Effect of solids concentration on removal of heavy metals from mine tailings via bioleaching

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Abstract

Mining of mineral ore and disposal of resulting waste tailings pose a significant risk to the surrounding environment. The objective of this work is to demonstrate the feasibility to remove heavy metals from mine tailings with the use of bioleaching and meanwhile to investigate the effect of solids concentration on removal of heavy metals from mine tailings by indigenous sulfur-oxidizing bacteria and the transformation of heavy metal forms after the bioleaching process. This work showed the laboratory results of bioleaching experiments on Pb–Zn–Cu mine tailings. The results showed that 98.08% Zn, 96.44% Cu, and 43.52% Pb could be removed from mine tailings by the bioleaching experiment after 13 days at 1% (w/v) solids concentration and the rates of pH reduction, ORP rise and sulfate production were reduced with the increase of solids concentration, due to the buffering capacity of mine tailing solids. The results also indicated that solid concentration 1% was found to be best to bacterial activity and metal solubilization of the five solids concentration tested (1%, 2%, 5%, 8% and 10%) under the chosen experimental conditions. In addition, the bioleaching had a significant impact on changes in partitioning of heavy metals.
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Keywords: Bioleaching; Mine tailings; Heavy metal; Solids concentration; Partitioning

1. Introduction

In Hunan Province, China, there are about 474 metal mines. The mining and metallurgical activities that were particularly intensive during the last century resulted in the generation of huge amounts of mine tailings, including acid-generating sulfidic tailings. Most of the tailings have been left without any management in these mines. Their improper management in the past resulted in the migration of heavy metals to the surrounding environment, contributing to soil substrates contaminated, soil texture destroyed, short of nutrient, ecological landscape destroyed, groundwater pollution and biological diversity decrease etc. The presence of toxic heavy metals in mine tailings caused lots of serious environmental problems. In order to resolve the above problems, it is important to develop a suitable and economical technology for removal of heavy metals from mine tailings.

Bioremediation of heavy metals has gained increased attention since it is innovative, environmentally friendly and eco-nomical [1]. The bioleaching process may be defined as the solubilization of metals from solid substrates either directly by the metabolism of leaching bacteria or indirectly by the products of metabolism [2]. Bioleaching processes are based on the ability of microorganisms to transform solid compounds and result in soluble and extractable elements, which can be recovered. Metal solubilization from solid wastes or other solids is achieved through a variety of acidophilic and chemoautolithotrophic bacteria such as *Thiobacillus thiooxidans* and *Thiobacillus ferrooxidans*. Other bacteria such as *Leptospirillum ferrooxidans* was reported to bioleach zinc from the marmatite flotation concentrate effectively [3].

Because of the advantages of low cost and environment friendliness and better efficiency, bioleaching technology has been a great success for the mining industry [4]. In recent years, bioleaching has also proved to be a possible way to remove heavy metals from metal contaminated materials such as anaerobically digested sewage sludge, contaminated river sediment, spent nickel–cadmium batteries, and incinerator fly ash [5–10]. In general, mine tailings are similar to these materials with respect to the physical and chemical characteristics. It shows that technologies potentially applicable for

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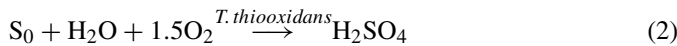*E-mail address:* axore@163.com (Y.-G. Liu).

treatment of these materials can be adapted to treat the mine tailings.

In this work we want to carry out bioleaching experiment of mine tailings by indigenous sulfur-oxidizing bacteria. The bacterium *T. thiooxidans* is active at low pH and can endure harsh conditions that exist in concentrated solutions of heavy metals. It can catalyze the oxidation of elemental sulfur or reduced sulfur compounds to sulfuric acid and obtain energy from the oxidation of elemental sulfur or reduced sulfur compound, and cause bioacidification and solubilization of heavy metals. The oxidation and acid producing activity of sulfur-oxidizing bacteria are the primary mode of solubilization of heavy metals in the bioleaching process. The solubilization mechanism of bioleaching by *T. thiooxidans* can be direct and indirect. In the direct mechanism, metal sulfides can be oxidized into sulfates by these acidophilic bacteria. In the indirect mechanism, H_2SO_4 generated from the oxidation of elemental sulfur or reduced sulfur compounds by *T. thiooxidans*. The direct and indirect mechanism can be described by the following equations [11]:

