BIOLEACHING OF PRINTED CIRCUIT BOARD BY *Acidithiobacillus ferrooxidans*

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ABSTRACT

The present work investigated the influence of Fe^{+2} supplementation on bioleaching process to recover copper from printed circuit board using *Acidithiobacillus ferrooxidans* LR. Printed circuit boards were collected from obsolete computers and mechanically processed through size reduction followed by magnetic separation. The bacteria *Acidithiobacillus ferrooxidans* LR were grown and adapted in presence of printed circuit boards. A shake flask study was carried out on the printed circuit boards samples using a rotary shaker under the following fixed conditions (160 rpm, 30℃). The influence of pH of the medium and concentration of ferrous iron produced were investigated. The bioleaching process efficiency was evaluated by comparison between the concentration in the initial sample and in the medium. Inductively coupled plasma optical emission spectrometry (ICP-OES) was used to determine the metals concentration. The results showed that *Acidithiobacillus ferrooxidans* LR can leach metals from printed circuit boards.

Keywords: bacteria, bioleaching, comminution, recycling, printed circuit boards

1. Introduction

The waste of electric and electronic equipment is generated by discarding of equipment such as obsolete computers and cell phones which is stimulated by the technology innovations and it has been growing each year.

The printed circuit boards are found in almost all electric and electronic equipment being composed of polymers, ceramics and metals and can be an alternative source to recover base and precious metals (Tenório et al., 1997).

The bioleaching can be an alternative method to recover base metals such as copper inside the printed circuit boards (Yang et al.,2009).

The bioleaching works at room temperature and normal atmospheric pressure, not needing high temperature and pressurization, which reduces the energy cost and avoids the emission of gas pollutants (Mousavi et al., 2006).

Advances in knowledge about bioleaching as an economically viable process to recover metals are also attributed to the depletion of high grade ores (Valdívia and Chaves, 2001).

Today, bioleaching is applied on a commercial scale for the recovery of copper and uranium from low levels ores and sulfide minerals (Garcia Jr. et al., 2007; Sepulveda et al., 2010).

Studies (Valdívia and Chaves, 2001; Brandl and Faramarzi, 2006; Olson et al., 2003) also apply bioleaching for the metals extraction such as copper, for subsequent recovery of precious metals like gold, using the bacterial leaching as a pretreatment step.

Bioleaching can be an alternative to conventional methods in the recovery of base metals contained in the printed circuit boards.

Experiments can use percolation columns, leaching columns, shake flasks and bioreactors (Olson et al., 2003).

Solubilization of copper from printed circuit boards using *Acidithiobacillus ferrooxidans* isolated from acid mine drainage can reach 75% copper leaching rates (Rao, 2006).

The mechanism of copper leaching from printed circuit boards by *A. ferroxidans* is similar to bioleaching from metal sulfide. For example, the $Fe_2(SO_4)_3$ formed by *A. ferrooxidans* oxidizes elemental copper contained in the printed circuit board to copper ion resulting in the copper solubilization. The regeneration of $FeSO_4$ suggests that the bioleaching process is cyclic (Choi et al., 2004).

Ilyas et al. (2007) demonstrated that metals can also be recovered from electronic scrap by bacterial leaching using thermophilic bacteria such as *S. thermosulfidooxidans.*

Choi et al. (2004) and Yang et al. (2009) studied the bacterial recovery of copper from printed circuit boards scrap computers using *Acidithiobacillus ferrooxidans* in shake flasks getting copper extraction rates higher than 70%.

2. Objective

The aim of this work was to investigate the influence of Fe^{+2} supplementation on bioleaching process to recover copper from non-magnetic fraction of printed circuit boards of obsolete computers using *Acidithiobacillus ferrooxidans* LR bacteria.

3. Materials and methods

3.1. Printed circuit boards

The printed circuit boards from obsolete computers was comminuted (<2mm) in hammer mill and taken in the magnetic cross-belt separator. Samples obtained through quartering of non-magnetic fraction from magnetic separator were used in bioleaching experiments and analysis of metals' concentration.

