Optimization of Modified Fenton (FeGAC/H$_2$O$_2$) Pretreatment of Antibiotics

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ABSTRACT

The study examined modified Fenton (FeGAC/H$_2$O$_2$) pretreatment of the antibiotics amoxicillin and cloxacillin in aqueous solution for biological treatment. The treatment was optimized by the response surface methodology (RSM). The optimum operating conditions at pH3 were H$_2$O$_2$/COD molar ratio 2.0, FeGAC dose 3.5 g/L and reaction time 90 min for 87.53% removal of COD, 78.01% removal of TOC, and 98.24% removal of NH$_3$-N. Biodegradability (BOD$_5$/COD ratio) improved from zero to 0.36, indicating the effluent was amenable to biological treatment. Meanwhile, FTIR spectra indicated degradation of the antibiotics. Compared with Fenton or photo-Fenton, modified Fenton (FeGAC/H$_2$O$_2$) was more effective in the pre-treatment of the antibiotics amoxicillin and cloxacillin in aqueous solution for biological treatment.

Keywords: Modified Fenton (FeGAC/H2O2), antibiotic aqueous solution; amoxicillin, cloxacillin, response surface methodology (RSM)

INTRODUCTION

The ultimate fate of pharmaceutical compounds after their intended use was not monitored until their presence in the aquatic system was detected. Consequently, these compounds have attracted increased attention as potential water pollutants (Jones *et al.*, 2007). Over 60 pharmaceutical compounds from a variety of therapeutic groups (including but not limited to, analgesics, antibiotics, anti-epileptics, β-blockers, β$_2$-sympathomimetics, blood lipid regulators and X-ray contrast media) have been found in the aquatic matrix (Jones *et al.*, 2001). These compounds have also been reportedly found in surface water, groundwater, sewage effluent, and in drinking water (Stackelberg *et al.*, 2004). The concern

about the presence of antibiotics in low concentrations in the aquatic environment is the development of antibiotic resistant bacteria (Walter & Vennes, 1985). Degradation of these antibiotics is a current challenge, especially as the conventional wastewater treatment plants (STPs) are not designed to remove them (Gulkowska *et al.,* 2008). Amoxicillin and cloxacillin antibiotics are broad beta-lactam antibiotic that belongs to the class of penicillin. They are used in veterinary and human medicine as initial medicine in the treatment of majority of infectious diseases (Anacona & Figueroa, 1999).

Advanced oxidation process (Fenton or photo-Fenton) pre-treatment of antibiotics in aqueous solution enhances biodegradability and produces a new effluent that is amenable to biological treatment.

Fenton and photo-Fenton pretreatments of the antibiotics amoxicillin, ampicillin and cloxacillin in aqueous solution have been reported (Elmolla & Chaudhuri, 2009a, 2009b) as advanced oxidation strategies for enhancement of antibiotics aqueous solution.

Activated carbon (AC) is known to decompose hydrogen peroxide (H_2O_2). Presumably, the process involves the exchange of a surface hydroxyl group with hydrogen peroxide anion (Eq. 1), according to Bansal *et al.* (1998, as cited in Khalil *et al.,* 2001). The formed surface peroxide is regarded as having an increased oxidation potential which enables the decomposition of another hydrogen peroxide molecule with release of oxygen and regeneration of the AC surface (Eq. 2).

$$AC-OH + H^+ OOH^- \longrightarrow AC-OOH + H_2O \qquad (1)$$

$$AC-OOH + H_2O_2 \longrightarrow AC-OH + H_2O + O_2 \qquad (2)$$

Beside this decomposition reaction, hydrogen peroxide can obviously be activated on the AC surface involving the formation of $OH^•$. AC is considered to function as an electron-transfer catalyst similar to the Haber–Weiss mechanism known from the Fenton reaction, with AC and AC^+, as the oxidized and reduced catalyst states (Eqs. 3-4) (Kimura & Miyamoto, 1994).

$$AC + H_2O_2 \longrightarrow AC^+ + HO^- + OH^• \qquad (3)$$

$$AC^+ + H_2O_2 \longrightarrow AC + HO_2^• + H^+ \qquad (4)$$

In the $FeGAC/H_2O_2$ process, activated carbon acts as an adsorbent, and yet at the same time, the presence of ferrous iron enables the adsorbent (FeGAC) to interact as a catalyst to degrade adsorbed or dissolved organic matter. GAC is also involved in the catalytic activity due the graphitic structure and functional groups on their surface (Bach & Semiat, 2010). Therefore, FeGAC has high adsorption capacity due to GAC surface and coating of iron oxide. In addition, the oxidation strength of H_2O_2 is enhanced by the catalytic properties of FeGAC in the removal of antibiotics from the aqueous solution. There is no detailed study which has reported contaminants removal mechanism in the $FeGAC/H_2O_2$ process. However, Fenton mechanism combined with adsorption process is basic for the $FeGAC/H_2O_2$ process to proceed. Oxidation with Fenton's reagent is based on hydroxyl radicals produced by catalytic decomposition of hydrogen peroxide (H_2O_2) in reaction to ferrous ions (Fe^{2+}) (Chamarro *et al*., 2001). In the photo-Fenton process, additional reactions occur in the presence of light that

produce hydroxyl radicals or increase the production rate of hydroxyl radicals (Pignatello *et al.*, 1999) and thus, increasing the efficiency of the process. Adsorption studies have been reported using the FeGAC/H$_2$O$_2$ process (Fan *et al.,* 2007; C-C Chen *et al.,* 2011).