The direct mechanism:

$$MS + 2O_2 \overset{T.\ thiooxidans}{\longrightarrow} MSO_4 \tag{1}$$

The indirect mechanism:

$$S_0 + H_2O + 1.5O_2 \overset{T.\ thiooxidans}{\longrightarrow} H_2SO_4 \tag{2}$$

$$H_2SO_4 + \text{materials-M} \rightarrow \text{materials-2H} + MSO_4 \tag{3}$$

where M is a bivalent metal.

The effectiveness of bioleaching is highly dependent on the physical, chemical and biological factors in the system [11]. Among these affecting factors such as nature of contaminated material, solids concentration, temperature, oxygen, pH, oxidation–reduction potential (ORP), composition of the medium, bacterial strain and cell concentration [12], solids concentration plays an important role in optimization of the bioleaching process. A higher solid concentration results in a higher solid to liquid ratio. Although this increases the concentration of heavy metals in the leaching environment, the increase in concentration of the leached metals and the resultant toxicity may inhibit the growth of the microorganisms and the production of acids, and moreover, pH variation in the bioleaching process are influenced by solids concentration in the system because of buffering capacity [13]. However, solubilization of heavy metals in bioleaching is highly pH-dependent [14], pH is the single most important factor that influences metal solubilization during the metal bioleaching. Therefore, a complete understanding of the factor solids concentration that affects the bioleaching process is very important to optimize the bioleaching process.

The objective of this work is to demonstrate the feasibility to remove metal ions from mine tailings with the use of bioleaching and this study also investigates the effect of solids concentration on remobilization of heavy metals from mine tailings by indigenous sulfur-oxidizing bacteria and the transformation of heavy metal forms after the bioleaching process.

2. Materials and methods

2.1. Sample and source and its characterization

Mine tailings and bacteria-containing sludge used in this study were collected from the Shuikoushan tailing dam of located 40 km south of Hengyang City, central China's Hunan province. The Shuikoushan mine mainly produced lead, zinc and copper and the large-scale Kangjiawan mine of the Shuikoushan mine was the fourth largest lead and zinc mine in China with an annual ore-producing capacity of more than 400,000 t and reserves 16,000,000 t of lead and zinc. The mine tailings samples and bacteria-containing sludge were transported back to lab and stored at 4 °C prior to their use. Mine tailings samples were dried in a glove box, which was continuously purged with oxygen-free nitrogen. Then mine tailings were characterized before its use in the bioleaching experiment. The measured characteristics of mine tailings were listed in Table 1.

In addition to the total metal content, the distribution of metals among various fractions in the mine tailings is a useful measure as partitioning information determines the behavior of the metal in the environment and it allows an investigation of which metals, bound to which fractions, are solubilized during bioleaching. So it was important to describe the partitioning of heavy metals into different fractions before and after their biological removal from mine tailings. Four binding fractions (F_1: exchangeable, F_2: reducible fraction or fraction associated with Fe and Mn oxides, F_3: oxidizable-fraction associated with sulfides and organic matter, F_4: residual fraction.) of heavy metals (Cu, Zn, and Pb) were analyzed using the Four-step procedure recommended by Community Bureau of Reference (BCR) [15].

In this study, the amounts of metals were determined by atomic adsorption spectro-photometer (AAS). Binding fractions of heavy metals in mine tailings were listed in Table 2.

Table 1
Measured characteristics of mine tailings

Parameter	Values
pH	6.30
Water content (%)	1.32
Organic matter (%)	1.37
Cu (mg/kg dry weight)	1332.4
Zn (mg/kg dry weight)	1169.1
Pb (mg/kg dry weight)	153.8
Fe (mg/kg dry weight)	4756.8
Ca (mg/kg dry weight)	38749
Carbonates (as $CaCO_3$ mg/kg)	97665
S_{total}	132470
Sulfides (mg/kg dry weight)	184892
Sulfates (mg/kg dry weight)	3208

