Optimization and modelling using the response surface methodology (RSM) for ciprofloxacin removal by electrocoagulation

Sibel Barışçi and Ozge Turkay

ABSTRACT

In this study, response surface methodology (RSM) was used to investigate the effects of different operating conditions on the removal of ciprofloxacin (CIP) by the electrocoagulation (EC) with pure iron electrodes. Box-Behnken design was used for the optimization of the EC process and to evaluate the effects and interactions of process variables such as applied current density, process time, initial CIP concentration and pH on the removal of CIP by the EC process. The optimum conditions for maximum CIP removal (86.6%) were found as pH = 4; C0 = 5 mg L⁻¹; I = 4.325 mA cm⁻²; t = 10 min. The model adequacy and the validity of the optimization step were confirmed with additional experiments which were performed under the proposed optimum conditions. The predicted CIP removal as 86.6% was achieved at each experiment by using the optimum conditions. These results specify that the RSM is a useful tool for optimizing the operational conditions for CIP removal by the EC process.

Key words | ciprofloxacin, electrocoagulation, process optimization, RSM

INTRODUCTION

Wide usage of pharmaceuticals has resulted in their detection in wastewater effluents. They are classified as emerging micro-pollutants as the residuals may cause significant adverse effects to the aquatic environment and human health. Antibiotics have special attention among the pharmaceuticals, since bacterial resistance and toxic effects on several organisms have been found not only at high concentrations, but also at low concentrations, resulting in chronic effects (Andreozzi et al. 2004). Quinolone antibiotics are an important type with non-biodegradability featuring among commonly used antibiotics. Ciprofloxacin (CIP) is a second generation fluoroquinolone antibiotic of high use in medicine and veterinary practice. CIP has a high aqueous solubility under various pH conditions and a high stability in soil and wastewaters (Golet et al. 2003).

Given that wastewater treatment plants do not seem to eliminate pharmaceuticals completely, a number of studies (Alaton et al. 2004; Klavarioti et al. 2009; Dimitrakopoulou et al. 2012; Barışçi et al. 2015a, 2015b) have been carried out to determine the most suitable technologies to treat pharmaceutical residuals from water and wastewater. Electrocoagulation (EC) can be an alternative process to remove emerging pollutants from fresh waters and wastewaters. Studies showed that pharmaceutical residuals have been removed successfully by EC (Martins et al. 2011; Ren et al. 2011; Arsan et al. 2013; Liu et al. 2013). Various electrode types such as iron and aluminium can be used in the EC process. When a current is applied, the electrodes start to dissolve and cations such as Al³⁺, Fe²⁺ and Fe³⁺ are produced. These cations then form metal hydroxides (Al(OH)₃, Fe(OH)₂ and Fe(OH)₃, etc.) in water. Metal hydroxide species are effective for destabilization of pollutants. Different mechanisms such as double layer compression, adsorption, charge neutralization and sweep coagulation can take place in the EC process. The mechanism of EC strongly depends on parameters such as pH, current density and conductivity (Yılmaz et al. 2005; Daneshvar et al. 2006; İrdemez et al. 2006; Arroyo et al. 2009).

Response surface methodology (RSM) is a mathematical and statistical technique for determining the effects of several independent variables on processes. It can be used also for evaluation of interaction between individual variables. Recent studies have shown that RSM could be useful for optimization of the factors influenced on the chemical processes (Baş & Boyacı 2007).

In this study, degradation of CIP by EC was investigated and the effects of process parameters on the mechanism were
evaluated with RSM. A Box-Behnken design was used for optimization of the process according to ultraviolet (UV) absorbance (UV_{abs}) reduction for the key parameters such as pH, current density (I_d), initial CIP concentration (C_0) and process time (t_{process}).

