	–	–	–	–	–

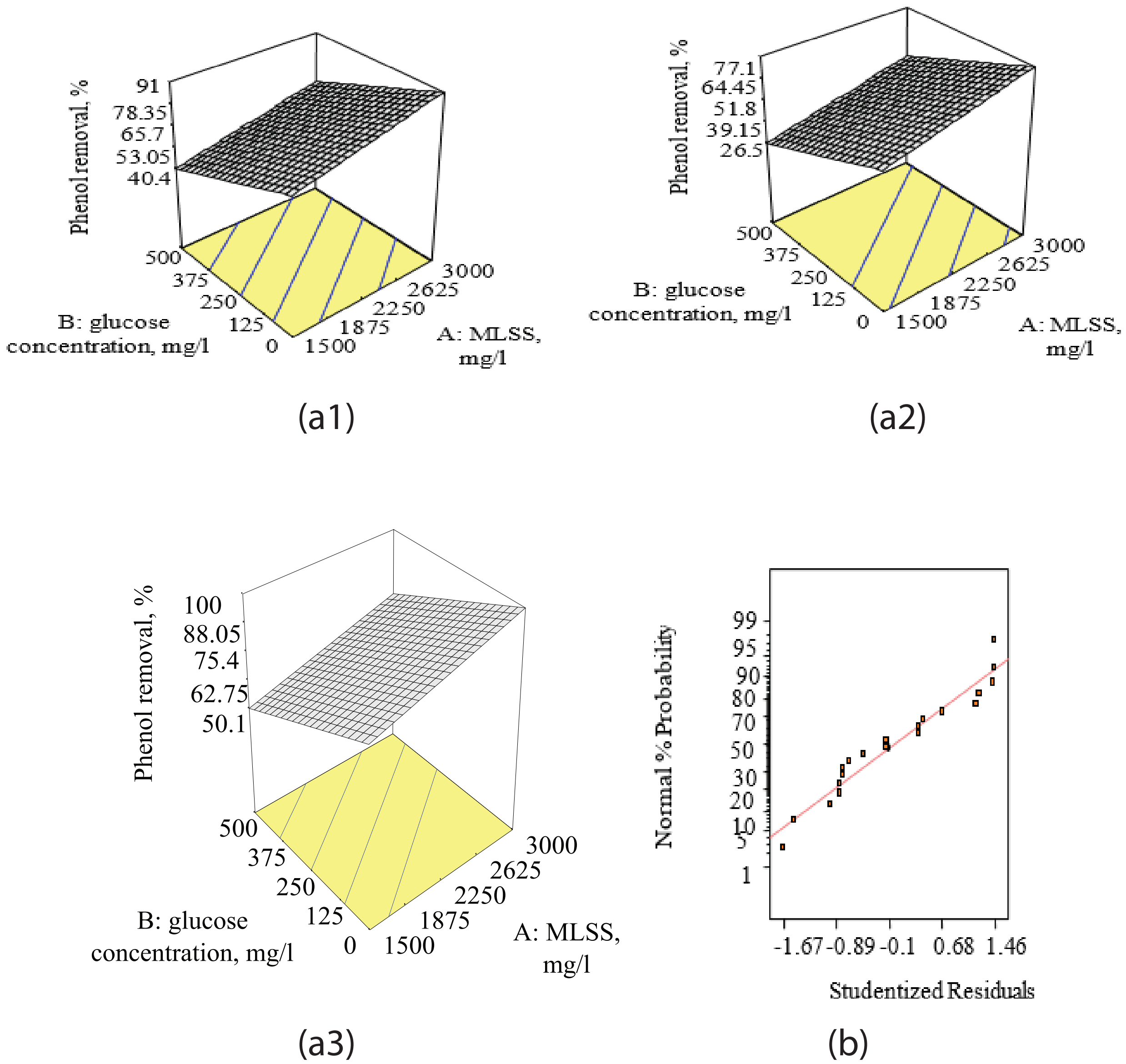

Fig. 7. Three-dimensional plan (a) interaction impact of MLSS concentration and glucose for phenol removal through BF/AS system and dissolved oxygen concentration (a1) 1 mg/L, (a2) 2 mg/L, (a3) 3 mg/L and (b) normal plot of residuals.

Table 3
Phenol removal model characteristics based on central composite design

Response	Phenol removal, %
Type of model	Quadratic
Degrees of freedom	4
Sum of squares	4,725.35
Mean square	1,181.34
F value	1,342.43
Prob > *F*	0.0001
Adequate precision	158.19
SD	0.94
CV	1.45
PRESS	22.93

solids had a direct relationship with removal efficiency ($P < 0.001$). By a factor of 10.3 the concentration of mixed liquor suspended solids in the bio-reactor had minimal impact on performance ($P < 0.001$).

$$\text{Phenol removal, \%} = +66.7 + 10.3A - 15B + 11.8C - 2.1C^2 \quad (1)$$

In present study, predicted R^2 was 0.995 and in accordance with adjusted R^2 (0.996). Results of ANOVA are presented in Table 3. In order to evaluate the model accuracy, among studied laboratory conditions, one run was selected and real amounts were compared by predicted ones. Optimum conditions accuracy for each response was evaluated by DES software through SD and real amounts obtained were very close to predicted amounts by model (Table 4).

Table 4
Validity assessment test

Phenol removal, %			
Optimum condition	Experimental level	Response model with 95% confidence interval	Standard error
MLSS = 2,250 mg/L DO: 3 mg/L Glucose concentration: 250 mg/L	75	76.29	0.42

4. Conclusion

This study aimed at investigating the efficiency of BF/AS combined process in phenol removal from aqueous solution. The CCD was found out to be a valuable tool in establishing optimal conditions through a response surface study. Four variables, that is, dissolved oxygen concentration, hydraulic residence time, MLSS and glucose concentration on phenol removing efficiency were evaluated. The maximum removal efficiency of the phenol was determined at the following conditions: 3 mg/L dissolved oxygen content, 3,000 mg/L MLSS, 250 mg/L of glucose and hydraulic residence time of 7 h. The findings in this study strongly suggested that the combination of conventional biotechnologies (AS/BF) proved to be promising techniques for the treatment of wastewater and aqueous solution containing phenol.

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