Modified Fenton process could be in terms of replacement of the standard catalysts such as ferrous and/or ferric ions with novel adsorbents, and an increase in process temperature up to 80–120°C, i.e. the so-called catalytic wet hydrogen peroxide oxidation process, application of novel, highly stable and active heterogeneous catalysts (Fe$_2$O$_3$ or Fe supported into SBA-15 mesostructured material, etc., and the application of iron chelates (typically organic acids) can promote chemical oxidation of contaminants by formation of iron–chelate complexes among others. The primary advantages of some modifications include effective generation of hydroxyl radicals at near-neutral pH and reduction of iron sludge production in other cases (Goi *et al.*, 2008).

There is insufficient literature to buttress the determination of the equilibrium proportions of ferric species as a function of pH and concentration or less presence of iron-related ionic species with respect to ratio, dosage and time in the FeGAC/H$_2$O$_2$ process. The species of impregnated iron were affected by impregnation methods and synthesizing conditions (Jang *et al.*, 2008; Thirunavukkarasu *et al.,* 2003) and that at different temperatures, the impregnated iron oxide manifested a more crystalline form (Jang *et al.*, 2008). In another study, the initial amount of iron oxides on FeGAC surface was approximately 38 mg Fe/g GAC, and after the treatment, the loss of iron oxide became less than 5% (Chen *et al.,* 2011). Acid black decolorization rate of 8.71 mg/min by the FeGAC/H$_2$O$_2$ process has been reported by Fan *et al.* (2006).

Generally, it is known where ferrous ions and hydrogen peroxide are used to generate OH, and production of iron sludge is inevitable. Nonetheless, this may cause additional cost with regard to handling and disposal. To circumvent the problem of iron sludge production in the processes reported, a modified Fenton process − Fe-granular activated carbon/hydrogen peroxide (FeGAC/H$_2$O$_2$) process − was proposed (Fan *et al.*, 2007). This is possible by coating iron oxides on the surface of granular activated carbon (GAC) and could minimize the production of sludge. This has been applied in the decolourization of Acid Black 24 (Fan *et al.,* 2006) removal of humic substances, namely, humic and fulvic acids in municipal landfill leachate (Fan *et al.,* 2007) and degradation of crystal violet (Chen *et al.,* 2011). However, the application of the treatment process of antibiotics in aqueous solution has not been reported.

The present study examined the application of the FeGAC/H$_2$O$_2$ process in the pretreatment of the antibiotics amoxicillin and cloxacillin in aqueous solution for biological treatment. The treatment was optimized by response surface methodology for the removal of chemical oxygen demand (COD), total organic carbon (TOC), and ammonia-nitrogen (NH$_3$-N).

MATERIALS AND METHODS

Fe-Granular Activated Carbon (FeGAC)

Granular activated carbon (GAC) was obtained from the Calgon Corporation, Pittsburgh, PA, and was ground to a size of 425 μm. FeGAC was prepared by mixing GAC in a solution of ferrous sulfate (FeSO$_4$•7H$_2$O) for 24 h at 40 mg ferrous sulphate/g GAC, followed by drying at 105ºC. The resulting FeGAC was washed several times with distilled water, dried at 105ºC and stored in a stopper glass bottle.

Antibiotic Aqueous Solution

The aqueous solution of the antibiotics amoxicillin (AMX) and cloxacillin (CLX) was prepared weekly by dissolving 150 mg each of the antibiotics in 1000 mL distilled water and stored at 4ºC. AMX and CLX were obtained from a commercial source (Farmaniaga Company, Malaysia). The characteristics of the antibiotic aqueous solution are as follows: COD 390 mg/L, TOC 168.8 mg/L, NH_3-N 20.6 mg/L and BOD_5/COD ratio zero.

TABLE 1
Characteristics of antibiotics solution

Parameter	Range
COD	390.0 ± 4.0
TOC	168.8 ± 2.5
NH_3-N	20.6 ± 1.2

Analytical Methods

Chemical oxygen demand (COD) was measured according to Method 5220D (closed reflux, colorimetric method) of the Standard Methods (APHA, 2005). If the sample contained hydrogen peroxide (H_2O_2), to reduce interference in COD determination, pH was increased to above 10 to decompose H_2O_2 to oxygen and water (Talinli & Anderson, 1992; Kang *et al*., 1999). A TOC analyzer (Model 1010; O & I Analytical) was used in determining total organic carbon (TOC). Meanwhile, NH_3-N was measured according to Method 8038 (Nessler Method) of the Water Analysis Handbook (Hach, 2002). Five-day biochemical oxygen demand (BOD_5) was measured according to Method 5210B (seeding procedure) of the Standard Methods (APHA, 2005). The bacterial seed for BOD_5 test was obtained from a municipal wastewater treatment plant. DO was measured using YSI 5000 dissolved oxygen meter. FTIR spectra of the untreated and treated antibiotic aqueous solution were taken by using *Shimadzu FTIR-8400S.*

FeGAC/H_2O_2 Treatment

Batch FeGAC/H_2O_2 treatment was performed in 250-mL conical flasks with 200 mL of the antibiotic aqueous solution adjusted to pH3, which is the optimum pH for Fenton treatment of antibiotic aqueous solution (Elmolla & Chaudhuri, 2009a). H_2O_2 and FeGAC were added simultaneously according to the selected H_2O_2/COD molar ratio and FeGAC dose and the flasks were placed on an orbital shaker. At the selected reaction time for each run 1 to 20 as obtained from the central composite design (CCD) in Table 3, a flask was removed from the orbital shaker and an aliquot of the supernatant was filtered through 0.45 µm membrane filter for measurement of COD, TOC and NH_3-N, and filtered through 0.20 µm membrane filter for FTIR spectra.