**EXPERIMENTAL**

**Chemicals**

CIP (namely, 1-cyclopropyl-6-fluoro-1,4-dihydro-4-oxo-7-(piperazin-1-yl)quinoline-3-carboxylic Acid) was purchased from Sigma-Aldrich (≥98%, HPLC grade). The buffers used in this study for pH adjustment included: C8H5KO4-HCl solution (pH 4); KH2PO4-NaOH solution (pH 6.5) and Na2B4O7·10H2O-NaOH solution (pH 9). Stock solutions for the treatment study (100 mg L⁻¹ of CIP) were prepared weekly in high quality pure water using the Millipore Water Purification System and stored at 4 °C.

**Electrocoagulation system and procedures**

The EC process experiments were conducted in a batch system. A plexiglass reactor with the dimensions of 12 × 7 × 5 cm was used in the EC process. The EC experiments were conducted using high purity iron electrodes with the dimensions of 10 × 5 × 0.2 cm which were supplied by Artı Makina, Turkey. The gap between electrodes was 2 mm and they were connected to a digital power supply (GW Instek PSP-405 Programmable DC) as a monopolar connection mode. Currents were held constant for each run. 300 mL of CIP solution was placed into the EC reactor in each run. The solution was stirred with a magnetic stirrer at a speed of 150 rpm during operating time. The samples were taken from the EC reactor at the end of operating time then the suspension was allowed to settle for 30 min. The samples were filtered through 0.45 μm pore size membrane before UV/VIS analysis. Prior to the experiments, electrodes were washed with 0.5 N H2SO4 solutions for 2 min and rinsed with deionized water, then dried in the drying-oven and placed in a desiccator to cool down. The electrodes were washed with 0.5 N H2SO4 solutions for 2 min and rinsed with deionized water, then dried in the drying-oven and placed in a desiccator to cool down.

The samples were filtered through 0.45 μm pore size membrane before UV/VIS analysis. Prior to the experiments, electrodes were washed with 0.5 N H2SO4 solutions for 2 min and rinsed with deionized water, then dried in the drying-oven and placed in a desiccator to cool down. The same process was also applied after each experiment.

The active total surface area (S_{total}) of the electrodes was 81.25 cm². Current densities were calculated for each applied current by dividing the applied current by S_{total}. Different current densities were applied to see the effect on UV_{abs} reduction.

**Analytical methods**

The equilibrium concentrations of CIP were measured by HACH Lange DR 5,000 UV-VIS spectrophotometer at a pre-determined wave length of 272 nm. First, the calibration curve was established by nine standards which were chosen for gaining better linearity in the range of 1–50 mg/L. The coefficients of regression (R²) was 0.997 and the mean linear regression equation was determined as \( y = 0.0941x + 0.1186 \). This method was applied according to the previous studies (Bhalerao & Rote 2012; Jiang et al. 2013). The zeta potential (ZP) measurements of the particles in suspensions were determined from their electrophoretic mobility based on the Smoluchowki model using a Malvern Zetasizer Nano-ZS (Malvern, USA). The cell was washed with deionized water and ethanol before taking the measurements.

**Mathematical and statistical procedures**

In the present study, a Box-Behnken experimental design was employed to evaluate four selected factors (A: pH, B: initial CIP concentration, C: current density and D: process time) for the optimization of CIP degradation by EC process according to the response of UV_{abs} reduction.

The removal efficiencies were fitted to a general function indicating the interaction between dependent and independent variables using second-order polynomial equation. The employed model of the second order polynomial is:

\[
R = \beta_0 + \sum \beta_i X_i + \sum \beta_{i2} X_i^2 + \sum \beta_{ij} X_i X_j
\]  

(1)

where R is the predicted response, \( X_i \) and \( X_j \) are independent factors, \( \beta_0 \) is the intercept, \( \beta_i \) is the linear coefficient, \( \beta_{ij} \) is the interaction coefficient.

Different levels were chosen for each parameter to determine the influence of selected parameters on the process efficiency for CIP removal. The selection of operating levels is summarized in Table 1.