Table 2
Binding fractions of heavy metals in mine tailings

Heavy metals	F_1 (%)	F_2 (%)	F_3 (%)	F_4 (%)
Cu	19.94	11.77	46.13	22.16
Zn	15.52	9.90	38.77	35.81
Pb	15.02	18.60	10.40	55.98

Table 3
Experimental conditions for bioleaching

Run no	Solids concentration (w/v, %)	Temperature (°C)	rpm	Inoculum dosage (v/v, %)
A_1 (blank)	1	28	180	No
A	1	28	180	2
B	2	28	180	2
C	5	28	180	2
D	8	28	180	2
E	10	28	180	2

2.2. Enrichment and acclimation of indigenous sulfur-oxidizing bacteria

Indigenous sulfur-oxidizing bacteria were enriched by adding bacteria-containing sludge from the Shuikoushan mine to 250 mL Erlenmeyer flask with 100 mL culture medium (2.15 g/L sulfur [prepared from $Na_2S_2O_3 \cdot 5H_2O$], 1 g/L NH_4Cl, 1 g/L KH_2PO_4, and 0.5 g/L $MgCl_2 \cdot H_2O$), at 2% (v/v). The medium provided nutrition, a sulfur substrate, and buffer capacity, for the growth of sulfur oxidizing bacteria in the sludge. This Erlenmeyer flask was placed in gyratory shaker and mixed at 180 rpm and 28 °C. The pH of the sludge was monitored. When the pH value of sludge dropped under 2.0, 2 mL acidified sludge was then transferred to 100 mL fresh culture medium under the same conditions. The inoculum for the bioleaching experiment was obtained by repeating the above procedures until the indigenous sulfur-oxidizing bacteria in the sludge had the highest rate of acidification.

2.3. Bioleaching experiment

The bioleaching was conducted in 250 mL Erlenmeyer flasks with 100 mL same culture medium with cotton-plug at five controlled solids concentration (w/v) (1%, 2%, 5%, 8%, and 10%). The flasks were agitated at 28 °C, 180 rpm on a gyratory shaker. A blank Run (A_1) without either inoculum of bacteria was also carried out in order to compare the results. Bioleaching conditions were tabulated in Table 3. During the bioleaching process the water loss due to evaporation was replenished with distilled water daily.

2.4. Chemical analysis

During the experiment, the pH and ORP value were measured at half-day intervals, whereas sulfate and soluble heavy metals (Cu, Zn, Pb) were measured at 1-day interval. Binding fractions of heavy metals were measured after the bioleaching using the Four-step procedure recommended by BCR. The pH and ORP values were measured by pH and ORP analyzer (Model K100, Dr-Kornder). Total sulfur of the tested tailings sample were determined by dry combustion [16]. Sulfides of the tailings (acid volatile sulfide, AVS) were determined by acidifying tailings sample under N_2 environment; and collecting the evolved H_2S in a trap containing excess zinc acetate and NaOH [17]. The approximate gravimetric method [18] was used for carbonates of the tailings and the Walkley–Black wet combustion method

[19] for organic matter of the tailings. The mine tailings slurry sample taken from the reactor was centrifuged at a speed of 10,000 rpm for 20 min. The filtrate was analyzed for its sulfate concentrations according to the standard methods [20]. Heavy metal (Cu, Zn, Pb, Ca, Fe) concentrations by atomic absorption spectrophotometer (Model ZEEnit700, Analytik Jena AG). The total heavy metals in the mine tailings were determined after HF–HNO₃–HCl digestion method [21]. All treatments and controls were done in duplicate.

3. Results and discussion

3.1. Effect of different solids concentration on pH and ORP

During the bioleaching process, elemental or reduced sulfur compounds was oxidized to sulfuric acid by the indigenous sulfur-oxidizing bacteria, resulting in a decrease in pH and solubilization of metals. The variations of pH during the bioleaching process with different solids concentration were shown in Fig. 1.

From the figure, there was an initial rise in pH between day 0 and day 2, perhaps caused by the buffer or by the release of alkaline from mine tailings during the initial stage of bioleaching. In the control Run A_1 without inoculation of indigenous sulfur-oxidizing bacteria, the pH increased from 6.57 to 6.74 in the first 2 days, then the final pH was stabilized at about 6.58. In the other runs, the initial pH were under 7.0, because the 2% of inoculum with the pH value under 2.0 was added, and a faster decrease in pH was observed after 2 days and the final pH was much lower than that of Run A_1 due to the indigenous sulfur-oxidizing bacteria oxidation of sulfur.