TABLE 2
The actual and coded values for the independent variables of the CCD design

Independent variable values		Coded		
		−1	0	+1
	Units	Actual value		
A: H$_2$O$_2$/COD	molar ratio	1.0	2.0	3.0
B: FeGAC	g/200ml	0.4	0.7	1.0
C: Time	min	60	90	120

TABLE 3
Experimental design and observed removal

Experimental Design			Removal (%)		
A: H$_2$O$_2$/COD (Molar ratio)	B: FeGAC (g/200 mL)	C: Reaction Time (min)	COD	TOC	NH$_3$-N
2.00 (0.0)	1.20 (1.68)	90.00 (0.0)	67.0	63.45	89.0
3.68 (1.68)	0.70 (0.0)	90.00 (0.0)	82.1	43.55	100.0
3.00 (1.0)	0.40 (-1.0)	120.00 (1.0)	77.0	29.32	96.0
2.00 (0.0)	0.70 (0.0)	90.00 (0.0)	85.1	88.75	97.0
2.00 (0.0)	0.70 (0.0)	39.55 (-1.68)	83.0	55.81	96.0
0.32 (-1.68)	0.70 (0.0)	90.00 (0.0)	98.2	74.42	97.0
2.00 (0.0)	0.20 (-1.68)	90.00 (0.0)	63.3	39.31	96.6
3.00 (1.0)	0.40 (-1.0)	60.00 (-1.0)	85.8	56.95	100.0
1.00 (-1.0)	1.00 (1.0)	60.00 (-1.0)	67.0	53.77	93.0
1.00 (-1.0)	0.40 (-1.0)	120.00 (1.0)	75.0	69.65	97.0
2.00 (0.0)	0.70 (0.0)	90.00 (0.0)	100.0	76.82	100.0
2.00 (0.0)	0.70 (0.0)	90.00 (0.0)	89.0	74.44	97.0
2.00 (0.0)	0.70 (0.0)	90.00 (0.0)	83.0	66.45	100.0
2.00 (1.0)	0.70 (0.0)	140.45 (1.68)	97.4	68.85	98.0
3.00 (1.0)	1.00 (1.0)	60.00 (-1.0)	84.0	47.35	96.0
1.00 (-1.0)	0.40 (-1.0)	60.00 (-1.0)	86.7	57.99	97.0
3.00 (1.0)	1.00 (1.0)	120.00 (1.0)	93.8	61.28	96.0
2.00 (0.0)	0.70 (0.0)	90.00 (0.0)	83.4	68.53	97.0
1.00 (-1.0)	1.00 (1.0)	120.00 (1.0)	96.0	66.62	94.0
2.00 (0.0)	0.70 (0.0)	90.00 (0.0)	85.0	92.89	98.5

Optimization and Response Surface Modelling

Design expert software (version 6.0.7) was used for the statistical design of experiment and data analysis. Central composite design (CCD) of the response surface methodology (RSM) (Khuri & Cornell, 1996; Bezerra *et al*., 2008) was used to optimize the operating conditions (variables) of the FeGAC/H$_2$O$_2$ treatment. The variables were simultaneously changed in a central composite circumscribed design. The coded values of the variables were H$_2$O$_2$/COD molar ratio (A), FeGAC dose (g/200 mL) (B), and reaction time (min) (C), and they were varied over three levels: 1 (low), 0 (central) and +1 (high). The variables H$_2$O$_2$/COD molar ratio,

FeGAC dose and reaction time were studied in the range of 1.0-3.0, 0.4-1.0 g/200 mL and 60-120 min, respectively (Table 2). The data range for the independent variables was obtained from previous studies (Elmolla & Chaudhuri, 2009a, 2009b) and hence, no preliminary study was conducted. The significance of the chosen variables includes that H_2O_2/COD molar ratio relates to the approximate H_2O_2 concentration required based on the stoichiometric ratio with respect to COD of the antibiotics aqueous solution and was calculated by assuming that a complete oxidation of COD occurred. H_2O_2 ensures the oxidation of the antibiotic in the aqueous solution. The amount of H_2O_2 influences the modified Fenton process because excessive H_2O_2 will pose an inhibitory effect in the degradation of antibiotics. This could be due to $OH^\bullet$ scavenging and the formation of another radical ($HO_2^\bullet$), which has an oxidation potential considerably smaller than $OH^\bullet$, as in Equation (5) (Arslan-Alaton *et al*., 2009).

