*Indian Journal of Fundamental and Applied Life Sciences ISSN: 2231– 6345 (Online)*
*An Open Access, Online International Journal Available at www.cibtech.org/sp.ed/jls/2015/01/jls.htm*
*2015 Vol.5 (S1), pp. 2861-2867/Roya et al.*

***Research Article***

# KINETIC STUDY OF BIOLOGICAL RECOVERY OF HEAVY METALS FROM SPENT CATALYSTS USING ACIDITHIOBACILLUS FERROOXIDANS

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## ABSTRACT

This study attempted to conduct a synthetic study on biological recovery of heavy metals from spent catalysts using Acidithiobacillus ferrooxidans. This bacterium produces sulfuric acid and helps recovering metals at the same time. It is easily adapted to growing concentrations of heavy metals under optimized conditions (temperature 30°C, Ph 1.9, pulp density 120 μm, rotation speed 160). This study measured Ph, Eh, and cellular mass, and ferrous and ferric iron concentrations during bioleaching and also bacterium-mediated bioleached content from the spent catalyst. The results revealed that the higher concentrations of spent catalysts could act as a reproduction inhibitor and as a result lower the growth rate of the bacterium. For example, at 200 g/L, bacterium's cell division occurred once in 8 days and the kinetics of growth was 0.093 per day. Bacteria's population rose to $504\times 10^{7}$ from the initial $1\times 10^{7}$ and then fell to $3.6\times 10^{7}$ following the death phase. Reaction rate of culture batch containing 200 mg/L Ni followed 2nd order rate (in the presence of Acidithiobacillus ferrooxidans) and the kinetics of reaction was the slope of the line (0.005). Also, reaction rate of culture batch containing 200 mg/L V followed 1st order rate (in the presence of Acidithiobacillus ferrooxidans) and the kinetics of reaction was the slope of the line (-0.086).

***Keywords:*** *Biological Recovery, Heavy Metals, Spent Catalysts*

## INTRODUCTION

### *Bioleaching*

*Cycling Nature of Bioleaching*

Reactions 1, 2 and 3 show the cycling nature of bioleaching. Under optimum conditions, ferrous sulfate is biologically oxidized and becomes available for additional ores to undergo oxidization (Nemati *et al.*, 1998).

1. $H_2S+Fe_2(SO_4)_3 \rightarrow S\downarrow +2FeSO_4+H_2SO_4$

2. $MeS+Fe_2(SO_4)_3 \rightarrow MeSO_4+2FeSO_4+S^O$

3. $2S^O+ 3O_2+2H_2O \rightarrow T.F \rightarrow 4H^+ +2SO_4^{-2}$

*Activities of Microorganisms*

Acidophilic bacteria's levels of activity could be indirectly determined through iron and sulfur oxidation rate measurement. *Adaptation* is an option for stepping up the activities of acidophilic microorganisms. Most metallic ions are toxic to microorganisms and therefore *adaptation method* could be utilized to help them survive these harsh environments. Following *adaptation, Acidithiobacillus ferrooxidans* could grow and operate in the presence of various types of metallic ions. As a result, overall bioleaching synthesis could be accelerated through *adaptation* (Schinner, 1989).

## MATERIALS AND METHODS

***Microorganisms***

• The microorganism used in this study was *chomolitotrophic Acidithiobacillus ferrooxidans,* obtained via 10% inoculation of a pure strain of bacterium in batch culture.

***The Batch Culture of Bacterium***

Table 2.1 shows the specialized batch culture of *Acidithiobacillus ferrooxidans.* The batch pH was initially alkaline. Therefore, sulfuric acid was added to change Ph for bacterial growth.