As indigenous sulfur-oxidizing bacteria have grown on sulfur, sulfuric acid accumulated in Erlenmeyer flasks and the mine tailings pH decreased to the pH value of about 2.0 in 3, 3.5,
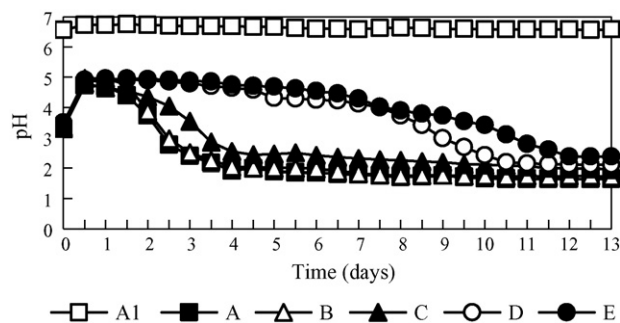

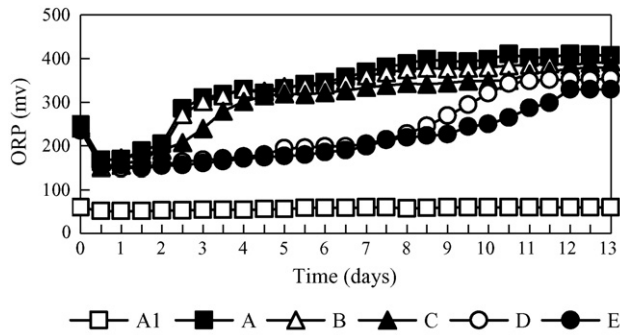Fig. 1. Effect of solids concentration on pH during bioleaching.

Fig. 2. Effect of solids concentration on ORP during bioleaching.

Fig. 3. Effect of solids concentration on sulfate production during bioleaching.

4.5, 10.5, and 13 days for solids concentrations of 1%, 2%, 5%, 8%, and 10%, respectively. It was apparent that the mine tailings solids concentration significantly affected the rate of pH decrease. The rate of pH reduction decreased with an increase in solids concentration, because mine tailings with higher solids concentration had a higher buffering capacity. Thus, it required more time to reach lower pH for mine tailings with higher solids concentration.

Changes in oxidation–reduction potential (ORP) also indicate the level of activity of sulfur-oxidizing bacteria. Fig. 2 showed the changes in ORP with bioleaching time at different solids concentration. From Fig. 2, the ORP of the control Run A_1 was stabilized at about 50 during the bioleaching period. In contrast, the ORP of the other runs increased by the oxidation of sulfur rapidly and then gradually approached a limiting value within 13 days. Compared to the variation of pH in bioleaching, the variations of ORP were much related to those of pH. The increase of ORP had a similar trend with decrease of pH. It can be also observed that the higher solids concentration of mine tailings induced a drop in the rate of ORP increase because of a higher buffering capacity.

In a word, at the higher solids concentration, system has higher buffering capacity and therefore more time was needed for sulfur-oxidizing bacteria to lower system pH, increase system ORP. So pH decreased faster at lower solids concentration than at higher ones and ORP variation significantly correlate with solids concentration.

3.2. Variation of sulfates in bioleaching process

The rate of sulfate production is a important indicator of bioleaching efficiency. Fig. 3 showed the effect of solids concentration on production of sulfate in the bioleaching process. It was found that the metabolic final product, sulfate was produced with time and the rate of sulfate production also decreased with the increase in solids concentration.