$$OH^\bullet + H_2O_2 \longrightarrow HO_2^\bullet + H_2O_2 \tag{5}$$

FeGAC is important for the catalysis of the process in the presence of H_2O_2. In addition, FeGAC serves as an adsorbent with GAC added to increase the available catalytic sites. The reaction time is a variable to observe and determine the best operating time for an optimum degradation of the antibiotics. The optimum H_2O_2/COD molar ratio 3.0 and reaction time 60 min for the Fenton treatment of antibiotic aqueous solution (Elmolla & Chaudhuri, 2009a) and FeGAC dose 0.8 g/200 mL for FeGAC/H_2O_2 treatment of a dye wastewater (Fan *et al.,* 2006) have been reported. CCD generally consists of a 2k factorial with nF factorial runs, 2k axial or star runs, and nC (i.e. centre runs). In this study, a total of 20 experiments were performed according to $2^k + 2k + 6$, where k is the number of independent variables (k=3). Fourteen experiments were enhanced with six replications to assess the pure error. The chosen response parameters of the FeGAC/H_2O_2 treatment were removal of COD, TOC and NH_3-N. Regression analyses, response surface plots, analysis of variance (ANOVA), perturbation plots and normal probability plot of the studentized residuals were carried out using the design expert software. The optimum operating conditions were identified from the response surface plots and the response equations simultaneously.

The following quadratic model Equation (6) was used to estimate the optimal values:

$$Y = \beta_o + \sum_{j=1}^{k} \beta_j X_j + \sum \beta_{jj} X^2_j + \sum_i \sum_{<j=2}^{k} \beta_{ij} X_i X_j + \varepsilon \tag{6}$$

where Y is the response; X_i and X_j are the variables; β_0 is a constant coefficient; β_j, β_{jj}, and β_{ij} are the interaction coefficients of linear, quadratic and second-order terms, respectively; k is the number of studied factors; and ε is the error. The quality of the fit was expressed by the coefficient of determination (R^2). The main indicators demonstrating the significance and adequacy of the model used include the F-value (Fisher variation ratio), probability value (Prob>F), and the Adequate Precision (Arslan-Alaton *et al*., 2009).

RESULTS AND DISCUSSION

Statistical Analysis

The results obtained were analyzed by ANOVA to assess the "goodness of fit". The models for COD, TOC, and NH$_3$-N removal (Y_1, Y_2 and Y_3) were significant by the F-test at 95% confidence level if Prob>F<0.05. The following fitted regression model (equations in terms of coded values) was obtained to quantitatively investigate the effects of H$_2$O$_2$/COD molar ratio (A), FeGAC dose (B), and reaction time (C) on COD, TOC and NH$_3$-N removal.

COD removal
$$Y_1 = 87.53 - 0.82A - 7.58B^2 + 7.41BC \tag{7}$$

TOC removal
$$Y_2 = 78.01 - 7.69A + 6.92A^2 - 9.61B^2 + 5.74C^2 \tag{8}$$

NH$_3$-N removal
$$Y_3 = 98.24 + 0.88A - 1.74B \tag{9}$$

In Equations 7, 8 and 9, the values of the sum of a constant (β_0), 87.53, 78.01 and 98.24 represent the percentage removal of COD, TOC and NH$_3$-N, respectively. The positive sign indicates that the variable is directly proportional to the response (COD, TOC and NH$_3$-N removal) and the negative sign indicates that the variable is inversely proportional to the response. Table 3 shows the experimental design, real and codified values (in parentheses) of the variables and response parameters (observed removal of COD, TOC and NH$_3$-N). Tables 4a-4c show the ANOVA for the response surface quadratic model. In this study, A, B^2 and BC; A, A^2, B^2 and C^2; and A and B; were found to be significant model terms for COD, TOC and NH$_3$-N removal, respectively, while others were discarded from the study to ensure an improved model. Adequate precision (AP) compares the range of the predicted values at the design points to the average prediction error. Ratios greater than 4 indicate adequate model discrimination and can be used to navigate the design space defined by CCD (Ghafari *et al.,* 2009). AP for all the responses was greater than 4. The probability of the lack of fit (PLOF) describes the variation of the data around the fitted model. This is significant when PLOF<0.05 for the overall model. PLOF for all the responses were <0.05. The R^2 values shown in Table 4 for the response parameters COD (R^2 = 0.7826), TOC (R^2 = 0.8034) and NH$_3$-N (R^2 = 0.8635) indicate that the model is good enough for the quadratic fits to navigate the design space defined by CCD (Ghafari *et al.,* 2009).

Process Analysis

Fig.1, Fig.2 and Fig.3 show the response surface plots for COD, TOC and NH$_3$-N removal. The maximum COD, TOC and NH$_3$-N removals were 87.53, 78.01 and 98.24%, respectively, under H$_2$O$_2$/COD molar ratio 2.0, FeGAC dose 0.70 g/200mL (3.5 g/L), pH 3 and reaction time 90 min. In the Fenton treatment of the antibiotics amoxicillin, ampicillin and cloxacillin in the aqueous solution, Elmolla and Chaudhuri (2009a) reported the maximum COD and DOC removal of 81.4 and 54.3%, respectively, under the H$_2$O$_2$/COD molar ratio 3.0, H$_2$O$_2$/

TABLE 4
The ANOVA for analysis and adequacy of the quadratic model

COD removal (%)

Source	Sum of Squares	DF	Mean Square	F Value	Prob > F
Model	1610.11	9	178.90	4.00	<0.0001
A	9.11	1	9.11	18.47	0.0016
B^2	826.09	1	826.09	9.83	0.0106
BC	439.56	1	439.56		
Residual	447.17	10	44.72		
Lack of Fit	239.64	5	47.93	1.15	<0.0001
Pure Error	207.53	5	41.51		