**Table 2.1: The Batch Culture of Acidithiobacillus Ferrooxidans (Silverman *et al.*, 1959)**

| Matter | g/1000 ml |
|---|---|
| $(NH_4)_2SO_4$ | 3 |
| KCl | 0.1 |
| $MgSO_4.7H_2O$ | 0.5 |
| $KH_2PO_4$ | 0.5 |
| $FeSO_4.7H_2O$ | 33.3 |
| $Ca(No_3)_2$ | 0.01 |

***Bioleaching of Spent Catalysts Using Acidithiobacillus Ferrooxidans***

Bioleaching took place in a number of 500 ml Erlenmeyer flasks each containing 200 ml batch culture and a spent catalyst with a pulp density of 1%. A number of bacterial populations with a concentration of 1x107cell/ml were inoculated in each flask and then cultured at 30c° with a rotation speed of 160/m. Adaptation to V and Ni took place gradually in several phases, from a concentration of 100 mg/l to 2000 mg/l. finally, the bacterial-mediated bioleached content was determined.

## RESULTS AND DISCUSSION

***Results***

*Adaptation of the Spent Catalyst to Acidithiobacillus ferrooxidans*

Catalysts inoculated in batch culture were 60, 120 and 230 µm in size. Using *spectrophotometer*, $Fe^{tot}$ and $Fe^{3+}$absorption rate and also ferrous-to-ferric conversion rate were calculated at given time intervals. The reaction cut-off point was set at reaching 80% iron conversion rate. Past this threshold, next culturing would start. To determine Ni and V absorption rate by batch culture in every adaptation stage, samples were tested by ICP. In this study, the bacterial maximum toleration rate to the spent catalyst in a given time interval was 200 mg/l.

Also, the overall time for adaptation to spent catalyst sized 120 µm was 123 days. The last batch culture i.e. 2000 mg/l concentration had the longest duration (30 days). Considering the fact that the highest iron conversion rate (92%) occurred in the presence of the 120µmspent catalyst, the bioleaching process could be termed successful compared to other similar studies. *120µm* could be as well considered the optimum size for bioleaching. Figure 3.1 shows the ionic concentrations of $Fe^{3+}$ and $Fe^{2+}$ in the presence of *Acidithiobacillus ferrooxidans* and also the 120µm spent catalyst in various concentrations. Results show that ferrous ion sees a downward trend and ferric ion does the opposite. In other words, with the gradual build-up of the ferric iron, ferrous iron concentration is depleted, indicating the progression of reaction and occurrence of bioleaching. In initial stages of bioleaching, almost all ferrous ions are converted into ferric ions (Roher *et al.*, 1983).

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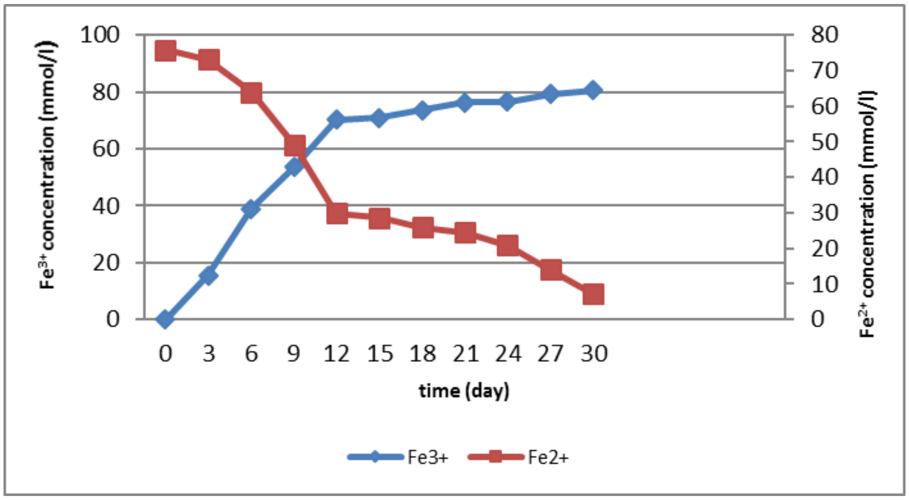


**Figure 3.1: Ferrous and Ferric ionic concentrations in different time periods in the presence of *Acidithiobacillus ferrooxidans* and 120 µm spent catalyst (in 30 days)**

Figure 3.2 shows the impact of various concentrations of 120 µm spent catalyst on ferrous oxidation potential in adapted batch culture. The highest and lowest iron conversion rates were observed at 2000 mg/l and 600 mg/l concentrations, respectively.