From Figs. 1–3 the results indicated that the rates of pH reduction, ORP rise and sulfate production were reduced with the increase of solids concentration, due to the buffering capacity of mine tailing solids. So we can draw a conclusion that the high solids concentration inhibited the growth of the microorganisms and decelerated the bioleaching process under the initial time period.
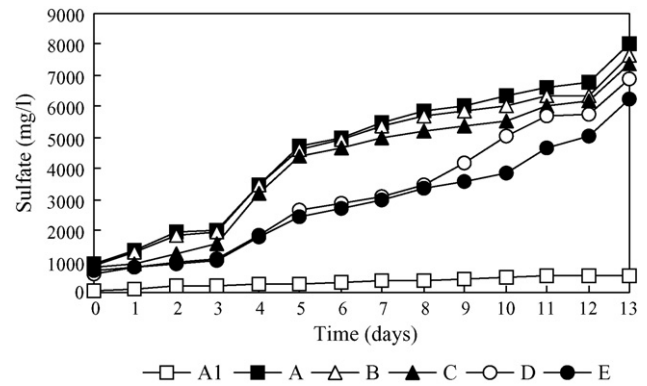
3.3. Metal solubilization in bioleaching

Fig. 4. showed the effects of solids concentration on the solubilization of heavy metals (Cu, Zn, Pb) from mine tailings during the bioleaching process. By comparison between tests A, B, C, D and test E, it is evident that heavy metals removal can be strongly influenced by solids concentration; lag phase was observed in the solubilization of Cu, Zn, Pb at high solids contents (Runs D and E) and the rate of metal solubilization decreased with increasing solids concentration. This is attributed to the fact that the final pH value was higher for the system with higher mine tailing solids concentration due to higher buffering capacity, however the percentage of metal solubilization was found to highly depend on the pH value during the bioleaching process.

Except for Pb, solubilization efficiency of Cu, Zn was satisfactory. The solubilization efficiency of bioleaching of Zn was highest at 98.08–75.93%, and that of Cu was higher at 96.44–65.26% and that of Pb was lowest at 43.52–14.48% of the three metals in the mine tailings. The solubilization of Zn, and Cu from mine tailings was significantly higher than that of Pb at the same solid concentration. The efficiency of Pb solubilization is not high in the bioleaching process because the solubilized Pb can form into low solubility of $PbSO_4$ ($Ksp = 1.62 \times 10^{-8}$) with sulfate.

In conclusion, solids concentration strongly affected the solubilization of heavy metals during the bioleaching process. From the experimental results, it was clear that the optimal solids concentration for solubilization of total extractable heavy metal was 1% under the chosen experimental conditions. At 1% (w/v) solids concentration the solubilization efficiency of bioleaching of Zn, Cu, Pb arrived at 98.08%, 96.44%, and 43.52%, respectively, within 13 days.