Std. Dev. = 6.69   PRESS = 2222.2 R^2 = 0.7826   Adj. R^2 = 0.5870 Adequate Precision = 6.985

TOC removal (%)

Source	Sum of Squares	DF	Mean Square	F Value	Prob > F
Model	3746.01	9	416.22	4.54	<0.0001
A	807.87	1	807.87	8.81	0.0141
A^2	683.08	1	683.08	7.45	0.0212
B^2	1330.31	1	13330.31	14.51	0.0034
C^2	467.99	1	467.99	5.11	0.0474
Residual	916.59	10	91.66		
Lack of Fit	342.17	5	68.43	0.60	<0.0001
Pure Error	574.42	5	114.88		

Std. Dev. = 9.57   PRESS = 3751.65 R^2 = 0.8034   Adj. R^2 = 0.6265 Adequate Precision = 6.726

NH_3-N removal (%)

Source	Sum of Squares	DF	Mean Square	F Value	Prob > F
Model	113.29	9	12.59	7.03	<0.0001
A	10.62	1	10.62	5.93	0.0351
B	41.32	1	41.32	23.06	0.0007
Residual	17.92	10	1.79		
Lack of Fit	7.04	5	1.41	0.65	<0.0001
Pure Error	10.88	5	2.17		

Std. Dev. = 1.34   PRESS = 70.87 R^2 = 0.8635   Adj. R^2 = 0.7406   Adequate Precision = 10.661

Fe^{2+} molar ratio 10.0, pH3 and reaction time of 60 min. In the photo-Fenton treatment of the antibiotics in aqueous solution, Elmolla and Chaudhuri (2009b) reported the maximum COD and DOC removal of 80.8 and 58.4%, respectively, under the H_2O_2/COD molar ratio 1.5, H_2O_2/ Fe^{2+} molar ratio 20, pH3 and irradiation time of 50 min. Thus, the modified Fenton (FeGAC/ H_2O_2) was more effective than Fenton or photo-Fenton in the pretreatment of the antibiotics in the aqueous solution. Fig.4, Fig.5 and Fig.6 show the perturbation plot for COD, TOC and NH_3-N removal, respectively. The adequacies of the models were also evaluated by the residuals, i.e. Normal probability plot of the studentized residuals (Fig.4 - Fig.6) and indicate