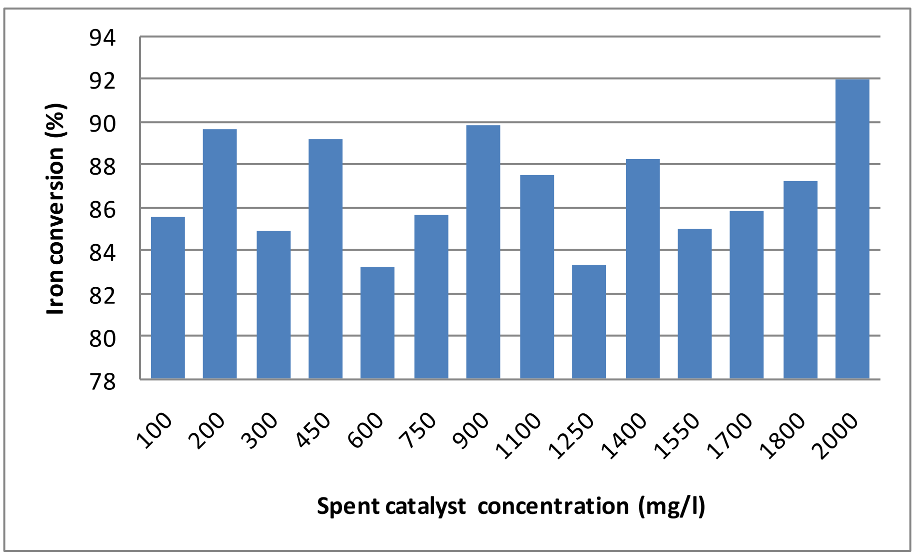


**Figure 3.2: The impact of various concentrations of120 µm spent catalyst on ferrous oxidation potential in adapted batch culture**

**Table 3.1: The Optimum Conditions of Bioleaching Using *acidithiobacillus ferrooxidans* and the Results**

| **Case** *A. ferrooxidans* | **pH** | **Temperature (°C)** | **Particle Size (µm)** | **Pulp Density (g/l)** | **Rotation Speed (rpm)** | **Recovery (%)** |
|---|---|---|---|---|---|---|
| **Ni recovery (%)** | 1.9 | 30 | 120 | 1.5 | 160 | 95.3 |
| **V recovery (%)** | 1.9 | 30 | 120 | 1.5 | 160 | 92 |

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The results indicate that the metal recovery outputs through bacterial-mediated (*Acidithiobacillus ferrooxidans*) bioleaching for Ni and V were 95.3% and 92% respectively. Table 3.1 shows the optimum conditions for maximum recovery of Ni and V using *Acidithiobacillus ferrooxidans.*
As shown by Figure 3.3, maximum Ni extraction in the presence of *Acidithiobacillus ferrooxidans* obtained 10 days after inoculation. However, V optimum extraction required 30 days. Ni and V toxicity to *Acidithiobacillus ferrooxidans* was Ni>V.