To determine the initial metals concentration, the sample was dissolved in aqua regia in 1:20 (Park and Fray, 2009) (1g of printed circuit board sample to 20ml of aqua regia solution). The contact between the printed circuit boards waste and aqua regia was 24 hours, follow by simple filtration with quantitative filter paper. The fraction leached was analyzed by inductively coupled plasma optical emission spectrometry (ICP-OES).

3.2. Cell cultures

The bacteria *Acidithiobacillus ferrooxidans* strain LR isolated from uranium mine effluents (Garcia Jr., 1991) was used in this work.

A. ferrooxidans LR was initially adapted through several subcultures with gradual increase of printed circuit boards concentration (non-magnetic fraction samples) and successive replacement of Fe^{+2}.

The *A. ferrooxidans* LR grew in 200ml T&K medium in 250ml shake sterilized flasks with 15g/l printed circuit boards' samples (non-magnetic fraction) at 30℃ and 180rpm.

3.3. Culture conditions

T&K medium that was adopted for the growth of bacterial inoculum (Tuovinen and Kelly, 1973) was composed from two solutions (A and B), being:

Solution A: $(NH_4)_2SO_4$: 0,625g/l; $MgSO_4.7H_2O$: 0,625g/l e K_2HPO_4: 0,625g/l.

Solução B: 166,5g/l $FeSO_4.7H_2O$

For the preparation of T&K medium, the solutions A and B were prepared, acidified with H_2SO_4 10N to pH 1,8 and sterilized separately.

The solution A was sterilized by autoclaving at 120ºC for 30min and 1atm. The solution B was filter-sterilized (0,45μm). Finally, the solutions A and B were mixed at proportion 4:1 respectively.

3.4. Bioleaching experiments

Bioleaching experiments were carried out in 200ml T&K medium in 250ml shake flasks containing printed circuit boards samples (15g/l) and inoculate with 5% (v/v) *A. ferrooxidans* LR adapted. The cultures were incubated at 160 rpm and 30℃.

The water evaporation was replenished at each sampling with sterile water acidic (pH=1,8) and the medium pH was adjusted with H_2SO_4 10N to 1,8-2,0. All conditions were performed in triplicate.

The experiments evaluated the influence of pH and ferrous ion concentration on copper bioleaching of non-magnetic fraction samples from printed circuit boards in complete T&K medium and T&K medium without Fe^{+2}. The conditions studied were:

- MP: T&K medium + printed circuit boards´ sample (control 1)
- MPBA: T&K medium + printed circuit boards´ sample + adapted bacteria
- AP: Solution A of T&K medium + printed circuit boards´ sample (control 2)
- APBA: Solution A of T&K medium + printed circuit boards´ sample + adapted bacteria

3.5. Analytical methods

For measurements of pH, a bench pH meter was used (an Ag^0/AgCl reference).

Leach samples were periodically withdrawn for chemical analyses (1, 2, 3, 4, 5, 6, 7, 10, 15, 20, 30, 40° days) and centrifug ed during 20min at 5000rpm.

10ml from supernatants was used to determine Fe^{+2} and 5ml was preserved with 1m HNO_3 concentrate at 4℃ for metal analyses (Cu and tota l Fe).

The metals concentration (Cu and Fe_{total}) were determine by inductively coupled plasma optical emission spectroscopy (ICP-OES).

The determination of Fe^{+2} concentrations was performed by titration with potassium dichromate ($K_2Cr_2O_7$). 10ml samples were acidified with 20ml acid mixture (H_2SO_4/H_3PO_4) and diluted to 50 ml. 3 drops of indicator (1% barium diphenylamina sulphonate) were added and the this solution was titrated with potassium dichromate until it turned from green to purple.

The Fe^{+2} concentrations were calculated through consumption of potassium dichromate solution, based in the reaction (1):

$$Cr_2O_7^{-2} + 6Fe^{+2} = 14H^{+} \rightarrow 2Cr^{+3} + 6\,Fe^{+3} + H_2O \qquad (1)$$

The Fe^{+3} concentrations were calculated through difference between Fe_{total} and Fe^{+2} concentrations.

The initial Cu concentration (Initial CC) was used to calculate the copper extraction rate using the following equation (1):

$$\%\ \text{Extraction of copper} = [(\text{Initial CC} - \text{Leach CC}) / \text{Initial CC}] \times 100 \qquad (1)$$

The leach copper concentration (leach CC) was determined in the leach samples taken from bioleaching experiments.