3.4. Variation in the binding forms of heavy metals

The different forms of heavy metals stand different energy states, and affect not only the efficiency of bioleaching but also the bio-availability of heavy metals after bioleaching. Metals in exchangeable, and Fe/Mn oxide-bound fractions are considered to be more mobile, dangerous and bioavailable. The organic matter/sulfide-bound and residual metals are considered to be more stable and nonbioavailable than metals in exchange-
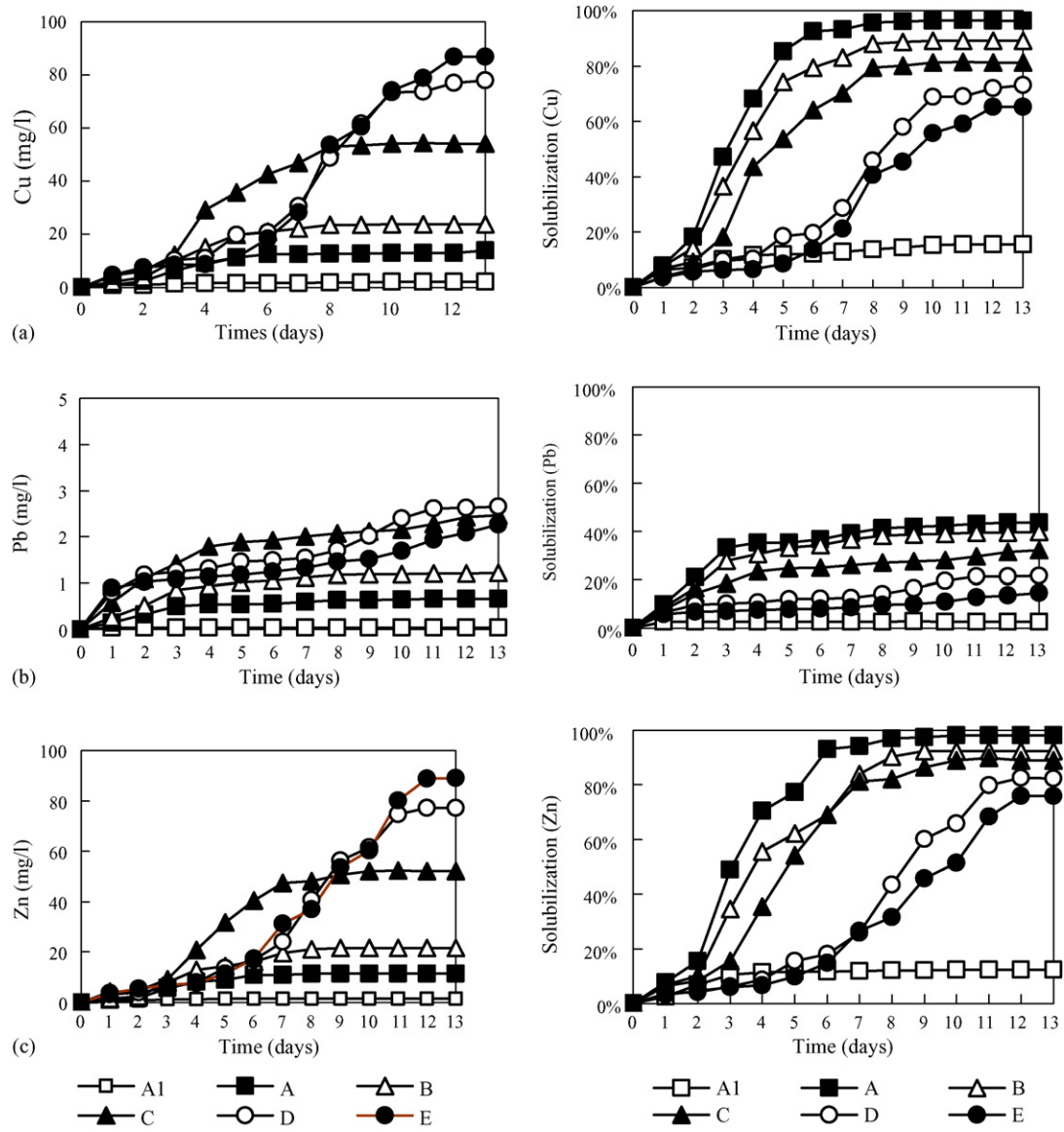
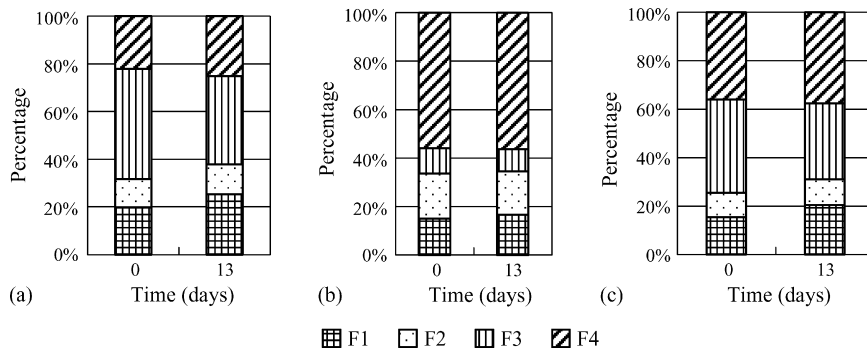Fig. 4. Bioleaching of Cu (a), Pb (b) and Zn (c) at different solids concentration.