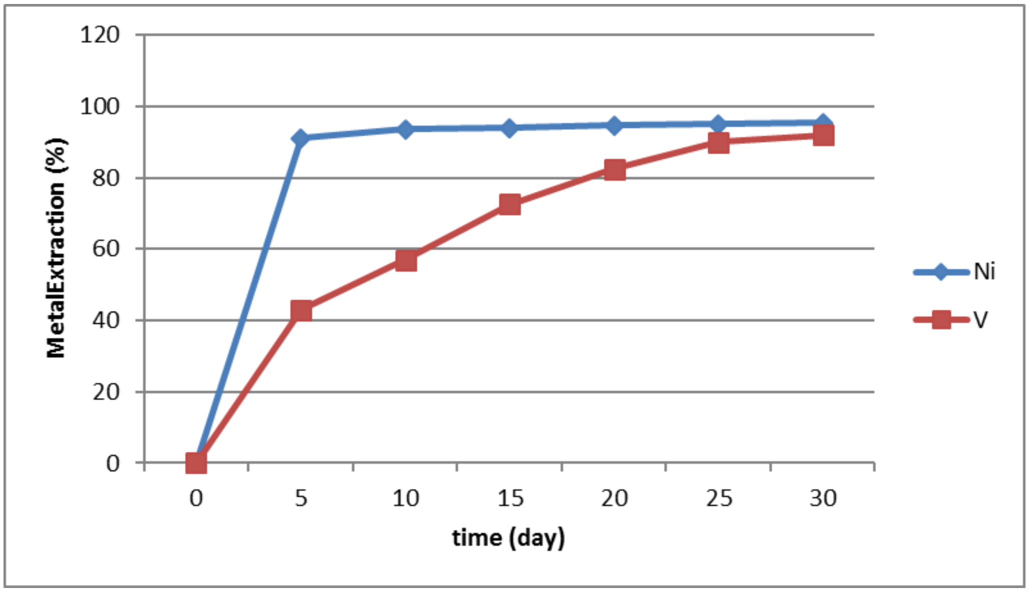


**Figure 3.3: The output of extracted Ni and V from 2000mg/l concentrated spent catalyst, using acidithiobacillus ferrooxidans at given time intervals (under optimum conditions)**

*Time-based Changes in Bacterial Population of Batch Cultures*
The numbers of bacteria in solution were counted macroscopically during adaptation phase. Bacterial population rose following ferrous-to-ferric oxidation. Bacterial population rose to $5.4 \times 10^7$ from the initial $1 \times 10^7$ and then fell to $3.6 \times 10^7$ following the death phase (Figure 3.4).

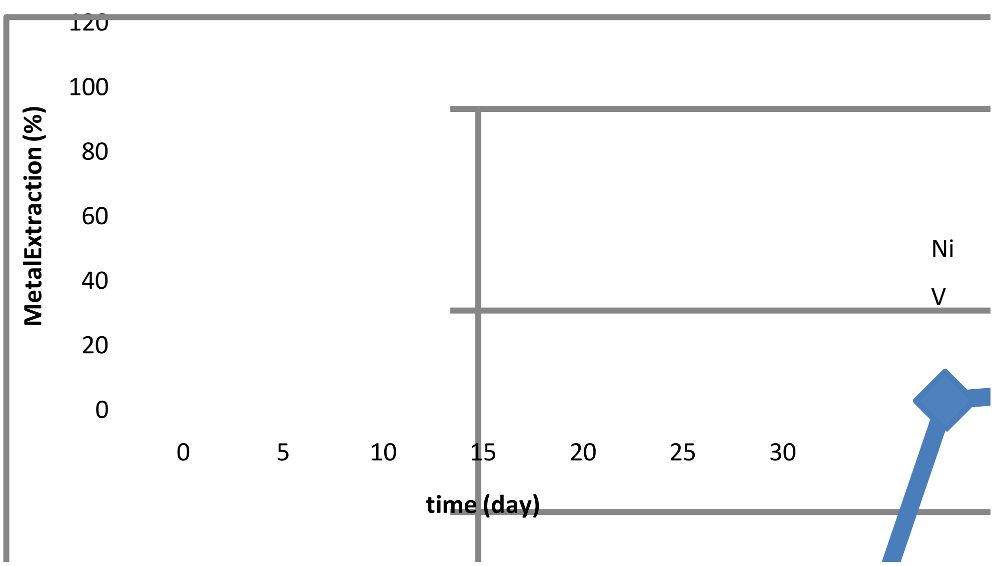


**Figure 3.4: Ferrous concentration and bacterial population changes during bioleaching using *acidithiobacillus ferrooxidans*, 120µm catalyst, 200 mg/l concentration (under optimum conditions)**

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*Bacterial Generation Time and Kinetics of Growth (K)*

Equations (1) and (2) have been used in calculation of generation time of the target bacterium (*Acidithiobacillus ferrooxidans*) and its kinetics of growth during bioleaching. Experiments revealed that high concentrations of the spent catalyst can act as growth inhibitor and slow the bacterial growth. For example, at 2000 mg/L, bacterium's cell division occurred once in 8 days and the kinetics of growth was 0.093(1/day).