4. Results and discussion

4.1. Printed circuit boards characterization

The non-magnetic fraction (82% w/w) of printed circuit boards containing (w/w) 18,3% copper was used in this work. The elemental analyses (%w/w) of this fraction are showed in Table 1.

Table 1 - Elemental analyses (%w/w) of non-magnetic fraction of printed circuit boards

Non-magnetic fraction									
Metals									Polymers and ceramics
Cu	Sn	Pb	Al	Zn	Fe	Ni	Ag	Au	41,8
18,3	7,8	4,9	4,5	3,9	0,4	0,2	0,1	0,1	

The non-magnetic fraction of printed circuit boards consists of 40% metals and 42% other materials (ceramics and polymers). Copper is the metal of the highest percentage in the printed circuit boards; this is because the printed circuit boards of computers are type FR2, which is a fiberglass layer or cellulose paper and phenolic resin. The surface contains copper layer to connect electrical contacts.

Small amounts (0,2% w/w) of precious metals were also present.

Studies by Tenório et al. (1997), Park & Fray (2009) and Veit et al. (2006) demonstrate that the printed circuit boards composition changes probably due to different methodologies applied in the works or because the composition has changed over time. Ilyas et al.(2007) also suggested that analytical methods and origin of the material can be attributed to this difference.

4.2. Bioleaching experiments

The results of copper extraction are presented in Fig. 2.

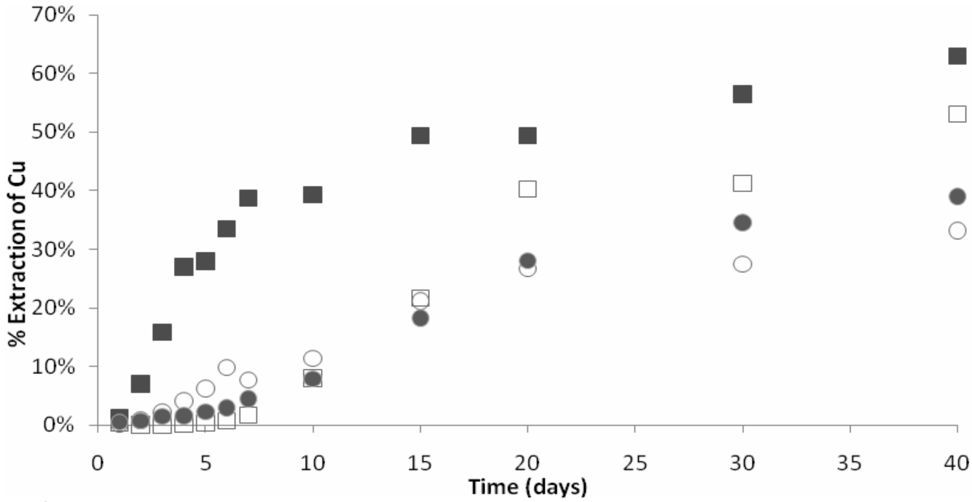

Fig. 1 – Cu^{+2} extraction vs time. Symbols: (□) abiotic control with complete T&K medium and printed circuit boards, (■) adapted *A. ferrooxidans* LR growing with printed circuit boards in complete T&K medium, (○) abiotic control with T&K medium without Fe^{+2} and printed circuit boards, (●) adapted *A. ferrooxidans* LR growing with printed circuit boards in T&K medium without Fe^{+2}.

As can be seen in Fig. 1, the results show that the copper extraction percentage with inoculated complete T&K medium was higher compared to inoculated T&K medium without Fe^{+2}, due the bacterial action is based on Fe^{+2} oxidation to Fe^{+3} which is an oxidant. Similar results were obtained in studies without addition of Fe^{+2} in the bioleaching process using shake flasks (Rohwerder et al., 2003; Xia et al., 2008).

