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ORIGINAL PAPER

The optimization of Cu and Fe bioleaching from converter slag using *Acidithiobacilus ferrooxidans*

Saeb Ahmadi[1], **Mohsen Vafaie Sefti**[1,✉], **Mohammad Mahdi Shadman**[1], **Zahra Azimi Dijvejin**[1], **Hoshyar Hossini**[2]

1. Chemical Engineering Department, Tarbiat Modares University, Tehran, Iran
2. Department of Environmental Health Engineering, Faculty of Health, Kermanshah University of Medical Sciences, Kermanshah, Iran

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ABSTRACT

The main objective of this work was to assess the optimization of Fe and Cu bioleaching from converter slag using *Acidithiobacillus ferrooxidans*. Important parameters that contribute to the bioleaching process include initial pH, initial Fe^{2+} concentration and pulp density. In order to optimize these parameters, the response surface methodology (RSM) was applied. The maximum simultaneous Fe and Cu recovery yields were 95% and 100%, respectively. The optimum conditions were initial pH 1.8, initial density 1.4 g/100 mL and initial Fe^{2+} 7.3 g/L. The comparison between chemical leaching and bioleaching results showed that bioleaching improved the recovery yields of Fe and Cu by 26% and 33%, respectively. The modified shrinking core model was used to determine the rate-limiting step of the process. It was found that diffusion through the product layer and chemical reaction are the rate controlling steps.

Keywords: Bioleaching; Converter slag; *Acidithiobacillus ferrooxidans*; Optimization; Kinetic

Introduction

Ash generated in industrial operations from burning fossil fuels, such as oil and coal, is a major environmental issue. Improper disposals cause problems, such as leakage of acidic liquids, dusting and pollution of ground water with heavy metals.[1,2] Steelmaking operations generate large volumes of converter slag that contain toxic heavy metal elements. Many researchers have been trying to recover valuable metals, such as Cu and Fe, from slag.[3,4]

The classical methods for metal extraction include pyrometallurgy and hydrometallurgy. They encounter problems, such as serious environment pollution, low recovery yields and high operational costs.[5,6] In order to decrease the cost of the leaching process, some studies have considered using a combination of precipitants for heavy metal removal.[7] Sadat et al used the Chemorex CP-150 to leach Cu from dilute Cu-Ni-Co bioleach solution. They found that Cu can extract above 90% using Chemorex CP-150.[8] Bioleaching offers an attractive alternative. It is based on the ability of microorganisms for the recovery of metals with many advantages, such as simplicity, low capital cost and less environmental hazard.[9,10]

Acidithiobacilus ferrooxidans (At. ferrooxidans) is a Gram-negative, mesophilic and iron-oxidizing proteobacterium that can transform non-soluble heavy and precious metals into soluble forms using following reactions:[11,12]

$$Fe^{2+}+\frac{1}{4}O_2+H^+ \xrightarrow{A.\ ferrooxidans} Fe^{3+}+\frac{1}{2}H_2O \qquad (1)$$

$$M + 2F^{3+} \rightarrow M^{2+} + 2F^{2+} \qquad (2)$$

where M is an insoluble metal in the slag.

To optimize the bioleaching process in laboratory testing, a statistical method is needed.[13] Conventional methods for optimization are one factor at a time, i.e., changing one independent variable at a time while the other variables remain fixed.[14]

✉ *Mohsen Vafaie Sefti*
vafaiesm@modares.ac.ir

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Statistical optimization is a better method that evaluates the effects of factors and the interactions among the factors, while greatly reducing the total number of tests needed to build models and search for the optimum conditions.[15,16] The response surface methodology (RSM) is based on the statistical optimization in which the optimal conditions of a multivariable system are determined.[17] In this work, Cu and Fe bioleaching using *At. ferrooxidans* was optimized using RSM according to examination of three main factors, including initial pH, initial Fe^{2+} concentration and pulp density. Additionally the kinetics of bioleaching was determined using the modified shrinking core model.

Materials and Methods

Slag sample

The converter slag used in this study was collected from a disposal site of waste materials of the Esfahan Steel Company, Iran. It was ground and passed through an No. 200 sieve to yield ash particle sizes less than 75 µm. This slag ash was used throughout this work. X-ray fluorescence (XRF) (Philips PW2404, *Netherlands*) was used to analyze the chemical composition in the slag ash samples, which is shown in Table 1. All chemical reagents were analytical grade, and all aqueous solutions were prepared using distilled water.