<table>
<thead>
<tr>
<th>pH</th>
<th>I_d (mA cm⁻²)</th>
<th>C_0 (mg L⁻¹)</th>
<th>t (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low</td>
<td>High</td>
<td>Low</td>
<td>High</td>
</tr>
<tr>
<td>(-1)</td>
<td>(+1)</td>
<td>(-1)</td>
<td>(+1)</td>
</tr>
<tr>
<td>4</td>
<td>9</td>
<td>2</td>
<td>6</td>
</tr>
</tbody>
</table>
Design-Expert, version 8.0.4.1 (STAT-EASE Inc., Minneapolis, USA) was used for the design of experimental parameters and statistical analysis of obtained data. The analysis of variance (ANOVA) was used to predict the statistical parameters. The design of experiments and the responses for each factor can be seen in Table 2.

Although initial CIP concentrations seem to be higher than they are found in surface waters and wastewater ef- fluents, its presence in antibiotic manufacturing ef- fluents may reach the mg/L level. There are many reported cases in the literature where it was found to be realistic to conduct investigations with high concentrations (Arslan-Alaton & Caglayan 2005; Brillas et al. 2005; Ciríaco et al. 2009; González et al. 2011; Dimitrakopoulou et al. 2012). In this study, we have chosen mg/L levels due to the planning of real scale applications for manufacturer ef- fluents. Besides this, these concentration ranges were chosen to allow for assessment of the statistical analysis model employed and correct determination of residual CIP with the analytical methods adopted.

| Table 2 | Experimental planning and responses obtained in the experiments |
| Run | Actual pH | Actual $C_0$ (mg.L$^{-1}$) | Actual $I_d$ (mA.cm$^{-2}$) | Actual $t_{process}$ (min) | R, CIP degradation (%) |
| 1 | 4.00 | 15.00 | 4.00 | 10.00 | 52.9 |
| 2 | 6.50 | 25.00 | 4.00 | 10.00 | 44.2 |
| 3 | 6.50 | 25.00 | 6.00 | 20.00 | 44.5 |
| 4 | 6.50 | 15.00 | 4.00 | 20.00 | 64.1 |
| 5 | 4.00 | 25.00 | 4.00 | 20.00 | 51.8 |
| 6 | 9.00 | 15.00 | 4.00 | 10.00 | 45.0 |
| 7 | 6.50 | 15.00 | 6.00 | 10.00 | 51.1 |
| 8 | 6.50 | 15.00 | 6.00 | 30.00 | 86.6 |
| 9 | 6.50 | 25.00 | 2.00 | 20.00 | 44.9 |
| 10 | 9.00 | 15.00 | 2.00 | 20.00 | 37.6 |
| 11 | 9.00 | 5.00 | 4.00 | 20.00 | 86.1 |
| 12 | 4.00 | 15.00 | 6.00 | 20.00 | 77.8 |
| 13 | 6.50 | 25.00 | 4.00 | 30.00 | 65.8 |
| 14 | 6.50 | 15.00 | 2.00 | 30.00 | 40.0 |
| 15 | 6.50 | 15.00 | 4.00 | 30.00 | 57.7 |
| 16 | 9.00 | 15.00 | 6.00 | 20.00 | 55.7 |
| 17 | 9.00 | 15.00 | 4.00 | 30.00 | 62.1 |
| 18 | 6.50 | 5.00 | 4.00 | 30.00 | 73.7 |
| 19 | 4.00 | 15.00 | 2.00 | 20.00 | 43.6 |
| 20 | 6.50 | 15.00 | 2.00 | 10.00 | 36.5 |
| 21 | 6.50 | 15.00 | 4.00 | 20.00 | 60.5 |
| 22 | 6.50 | 5.00 | 4.00 | 20.00 | 70.4 |
| 23 | 6.50 | 5.00 | 2.00 | 20.00 | 50.4 |
| 24 | 6.50 | 15.00 | 4.00 | 20.00 | 53.8 |
| 25 | 4.00 | 5.00 | 4.00 | 20.00 | 79.3 |
| 26 | 6.50 | 5.00 | 4.00 | 10.00 | 80.3 |
| 27 | 6.50 | 15.00 | 4.00 | 20.00 | 58.4 |
| 28 | 6.50 | 15.00 | 4.00 | 20.00 | 67.3 |
| 29 | 9.00 | 25.00 | 4.00 | 20.00 | 65.8 |

The model F-ratio (Fisher variation ratio), probability value (Prob > F) and adequate precision are the main pointers presenting the significance and acceptability of the employed model. According to the ANOVA report (see Table 3), the model F-ratio of 9.47 indicate the model is significant and value of ‘Prob > F’ < 0.0500 indicate that the employed model is significant.