The Fig. 1 also shows that the extraction is more pronounced in the first seven days of experiment in the inoculated complete T&K medium probably due the bacterial growth to be in exponential phase, resulting in exponential increase in the bacterial cells number by increasing the bacterial metabolic activity (oxidation of Fe^{+2} to Fe^{+3}) and consequent copper dissolution, as show in reaction 2:

$$Cu^0 + 2Fe^{+3} \leftrightarrow 2Fe^{+2} + Cu^{+2} \quad (2)$$

From the 8th day, bacterial growth in the inoculated complete T&K medium enters in the stationary phase and the copper extraction becomes less intense because, at this stage, the number of bacterial cells remains constant until the death phase.

With the complete T&K medium, the bacteria leached about 60% more than in medium without iron supplementation. Similar result was obtained by Bevilaqua et al. (2002) in the biooxidation of chalcopyrite, in which the culture of *A. ferrooxidans* with additional iron leached twice as much copper as in the culture without additional iron.

Furthermore, the non-magnetic fraction of printed circuit boards did not have a high iron concentration (0,4% w/w), therefore the largest source of iron for the bacteria was the solution of ferrous sulphate (solution B of T&K medium).

The Fig. 2 shows the changes in pH before the adjustments to the range 1,8 to 2,0.

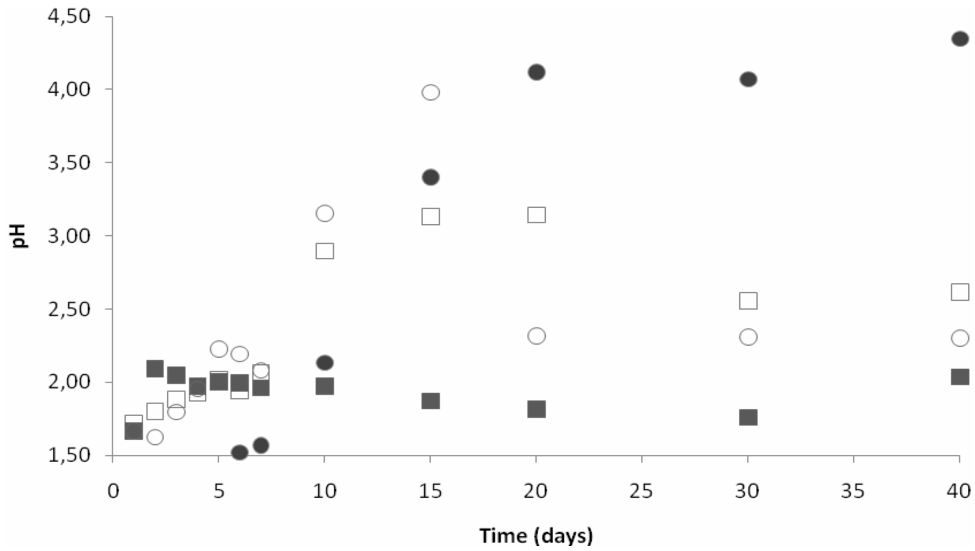

Fig. 2 – Changes in pH vs time. Symbols: (□) abiotic control with complete T&K medium and printed circuit boards, (■) adapted *A. ferrooxidans* LR growing with printed circuit boards in complete T&K medium, (○) abiotic control with T&K medium without Fe^{+2} and printed circuit boards, (●) adapted *A. ferrooxidans* LR growing with printed circuit boards in T&K medium without Fe^{+2}.

In the inoculated T&K medium without Fe^{+2} the pH increased (above 3,5) until the 15th day, suggesting the acid consumption during the oxidation of other metals from printed circuit board, such as Zn, Sn, Al and Pb.

In the control T&K medium without Fe^{+2} there was an increase of pH showing the consumption of acid during the oxidation process of printed circuit boards. Another factor that could explain the higher pH values of control even with the adjustments is the H^+ consumption from metals oxidation which was not reset through the Fe^{+2}/Fe^{+3} cycle.

The pH of inoculated complete T&K medium remained steady between 1,8 and 2,0 after 8th day, possibly due to the combinations of reactions that consume acid, such as chemical and microbiological oxidation of Fe^{+2} and the reactions that produce acid, as the hydrolysis of ferric ion (Yang et al., 2009; Ilyas et al., 2007). After 20 days was observed a pH decrease in both complete T&K medium (control and inoculated) probably because of jarosite precipitation, which produces acid.