Table 1. Chemical composition of the converter slag sample

Element	CuO	Ni	Fe_2O_3	CaO	MgO	CoO	Al_2O_3	SiO_2	LIO
Value (%)	4.2	1.98	38.8	4.01	2.6	0.48	0.08	34.3	13.55

Microorganism and culture medium

At. ferrooxidans (PTCC 1626) was provided by the Iranian Research Organization for Science and Technology (IROST), Tehran, Iran. To grow the bacterium, 50 mL of 9 K medium was added in each 250 mL Erlenmeyer flask. The culture medium (initial pH 2) contained 3 g/L $(NH_4)_2SO_4$, 0.5 g/L $MgSO_4.7H_2O$, 0.5 g/L $K_2HPO_4.3H_2O$, 0.1 g/L KCl, 0.01 $Ca(NO_3)_2$ and 44.22 g/L $FeSO_4.7H_2O$. The flasks were shaken in an orbital shaker at 150 rpm at 32 °C. The inoculum size was 10% (v/v) throughout this work.

Analytical methods

The pH and reduction potential (Eh) of the medium were monitored during the bioleaching process using a portable pH/Eh meter ((Model 713, Metrohm, Switzerland). The bacterial cell count in the liquid phase was obtained using a hemocytometer under a phase-contrast microscope at 40X (Zeiss Standard 25, Germany). Fe^{2+} concentration was measured with a spectrophotometer at a wavelength of 500 nm using 5-sulfosalicylic acid as an indicator.[18] Atomic absorption spectrometry was used to analyze the metals after filtering a solution through Whatman No. 42 filter paper (2.5 µm).

Experimental design and optimization

The experimental design of the bioleaching study was performed using Design Expert 7.1.4 software. The central composite design (CCD), based on RSM, was used extensively to produce a polynomial model. According to the CCD method, the total number of experiments was obtained using 2k + n_α + n_0, where k is the number of independent variables, n_α is the number of axial points and n_0 is the number of center points.[15,16] In this work, there were three variables of interest: initial pH, initial Fe^{2+} concentration and pulp density; and the number of center points was 6, which resulted in a total of 20 runs. The behavior of the system is explained by the polynomial empirical model below,

$$y = \beta_0 + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k} \beta_{ii} X_i^2 + \sum_{i=1}^{k} \sum_{j=1}^{k} \beta_{ij} X_i X_j + \varepsilon \qquad (3)$$

where y is the expected value of the response variable, β_0, β_{ii}, β_i are the model parameters, X_i and X_j are the coded factors evaluated. In this study, y represents the Cu or Fe recovery yield. The variables X_i were coded as x_i according to the following equation:[19]

$$x_i = \frac{X_i - X_0}{\Delta X} \qquad i=1.2.3.\ldots.k \qquad (4)$$

where x_i is a dimensionless coded value, X_i is the real value, X_0 is the value of X_i at the center point and ΔX_i is the unit change of the real value of the variable *i*. Each factor was varied at five different levels based on the CCD as shown in Table 2.

The analysis of variance (ANOVA) of the polynomial model was carried out to evaluate the significant parameters in this study. In order to validate optimum conditions, an experiment at the obtained optimum conditions using RSM was performed and the results were compared with model-predicted results.

Table 2. Levels of the variables used for experimental design

Factor	Code	Unit	Low axial (-α)	Low factorial (-1)	Centre point (0)	High factorial (+1)	High axial (+α)
Initial pH	A	-	1.5	1.8	2.2	2.7	3
Initial density	B	g/l00mL	0.5	1.4	2.75	4.0	5
Initial [Fe^{2+}]	C	g/L	0.5	2.2	4.7	7.2	9

Rate-controlling model

The bioleaching process has four sequential steps, including 1) the diffusion of the attacking chemical species (e.g., H^+ and Fe^{3+}) from the bulk solution to the reactant inside a solid (ash in this work); 2) the diffusion of the reactants through the solid; 3) chemical reaction; and 4) the transfer of the product species to the bulk solution.6 Because of the vigorous shaking condition in the bioleaching process, the first and last steps are usually not rate limiting. Due to pH variation over the process, the ferric ion (bioleaching agent) concentration varied. The modified shrinking core model for determination of diffusion Eq. (5) and chemical reaction Eq. (6) control were used, which showed in the following:[6]

$$E(X_B)=1-\frac{2}{3}X(t)-(1-X(t))^{2/3} \tag{5}$$

$$G(X_B)=1-(1-X_B(t))^{1/3} \tag{6}$$

where XB is the fractional conversion (meal recovery yield), t is the actual time and X (t) is the conversion of metal leached from convertor slag.