**Equation 1**: $N_t=N_0e^{+kt}$

**Equation 2**: $\text{Log } N_t=\text{Log } N_0+t/G \text{ Log}2$

**N**= number of bacteria in Time *t*

**$N_{0=}$** number of bacteria in time *0*

**t**= time

**K**= kinetics of growth

**G**= bacterial doubling time

*Reaction Order*

To choose an appropriate synthetic model, bioleaching synthetic has been fitted to following models: *$1^{st}$ order* and *$2^{nd}$ order*. A linear relation is established after diagramming remaining concentrations versus time (t). The slope of this line is read as the kinetics of growth (k).While *lnC* is plotted versus time in $1^{st}$ order reaction rate, 1/C is used in $2^{nd}$ order one. Furthermore, reactant coefficient ($R^2$) proves that bioleaching synthesis follows either $1^{st}$ or $2^{nd}$ order reaction rate (Mishera *et al.*, 2009). Table 3.2 shows that the reaction rate of the batch culture containing 2mg/l Ni (in the presence of *Acidithiobacillus ferrooxidans*) follows $2^{nd}$ order reaction. Table 3.3, however, shows that the reaction rate of the batch culture that contains 6mg/l V follows $1^{st}$ order reaction.

**Table 3.2: $2^{nd}$ order reaction rate of the batch culture containing 2mg/l Ni (in the presence of *Acidithiobacillus ferrooxidans*)**

| **Reaction Rate** | **$R^2$** | **Equation of Line (Y)** |
|---|---|---|
| Zero order | 0.410 | Y=-2.107x+51.11 |
| $1^{st}$ order | 0.558 | Y=-0.075x+3.310 |
| $2^{nd}$ order | 0.821 | Y=0.005x+0.60 |

**Table 3.3: 1st order reaction rate of the batch culture containing 6mg/l V (in the presence of *Acidithiobacillus ferrooxidans*)**

| **Reaction Rate** | **$R^2$** | **Equation of Line (Y)** |
|---|---|---|
| Zero order | 0.866 | Y=-2.823x+79.93 |
| $1^{st}$ order | 0.993. | Y=-0.086x+4.557 |
| $2^{nd}$ order | 0.904 | Y=0.003x-0.005 |

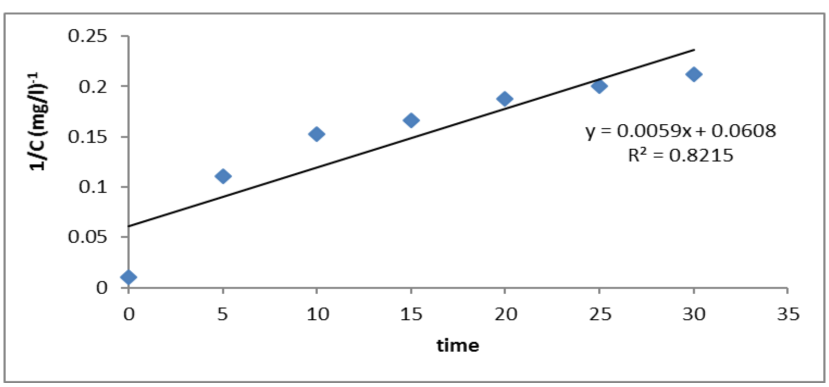


**Figure 3.5: $2^{nd}$ order reaction rate diagram of the batch culture containing 2mg/l Ni (in the presence of *Acidithiobacillus ferrooxidans*)**

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In Figure 3-5 the slope of the line plotted to represent the remaining reactant concentration versus time shows the 2nd order reaction kinetics. Therefore, reaction kinetics (k) is the slope of the line (0.005). Also, reaction kinetics (k) in first direction is the slope of the line (-0.086) in diagram 3.6.