The pH of control complete T&K medium presented values higher than inculated (Fig. 2), probably because the consumption of H^+ and O_2 (promoted by shaking) in the Fe^{+2} oxidation, as show in reaction 3:

$$O_2 + 4H^+ + 4\,Fe^{+2} \rightarrow 4\,Fe^{+3} + 2H_2O \qquad (3)$$

This also explains the copper leaching rate in the control complete T&K medium (Fig. 1) by Fe^{+3} produced (Eq. 3).

The Fig. 3 presents the results of changes in Fe^{+2} concentrations.

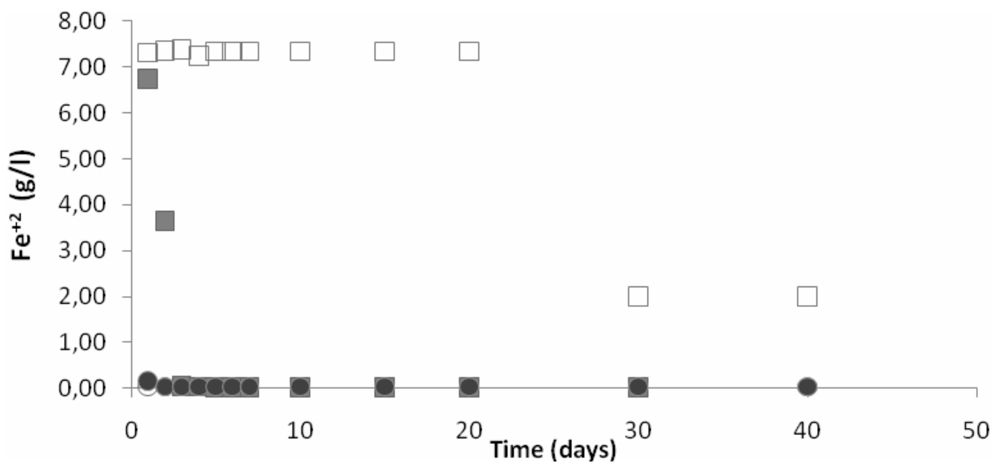

Fig. 3 – Changes in Fe^{+2} concentrations vs time. Symbols: (□) abiotic control with complete T&K medium and printed circuit boards, (■) adapted *A. ferrooxidans* LR growing with printed circuit boards in complete T&K medium, (○) abiotic control with T&K medium without Fe^{+2} and printed circuit boards, (●) adapted *A. ferrooxidans* LR growing with printed circuit boards in T&K medium without Fe^{+2}.

The ferrous iron concentrations obtained in the T&K medium without Fe^{+2} (control and inoculated) were insignificant (Fig. 3)

In the inoculated complete T&K medium was observed consumption of Fe^{+2} until the 10th day indicating that the bacterial activity was intense in this period. Same behavior observed by Bevilaqua et al. (2002) in the chalcopyrite bioleaching.

This behavior occurs due to the exponential phase of bacterial growth because of the increase in the number of bacterial cells while there is Fe^{+2} available.

In copper recovery study from printed circuit board using *A. ferrooxidans*, Choi et al. (2004) concluded that the addition of ferrous ion to the bioleaching process helps to promote the copper dissolution.

In the control complete T&K medium, the Fe^{+2} concentration starts to decline after the 20th day (Fig. 3). Similar behavior was observed by Francisco Jr. et al. (2007) in bacterial leaching of complex mineral sample containing pyrite, pyrrotite and molybdenite, which suggested that this fact may have occurred by natural oxidation of Fe^{+2}.

5. Conclusion

The results demonstrate that addition of Fe^{2+} in the leaching medium increases the percentage of copper extraction in the bioleaching process of non-magnetic fraction of printed circuit boards which strengthens the relevance of oxidation mechanism promoted by Fe^{+3}.

The changes in Fe^{+2} concentrations of inoculated complete T&K medium suggests that exponential phase of bacterial growth occurs until 5º day.

Based on the high values of pH observed in the T&K medium without Fe^{+2} other metals can be oxidized by sulfur acid, consuming H^+ which increase the pH, but analyses of other metals extractions are necessary to comprove this.

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