Results and Discution

Bacterial adaptation

Due to toxicity of the converter slag, adaptation of bacterium was done before bioleaching. Adaptation was done by step-wise increase of the slag ash concentration in the culture medium in a serial sub-culturing process.[20] Adaptation began by adding 0.5 g/100 mL of ash into the medium and continued until 5 g/100 mL. At higher concentrations (> 5 g/100 mL), the bacterial cell count decreased to < 10^6 cells/mL, indicating that the bacteria cannot tolerate the higher 5 g/100 mL of ash concentration. This adapted bacterium was chosen as the working cell culture for the remaining experiments.

pH and Eh variations

The pH and Eh variations with time at the ash concentration of 4% (w/v) (i.e., 4 g/100 mL) are shown in Fig. 1. After one day, the pH increased to 2.5 and the bacterial count decreased to 10^6 cells/mL. In the second day, solution pH started to decrease and reached 1.3 at the end of the 9-day bioleaching process. The bacterial count had a rapid increase and reached 10^9 cells/mL, which was accompanied by increased Eh from 340 mV to 550 mV.

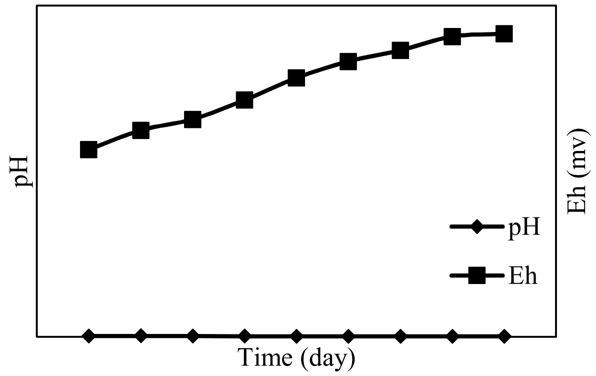

Fig. 1. pH-Eh variation in different time

Statistical analysis

The bioleaching experimental conditions and recovery yields are shown in Table 3. Analysis of variance (ANOVA)was used to analyze each response, including Fe and Cu recovery yields, as shown in Table 4. The quadratic model was used for Fe and Cu recovery yields, respectively. Results showed that the p-value for each model, including Fe and Cu recovery yields, were less than 0.05, meaning that the models were statistically significant with 95% confidence level. The R^2 values for Fe and Cu recovery yields were 0.96 and 0.92, respectively, suggesting that the models are a good fit with experimental data. The coefficient of variation (CV) is defined as the ratio of the standard deviation to the mean that measures the reproducibility of the model. If CV is less than 10%, a model can be considered reasonably reproducible.[19] The results in Table 4 show that both models had CV values less than 10%.[11] The adequate precision (signal to noise ratio) is desirable if it is greater than 4. In this work, it was 21.2 for Fe recovery and 16.7 for Cu recovery (Table 4), both much higher than 4.

Table 3. CCD design and results

Run	Initial pH	Initial ash density (g/100 mL)	Initial [Fe^{2+}] (g/L)	Fe Recovery (%)	Cu Recovery (%)
1	2.3	5.00	4.75	98	41
2	2.3	2.75	4.75	77	76
3	1.5	2.75	4.75	81	88
4	1.8	1.41	7.28	79	85
5	2.3	0.50	4.75	93	98
6	2.3	2.75	4.75	76	78
7	1.8	1.41	2.22	68	64
8	2.7	1.41	2.22	65	58
9	1.8	4.09	2.22	87	51
10	2.3	2.75	0.50	74	61
11	2.3	2.75	9.00	96	81
12	2.7	4.09	2.22	85	59
13	2.3	2.75	4.75	79	79
14	2.7	4.09	7.28	91	66
15	2.7	1.41	7.28	55	76
16	1.8	4.09	7.28	94	62
17	2.3	2.75	4.75	76	78
18	3.0	2.75	4.75	46	32
19	2.3	2.75	4.75	78	77
20	2.3	2.75	4.75	79	76