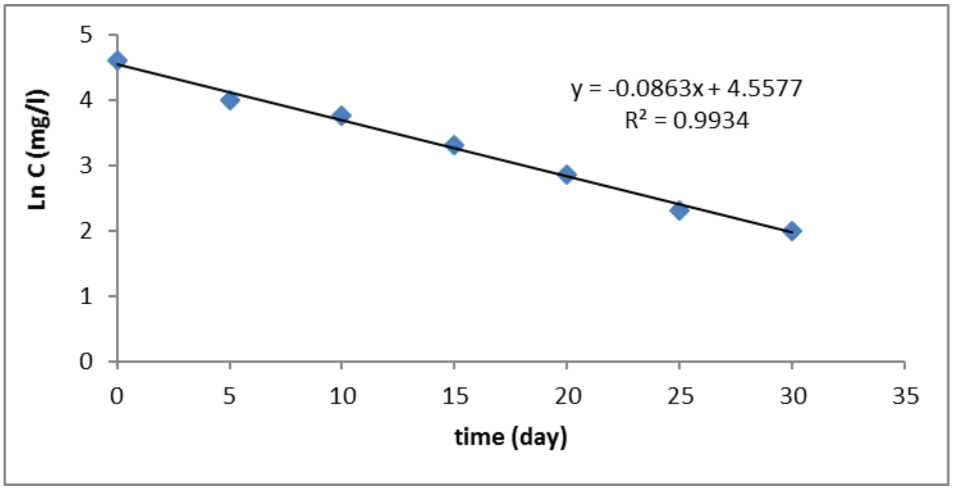


**Figure 3.6: 1st order reaction rate of the batch culture containing 6mg/l V (in the presence of *Acidithiobacillus ferrooxidans*)**

Table 3.4 shows reaction kinetics (k), $R^2$, line equation (Y), slope of the line and reaction order during bioleaching in two batch cultures containing 2mg/l Ni and 6mg/l V in the presence of *Acidithiobacillus ferrooxidans*.

**Table 3.4: Reaction kinetics (k), $R^2$, line equation (Y), slope of the line and reaction order during bioleaching in two batch cultures containing 2mg/l Ni and 6mg/l V ( in the presence of *Acidithiobacillus ferrooxidans*)**

| **Spent catalyst 2000( mg/l )** | **(K) (1/day)** | **$R^2$** | **(Y)Line Equation** | **Slope of the line** | **Reaction order** |
|---|---|---|---|---|---|
| Catalyst containing 2mg/l Ni | 0.005 | 0.821 | Y=0.005X+0.060 | 0.005 | 1st order |
| Catalyst containing 6 mg/l V | -0.086 | 0.993 | Y= -0.086X+4.557 | -0.086 | 1st order |

***Conclusion***

- The bioleached outputs of target heavy metals (Ni and V)were 95% and 92% respectively, could be deemed significant considering other similar studies
- Experiments revealed that high concentrations ( over 2000mg/l) of the spent catalyst may act as growth inhibitor and therefore slow the bacterial growth
- As Ph drops, bacterial populations begin to rise, indicating the bacterial activity and acid production. Also, ferrous-to-ferric oxidation prompted an increase in bacterial population
- Bacterial population rose to $5.4\times 10^{7}$ from the initial $1\times10^{7}$ and then fell to $3.6\times10^{7}$ following the death phase.
- The reaction rate of the batch culture containing 2mg/l Ni ( in the presence of *Acidithiobacillus ferrooxidans*) follows 2nd order reaction and the reaction kinetics (k) is the slope of the line( 0.005)
- The reaction rate of the batch culture containing 6mg/l V follows 1st order reaction and the reaction kinetics (k) is the slope of the line( -0.086)

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