Furthermore, the ANOVA analysis indicates the significance of each parameter and also the interaction between variables. The significance of the interaction between variables indicates that these factors have synergetic effect on the responses. In this case; B, C, D and C$^2$ are significant model terms as seen in Table 3.

| Table 3 | ANOVA results for response surface quadratic model |
| Factor | Sum of squares | F-ratio | p-value (Prob > F) |
| Model | 4696.27 | 9.47 | <0.0001 (significant) |
| A: pH | 24.37 | 0.39 | 0.5377 |
| B: $C_0$ | 1264.85 | 20.41 | 0.0002 |
| C: $I_d$ | 1476.30 | 23.83 | <0.0001 |
| D: $t_{process}$ | 563.07 | 9.09 | 0.0068 |
| BD | 198.81 | 3.21 | 0.0884 |
| CD | 256.00 | 4.13 | 0.0556 |
| B$^2$ | 211.84 | 3.42 | 0.0795 |
| C$^2$ | 587.04 | 9.47 | 0.0059 |
| Residual | 1239.23 | | |
| Lack of fit | 1151.24 | 2.62 | 0.1816 (not significant) |
| Pure error | 107.99 | | |
| Cor total | 5935.51 | | |
The regression factors \( R^2 \) and adjusted-\( R^2 \) were equal to 0.7912 and 0.7077, respectively. The predicted-\( R^2 \) of 0.5420 was in reasonable agreement with the adjusted-\( R^2 \) of 0.7077 as the difference between them was <0.2. When second order polynomial model is considered (Equation (1)), the following expressions were obtained according to RSM:

\[
UV_{\text{abs,removal}}(\%) = +63.24 - 10.27 \times C_o + 11.09 \times I_d + 6.85 \times t_{\text{process}} - 9.98 \times I_d^2
\]  

\( (2) \)

**Graphical analyses**

Figure 1 shows the perturbation plot of the model. The X axis signifies changes of the selected factors from minimum values to maximum values of levels. The proportional UV\(_{\text{abs}}\) removal was presented with the Y axis. The individual effects of each factor can be discussed according to Figure 1. The change of pH (A) did not affect the EC process compared to the other parameters. Therefore, it did not take place in Equation (2). The change of ZP according to pH can be seen in Figure 2. The ZP values showed a similar trend at the different pH values as seen in Figure 2. This information confirms that the change of pH did not affect CIP removal by EC. The ZP value at the beginning was measured as –13.38 mV. It reached to positive values (2.71, 5.71 and 3.99 for pH 4, 6.5 and 9, respectively) at 10 min which indicates that the solution went through rapid coagulation or flocculation according to ZP test methods (D4187-82 ASTM Zeta Potential of Colloids in Water and Wastewater). The ZP values then closed to zero with increasing process time.

Removal efficiency decreased with increasing initial CIP concentration (B) until the reference point and the efficiency was almost stable after the reference point. This means that the increase on initial CIP concentration until a certain point did not affect the EC process effectiveness.

Current density (C) was a highly effective parameter on CIP removal by the EC process according to Figure 1. When applied, current density increased to the reference point and the removal efficiency increased. After the reference point, the efficiency continued to increase but then there was a slight decrease on the efficiency. In all electrochemical processes, the current density is the most important parameter for controlling the reaction rate. The current density shows a strong effect on the EC process, especially on the kinetics of removal and shortening the treatment time. High current density affects the amount of anodic dissolution of iron and this provides a greater amount of precipitate for the removal of pollutants. Additionally, the bubble generation rate increases with the increasing current density which is beneficial for high pollutant removal via \( \text{H}_2 \text{flotation} \).

High removal rates of CIP were observed with increasing process time (D). A further increase of process time to the maximum point (+1) still provided high removal efficiency (see Figure 1).