Table 4. ANOVA for the response models applied

Response	Model	p-value	R-Squared	Adj. R^2-	CV (%)	Adeq. Precision
Fe recovery	Quadratic	< 0.0001	0.96	0.92	4.38	21.2
Cu recovery	Quadratic	< 0.0001	0.94	0.90	6.18	16.7

Fitted statistical Models for Fe and Cu recoveries

The quadratic empirical relationships between Fe and Cu recovery with the variables obtained from Design-Expert 7.1.4 software are showed in the Eqs. (7) and (8):

$$\text{Fe recovery (\%)} = 77.47 - 9.14\,A + 5.70\,B + 2.14\,C + 7.0\,AB - 1.50\,AC + 2.75\,BC - 4.77\,A^2 + 5.13\,B^2 + 4.07C^2 \quad (7)$$

$$\text{Cu recovery (\%)} = 77.41 - 6.70\,A - 11.75\,B + 6.42\,C + 7.50\,AB - 0.50\,AC - 2.25\,BC - 2.57\,A^2 - 1.87\,B^2 - 2.75\,C^2 \quad (8)$$

where A is initial pH, B initial density and C initial Fe^{2+} concentration. Fig. 2 shows the model predicted values versus experimental data. The data points gather around the 45° diagonal line closely, indicating that model predictions matched experimental data very well.

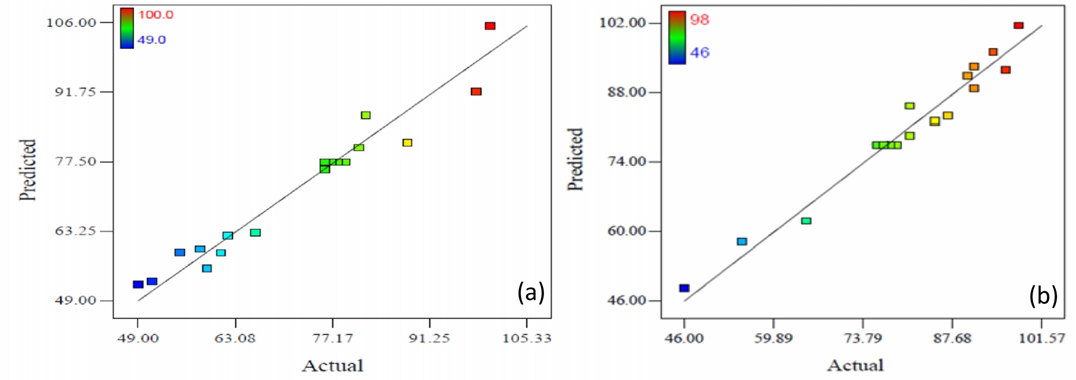

Fig. 2. Actual values vs. predicted values for (a) Fe recovery and (b) Cu recovery

Contour plots in for Fe and Cu recoveries

The bioleaching of Fe

Fig. 3 (a) shows the interaction between initial pH and initial ash density with a constant initial Fe^{2+} concentration of 7.3 g/L. Fig. 3(a) shows that Fe recovery increased from below 64.6% to above 93.3% when the initial pH decreased from 2.7 to 1.80 and the initial pulp density increased from 1.4 g/100 mL to 4 g/100 mL at the same time. Ash sample content Fe, which therefore increased initial density, led to release Fe as a substrate for *At. ferrooxidans*, leading to higher recovery of Fe from the ash.

Fig. 3 (b) shows a combined effect of initial pH and initial Fe^{2+} concentration at a constant initial pulp density of 1.4 g/100 mL. The decreasing initial pH had a positive effect on Fe recovery. *At. ferrooxidans* is an acidophilic bacterium, preferring a lower pH for optimum growth. This led to more proton production from this acid-producing bacterium and therefore increased the recovery of Fe. Increasing the initial Fe^{2+} concentration increased the availability of the substrate for metabolic activities. A high concentration of Fe^{2+} also helped to decrease mass transfer resistance. Fig. 3 shows that the maximum Fe recovery was 89% at the initial pH 1.8 and the initial Fe^{2+} concentration of 6 g/100 mL with an initial pulp density of 1.4 g/100 mL.