that there is no abnormality in the model as all the data were found around the line of the 'best fit'. Fig.4 - Fig.6 show the perturbation plots for COD, NH$_3$-N and TOC removals. The plots show how response (COD, TOC and NH$_3$-N removal) changes as each variable (A: H$_2$O$_2$/COD molar ratio, B: FeGAC dose, g/mL, and C: reaction time) moves from the chosen reference point, i.e. 0 (central) level, with other variables held constant. Thus, a comparative effect of all the independent variables on the removals was observed. A sharp curvature is seen in Figure 4, indicating that the COD removal was the most sensitive to variable B: FeGAC dose. The comparatively flat curvature from variable A: H$_2$O$_2$/COD molar ratio and C: reaction time shows less sensitivity on the COD removal. In Fig.5, all the variables (A: H$_2$O$_2$/COD molar ratio, B: FeGAC dose, g/mL and C: reaction time) had a moderately sharp curvature which showed that the TOC removal was sensitive to the three variables. Fig.6 shows that NH$_3$-N removal was most sensitive to variable B: FeGAC dose compared to A: H$_2$O$_2$/COD molar ratio, and C: reaction time during the treatment process. The normal probability plots of the studentized residuals for the COD, TOC and NH$_3$-N removal are shown in Fig.7- Fig.9. A normal probability plot graphically shows whether or not the residuals follow a normal distribution, hence, the points follow a straight line. The data showed a good plot although some scattering was also observed.
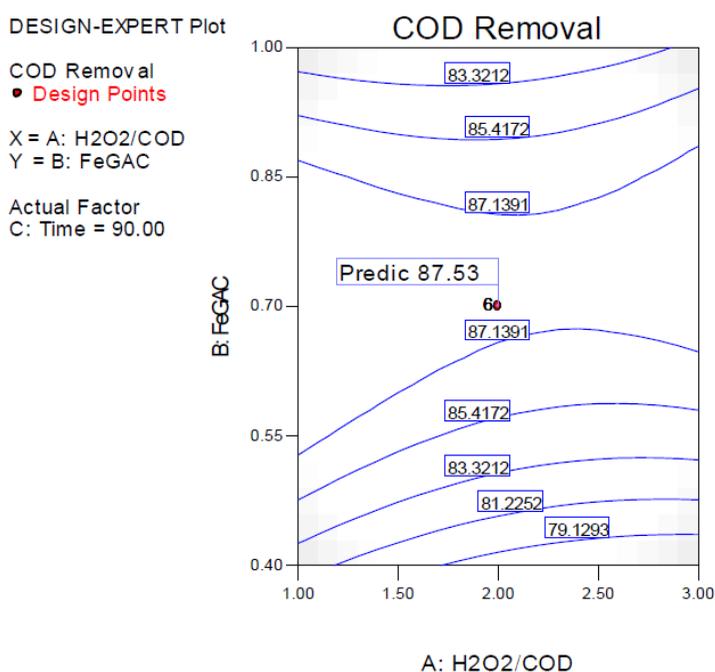
Fig.1: Response surface plot for COD removal
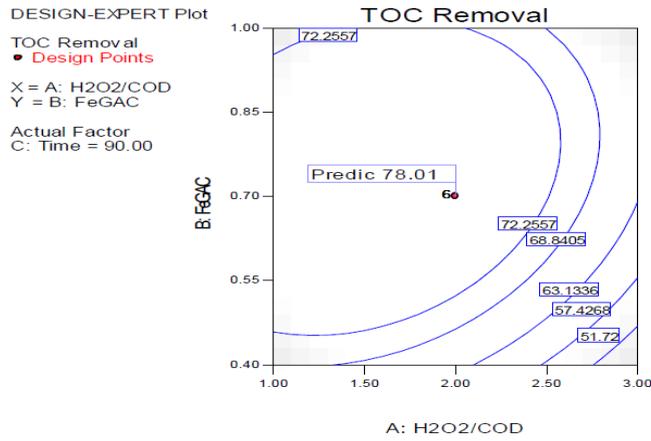
Fig.2: Response surface plot for TOC removal
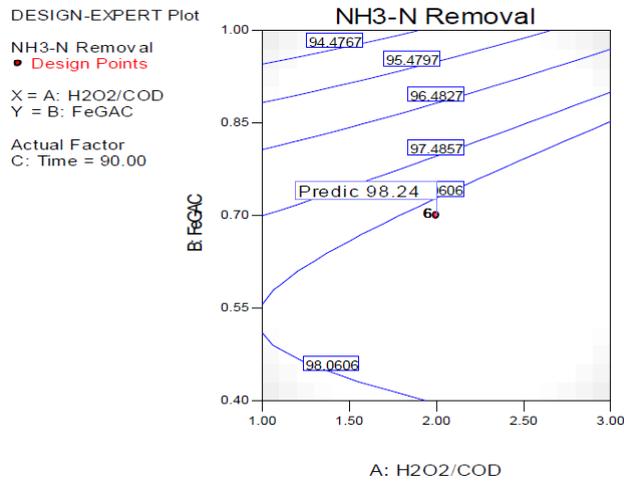
Fig.3: Response surface plot for NH$_3$-N removal
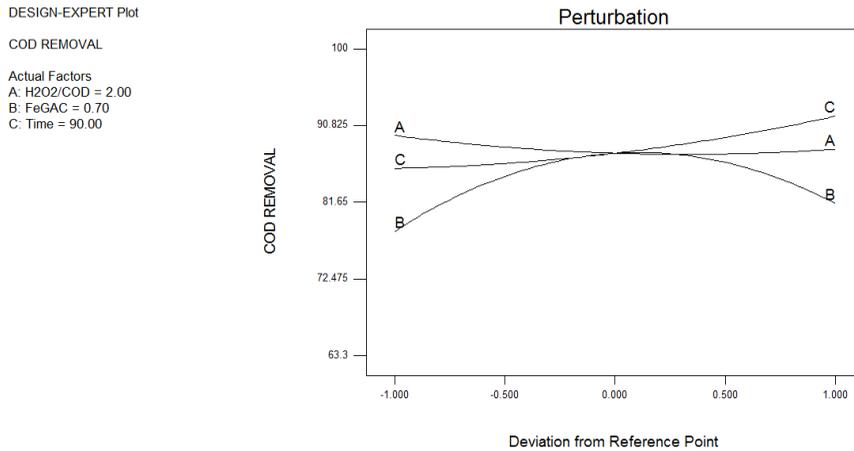
Fig.4: Perturbation plot for COD removal
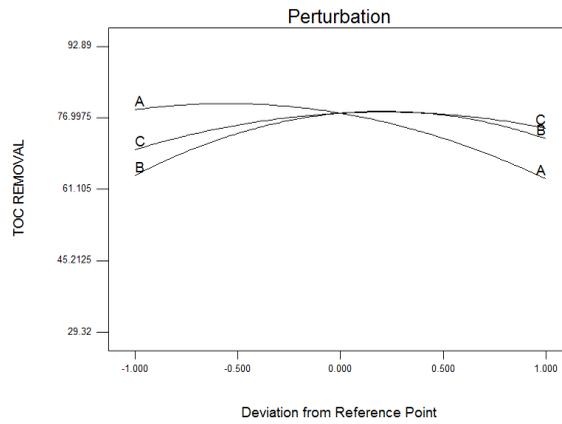
Fig.5: Perturbation plot for TOC removal
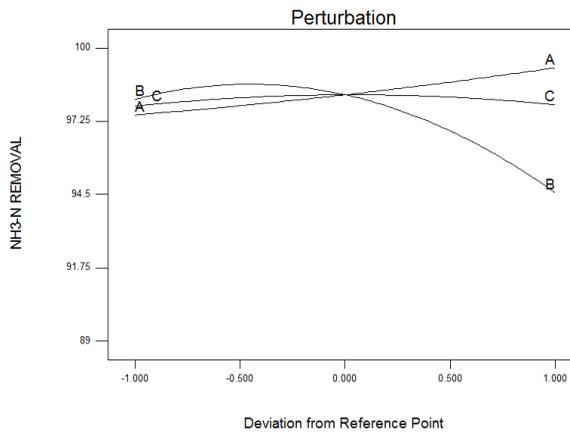
Fig.6: Perturbation plot for NH$_3$-N removal
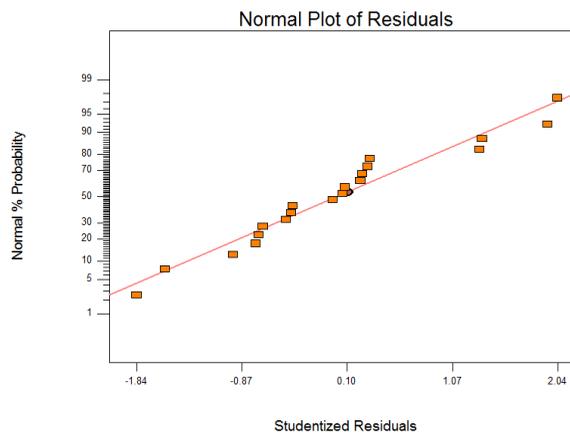
Fig.7: Normal probability plot of the studentized residuals for COD removal
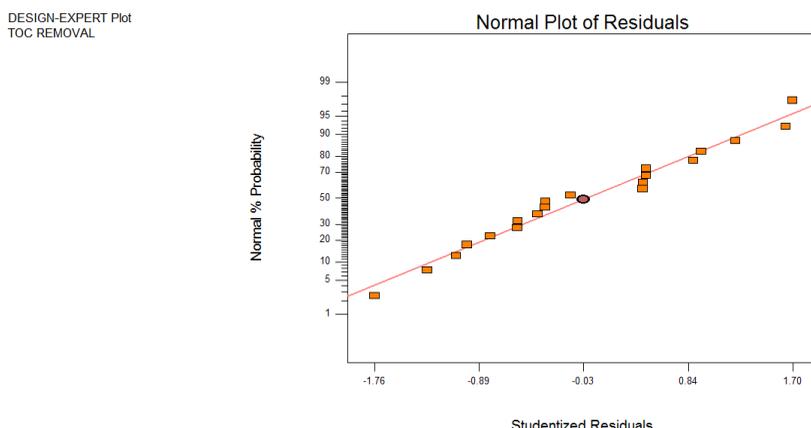
Fig.8: Normal probability plot of the studentized residuals for TOC removal
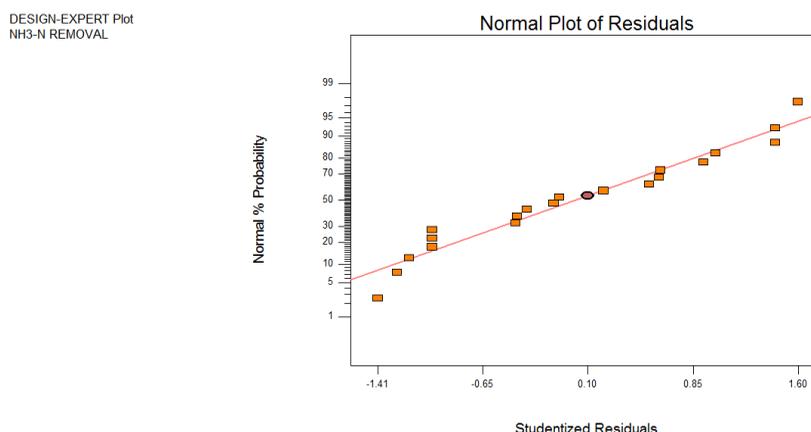
Fig.9: Normal probability plot of the studentized residuals for NH_3-N removal