Fig. 5. Variation in partitioning of chemical forms for Cu (a), Pb (b) and Zn (c) of control A_1 after the experiment.
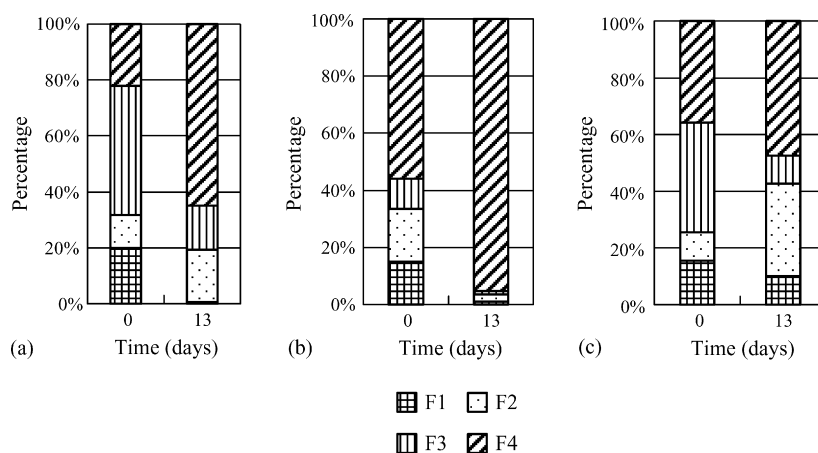
Fig. 6. Variation in partitioning of chemical forms for Cu (a), Pb (b) and Zn (c) of Run A after the bioleaching.

able, and Fe/Mn oxide-bound fractions. So it was necessary to describe the partitioning of heavy metals into different fractions before and after the bioleaching.

The partitioning of heavy metals in control A_1 and Run A was shown in Figs. 5 and 6. With the control A_1, for all of the metals of control A_1, F_2: the Fe/Mn oxide-bound fractions, and F_4: residual forms had little change during the running, while F_3: the sulfide-bound fractions of Cu, Pb and Zn showed a decrease because of the oxidizing environment caused by aeration with agitation. As for F_1 the exchangeable bound form, all of the metals showed an increasing trend during the experimental period.

The bioleaching had a significant impact on changes in partitioning of heavy metals. Fig. 6 showed variation in the binding forms of heavy metals in Run A before and after the bioleaching process. Cu, mainly bound to F_3: the sulfide-bound fractions and F_1 the exchangeable bound fractions in raw mine tailings, was mainly bound to F_4: residual fraction and F_2 the Fe/Mn oxide-bound fractions after the bioleaching process. Solubilization efficiency of Pb was somewhat unsatisfactory, but Pb was mainly bound to F_4: residual fraction after the bioleaching process. So Pb remaining in the mine tailings was stable and nonbioavailable after the bioleaching process. A transformation of binding fractions occurred during the bioleaching of Zn; mainly bound to F_3: the sulfide-bound fractions and F_1: the exchangeable bound fractions before the bioleaching process, it was mainly bound to F_4: residual fraction and F_2 the Fe/Mn oxide-bound fractions after the bioleaching process. After the bioleaching process, metals remaining in the mine tailings were mainly found in the stable fractions.

Table 4
The content of the remaining metals, S_{total} and sulfide in the tailings after leaching

	1%	2%	5%	8%	10%
Cu (mg/kg)	47.43	144.96	249.69	357.34	462.87
Zn (mg/kg)	22.45	87.21	127.43	204.71	281.4
Pb (mg/kg)	86.87	92.62	104.39	120.59	131.52
Sulfide (mg/kg)	30506	82517	121930	152687	161254
S_{total} (mg/kg)	67679	97341	109571	114472	124361

4. Conclusions

Experimental results showed solids concentration strongly affected the solubilization of heavy metals during the bioleaching process. Of the five solids concentration tested (1%, 2%, 5%, 8%, and 10%), results indicate that oxidizing activity of indigenous sulfur-oxidizing bacteria to mine tailings was greatest in 1% under the chosen experimental conditions. Except for Pb, solubilization efficiencies of total extractable Zn and Cu were very high (>96%) at this solid concentration, while that of Pb was 43.52%. After the bioleaching process, metals remaining in the mine tailings had relatively low contents. The contents of the remaining metals, S_{total} and sulfide in the tailings after leaching were shown in Table 4. This proved that it was feasible to remove heavy metals from mine tailings with the use of the bioleaching remediation method.

The results also showed that differences in heavy-metal binding before and after bioleaching were significant. After the bioleaching, metals remaining in the mine tailings were mainly found in the stable fractions process and were stable to the surrounding environment.

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