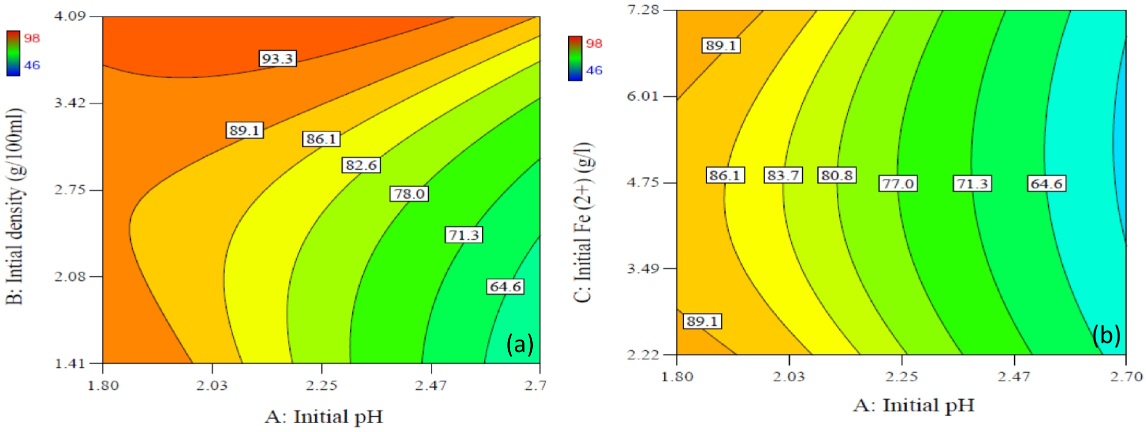

Fig. 3. Contour plots of the interaction effects for Fe recovery: (a) initial pH vs. initial density at a constant initial Fe^{2+} concentration of 7.3 g/L, (b) initial pH vs. initial Fe^{2+} concentration at a constant initial pulp density of 1.4 g/100 mL

The bioleaching of Cu

Fig. 4 shows the two dimensional contour plots between different variables for Cu recovery. According to Fig. 4 (a), the Cu recovery increased when the initial pH decreased and the initial Fe^{2+} concentration increased at a constant initial pulp concentration of 1.4 g/100 mL. This behavior is similar to that observed for Fe recovery in Fig. 3(b). Thus, the same arguments can be used to explain the phenomenon. Cu recovery reached 100% with an initial pH 1.8, an initial Fe^{2+} concentration of 7 g/L, and a pulp concentration of 1.4 g/100 mL. Fig. 4(b) shows interactions between the initial pulp density and the initial Fe^{2+}concentration and their effects on Cu recovery at a constant initial pH of 1.8. According to figure 4(b), decreasing the initial pulp density and increasing the initial Fe^{2+} concentration improved Cu recovery. At low initial pulp densities, oxygen and CO_2 were effectively dissolved and transferred to the bacterial cells, resulting in increased metabolic activities and, therefore, improved Cu recovery.[19] Fig. 4(b) indicates that 100% recovery of Cu was achieved with an initial pulp density of 1.4 g/100 mL and an initial Fe^{2+} concentration of 7 g/L at the constant pH of 1.8.

Hocheng et al. used the culture supernatants of *Acidithiobacillus thiooxidans* (*At. thiooxidans*), *At. ferrooxidans*, and *Aspergillus niger* (*A. niger*) to extract metal from the converter slag ash, and at the best condition, the efficiency was about 30%.[21]