Confirmatory Experiments

In order to validate optimization by RSM, three confirmatory experiments were conducted under the optimum operating conditions to verify the model prediction. As shown in Table 5, model prediction and experimental removal efficiency were in close agreement with less than 0.40% error.

TABLE 5
Model prediction and experimental removal efficiency

Parameter	Model prediction	Experimental removal	Error (%)
COD removal (%)	87.53	85.58,88.9, 89.20 (87.89)	0.36
TOC removal (%)	78.01	78.08,78.0,78.03 (78.04)	0.03
NH_3-N removal (%)	98.24	98.89,98.4,98.58 (98.62)	0.38

Antibiotic Degradation and Biodegradability

FTIR spectroscopy provides information on the chemical structure of organic bonds in a sample for complete structural analogy. Fig.10 and Fig.11 are important to present the analysis for the residual organic matter by the FTIR spectroscopy, and to ascertain the extent of antibiotic degradation of the organic bonds in the untreated and treated antibiotics under optimum operating conditions. Generally, when infrared light interacts or passes through an aqueous solution, the chemical bonds present in that solution stretches, contracts and/or bends. As a result, chemical functional group tends to absorb infrared radiation in a specific wavelength range, regardless of the structure of the rest of the molecule. The infrared spectra of organic bonds consisting of beta-lactam carbonyl group and aromatic ring could shift to specific bands due to degradation (Anacona & Figueroa, 1999; Rozas *et al.,* 2010). The IR features of penicillins are reported in the range of 1550 cm^{-1} and 1880 cm^{-1}, but these usually occur at about 1770 cm^{-1} as a result of the vibration of strong bands of the beta-lactam carbonyl group (Kukpa, 1997). The main difference between the beta-lactam carbonyl group and aromatic ring is located at 1596.95 cm^{-1}, a signal that can be attributed to conjugated carbonyl groups which only become visible in the intermediates formed by the modified Fenton reaction (Rozas *et al.,* 2010). The characteristic band which occurred at 1637.45 cm^{-1} for the untreated antibiotic aqueous solution (Fig.10) shifted and modified to 1596.95 cm^{-1} (see Fig.11). This can be further attributed to the pairing up of the organic group degradation intermediates (Rozas *et al.,* 2010). Meanwhile, Arjunan *et al*. (2012) reported that N–H stretching vibrations were observed between 3300-3500 cm^{-1} and subsequently led to deformation vibration in the short range of 1650-1580 cm^{-1} of the spectrum. In a study on amoxicillin FTIR/ATR, a stretch resulting from ν(N10H) and ν(O13H) occurred at 3451 and 3529 cm^{-1}, respectively (Bebu *et al.,* 2011). The band between 3361 and 3449 cm^{-1} in cloxalillin was due to ν(O-H) stretching (Adedibu *et al*., 2011). The observed shift and/or modification of the band in cloxacillin occurred through the oxygen atom of the lactam carbonyl group (Anacona & Carman, 2005).
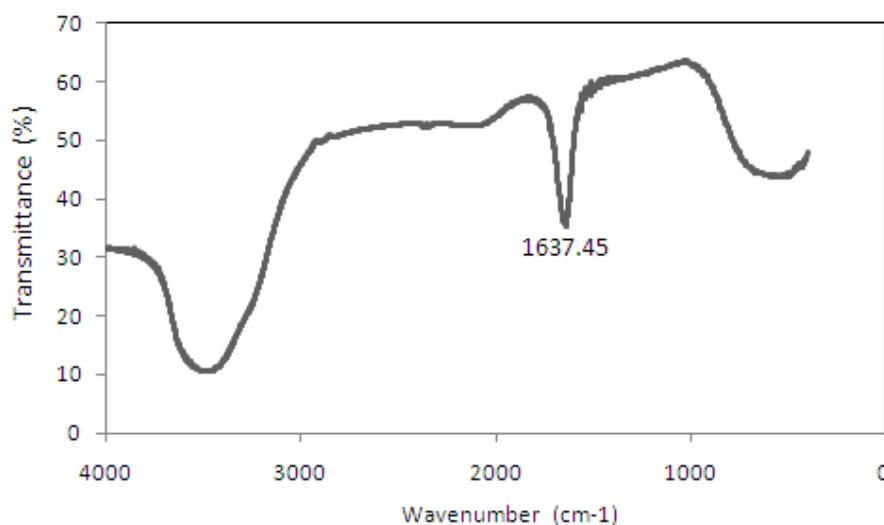

Fig.10: FTIR spectra of untreated antibiotic aqueous solution
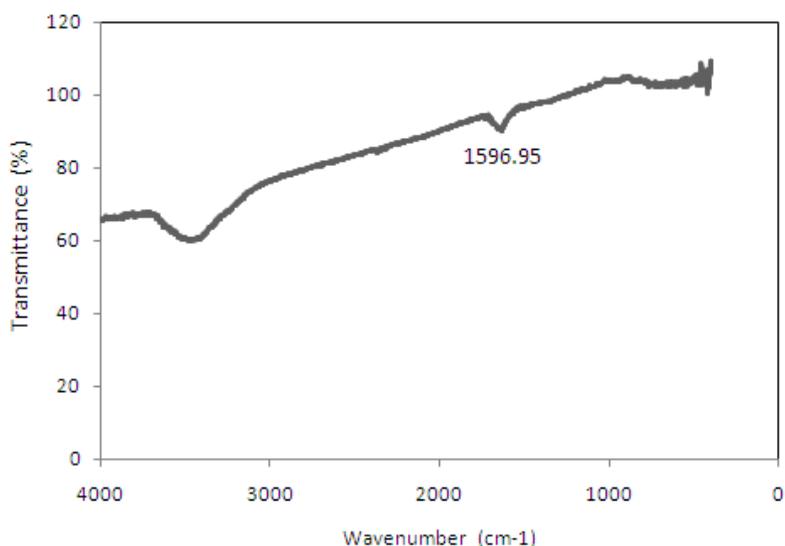
Fig.11: The FTIR spectra of treated antibiotic aqueous solution

In the modified Fenton (FeGAC/H_2O_2) treatment under the optimum operating conditions, biodegradability (BOD_5/COD ratio) of the antibiotic aqueous solution improved from zero (untreated) to 0.36 (treated), indicating that the effluent was amenable to biological treatment (Al-Momani *et al.*, 2002). A similar biodegradability improvement (zero to 0.37 and 0.40) was also observed in the Fenton and photo-Fenton treatment of the antibiotics amoxicillin, ampicillin and cloxacillin in the aqueous solution (Elmolla & Chaudhuri, 2009a; 2009b).

CONCLUSION

The optimum operating conditions of the modified Fenton (FeGAC/H_2O_2) treatment of the antibiotics amoxicillin and cloxacillin in aqueous solution at pH3 were H_2O_2/COD molar ratio 2.0, FeGAC dose 3.5 g/L and reaction time of 90 min for 87.53% removal of COD, 78.01% removal of TOC and 98.24% removal of NH_3-N. The biodegradability (BOD_5/COD ratio) of the antibiotic aqueous solution improved from zero to 0.36. Meanwhile, FTIR spectra indicated degradation of the antibiotics. Hence, the modified Fenton (FeGAC/H_2O_2) process could be an alternative in the pretreatment of the amoxicillin and cloxacillin antibiotics in aqueous solution for biological treatment.

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