According to ANOVA analysis, all the interaction terms including BD and CD were insignificant terms. However, UV removal efficiency was evaluated within these parameters. The contour plots of the quadratic model with two variables kept constant and the other two varying within the experimental ranges are shown in Figures 3–5. In Figure 3, the contour plot was developed as a function of...
of initial CIP concentration and applied current density, while the process time and pH were kept constant at 30 min and 6.5, respectively. 80% UV\textsubscript{abs} removal efficiency was gained at 30 min process time and pH 6.5, while applied current density was above 4.5 A and initial CIP concentration was almost between 5 and 10 mg.L\textsuperscript{-1}. As can be seen from Figure 3, the influence of these two individual variables on CIP removal efficiency was significant for higher values of applied current density and lower initial antibiotic concentration studied.

Figure 4 shows the relationship between initial CIP concentration and the EC process time on UV\textsubscript{abs} removal efficiency. In this case, current density and pH were kept constant. As seen in Figure 4, a longer EC process time provided higher efficiencies. Also, higher removal efficiencies were obtained with lower initial CIP concentrations, as indicated before. However, it does not mean that two parameters affect the removal efficiency synergistically, as is also understood from the discrete lines in Figure 4.

The contour plot for the variables of current density and EC process time is shown in Figure 5. Initial CIP concentration was 25 mg.L\textsuperscript{-1} and pH was 6.5. As seen from Figure 5, increasing the process time and the current density provided higher removal rates individually.

In order to achieve a high level of removal efficiencies (>70%), applied current density values become relatively important for application times >26 min. The interrelation between the variables, therefore, is important in terms of process optimization.

**Optimization of the electrocoagulation process for CIP removal**

The main objective of the optimization is to define the optimum values of variables for CIP removal through the EC process from the model obtained using experimental data. In the optimization step, the desired goal for the response (UV\textsubscript{abs} removal efficiency) was chosen to a maximum value of 86.6% and the variables of applied current density, process time, initial CIP concentration and pH were selected to be within range. The optimization results of the process variables for CIP removal are shown in overlay plots (see Figure 6).

The Design-Expert version 8.0.4.1 software programme found 100 solutions to achieve optimum conditions (the
The highest removal efficiency provided by the EC process (for CIP removal with high desirability range of 0.897–0.924). For instance, one of the solutions for optimal removal conditions was found as pH = 4; C₀ = 5 mg.L⁻¹; t = 4.325 mA.cm²⁻¹; t_process = 10 min (86.624% removal efficiency). The overlay plots for the optimal regions are presented in Figure 6. The yellow portions gave the allowable values of the two variables by maximizing removal efficiencies of CIP. According to Figure 6, when the process was conducted at pH 4 and initial CIP concentration of 5 mg.L⁻¹, applied current density and process time should be 4.24 mA.cm⁻² and at least 16 min, respectively, to maximize the CIP removal.

To confirm the model adequacy and the validity of the optimization step, further experiments were performed under the proposed optimum conditions based on the results obtained from the model. The predicted 86.6% CIP removal was achieved at each experiment by using the optimum conditions (data not shown). These results testify that the RSM is a powerful tool for optimizing the operational conditions for CIP removal by the EC process.

**CONCLUSIONS**

In the present study, the performance of EC process for CIP removal was studied focusing on the impact of operating parameters such as current density, process time, initial antibiotic concentration and pH by using RSM with Box-Behnken design. The results obtained from the experiments showed that RSM was a suitable method to optimize the operating conditions of EC for CIP removal. ANOVA showed a high correlation factor (R² = 0.7912), confirming an acceptable adjustment of the second-order regression model with the experimental data. Optimal removal conditions for 86.6% removal efficiency were found as pH = 4; C₀ = 5 mg.L⁻¹; I_d = 4.325 mA.cm²⁻¹; t_process = 10 min. The results of the study indicate that EC is an effective method for CIP removal.

**REFERENCES**


First received 16 June 2015; accepted in revised form 10 December 2015. Available online 28 December 2015