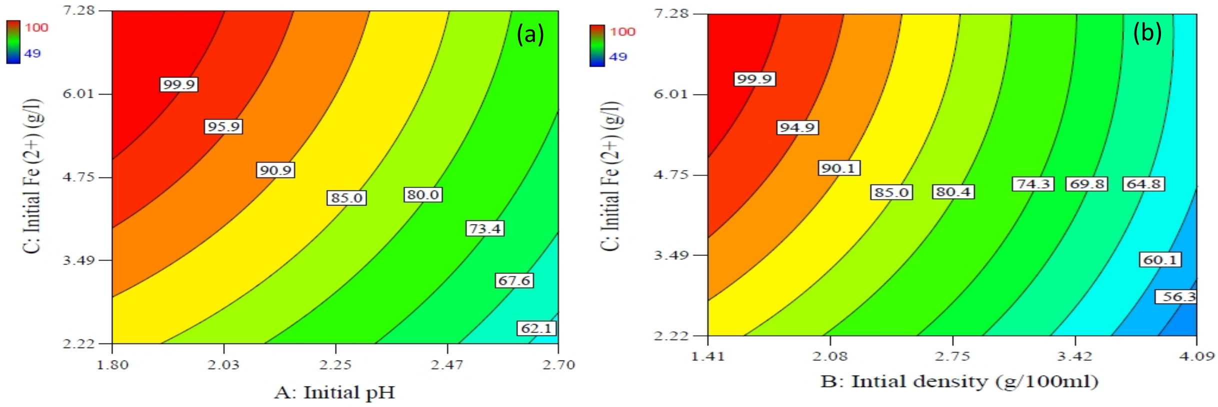

Fig. 4. Contour plots of the interaction effects for Cu recovery: (a) initial Fe^{2+} vs. initial pH at a constant initial pulp concentration of 1.4 g/100 mL, (b) initial Fe^{2+} concentration vs initial pulp density at a constant initial pH of 1.8

Optimization process

Optimization was based on the simultaneous maximization of Fe and Cu recoveries. It should be noted that the goal of optimization is to find a good set of conditions that will meet all of the goals (maximize both of Fe and Cu recoverirs). In numerical optimization, a minimum and maximum level must be provided for each parameter. The optimum conditions proposed by the statistical models in Equations (7) and (8) were initial pH 1.8, initial Fe^{2+} concentration 7.3 g/L and initial pulp density 1.4 g/100 mL, with which maximum recoveries of 100% Fe and 98% Cu were achieved. Panda et al, also at the optimum conditions, successfully extracted approximately 96% of Cu from the converter slag ash using a bioleaching method.[22]

Experimental confirmation

To test the validity of the optimized model conditions, an experiment was carried out. Table 5 shows that the experimental values were in close agreement to the predicted values and a 95% confidence interval (C.I.), and thus, confirming the validity of the model to predict optimal conditions. Under the model-predicted optimized conditions, the experimental recovery results for Fe and Cu were 98% and 100%, respectively, with the aforementioned confidence interval.

Table 5. Experimental confirmation test at the optimum conditions predicted by the models

Response (%)	Target	Predicted (%)	Experimental (%)	95% C.I. Low	95% C.I. High
Fe recovery	Maximize	93	100	87	100
Cu recovery	Maximize	100	98	97	100

Comparison of bioleach with chemical leaching

Chemical leaching of the sample was carried out using H_2SO_4 under the same conditions as abiotic bioleaching using deionized distilled water to make up the desired initial acid concentrations. A comparison between bioleaching and chemical leaching was done, and the results showed that bioleaching produced comparatively better results than chemical leaching (showed in the Fig. 5). The recovery of Fe increased from 74% (chemical leaching) to 100% (bioleaching) and Cu recovery increase from 62% (chemical leaching) to 95% (bioleaching). This was not a surprise, because in bioleaching, spent protons in Eq. (1) were able to be replenished by the acid-producing bacterium, while in chemical leaching, protons did not benefit from this mechanism.

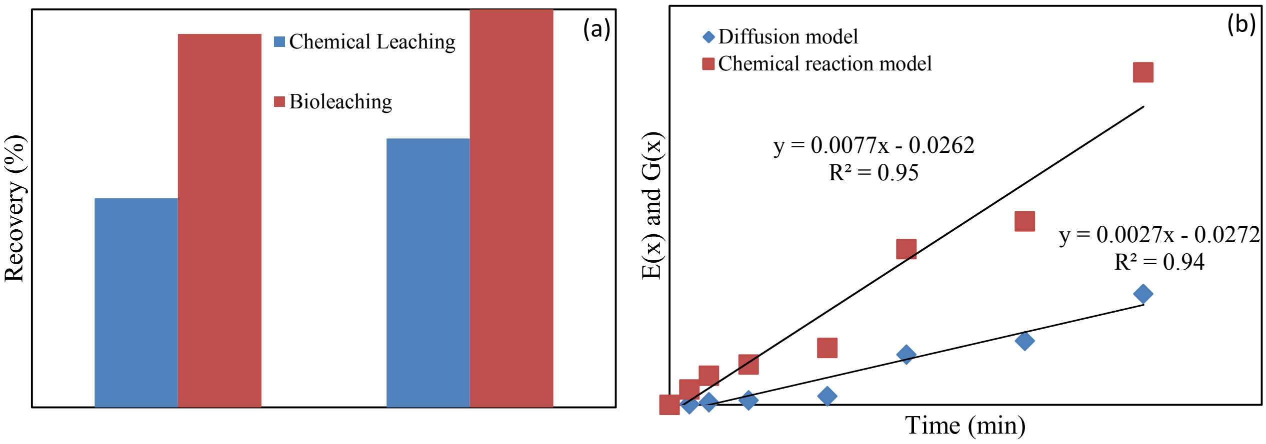

Fig. 5. Comparison between bioleaching and chemical

Determination of rate controlling step

The comparison between correlations expressing diffusion and chemical reaction-controlled regimes for bioleaching of Cu and Fe vs. time is shown in Fig. 6. The diffusion model produced $R^2 = 0.96$ for the Cu recovery and $R^2 = 0.99$ for the Fe recovery, while the chemical reaction had $R^2 = 0.94$ for the Cu recovery and $R^2 = 0.98$ for the Fe recovery. These R^2 values suggest that both models fit the experimental data equally well. Therefore, it is reasonable to argue that both diffusion and chemical reaction were rate controlling for Cu and Fe recoveries during bioleaching. This may be attributed to low porosity of the converter slag ash used in this work.

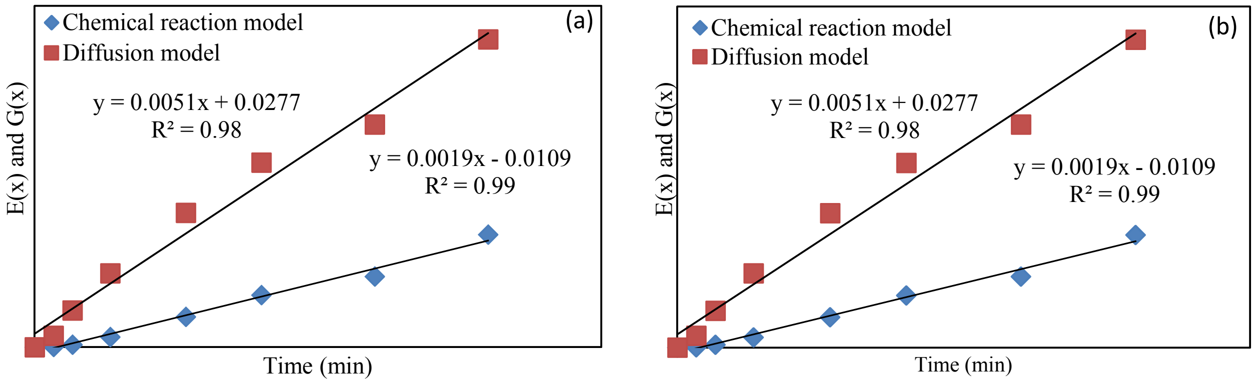

Fig. 6. Comparison between measurements and correlations expressing diffusion and chemical reaction-controlled regimes: (a) for the bioleaching of Cu, (b) for the bioleaching of Fe

Conclusion

Bioleaching of converter slag using *At. ferrooxidans* was investigated. The recovery yields of Fe and Cu were optimized using RSM. Under the optimum conditions with an initial pH = 1.8, an initial density 1.4 g/100 mL and an initial Fe^{2+} 7.3 g/L, bioleaching recovery yields of Fe and Cu reached 95% and 100%, respectively. Modeling of experimental data suggested that both diffusion and chemical reaction were rate controlling in the bioleaching process. The comparison between bioleaching at optimum conditions and abiotic chemical leaching showed that bioleaching improved the recovery of Fe by 26% and the recovery of Cu by 33% due to acid production by *At. ferrooxidans*.

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