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Дигитални репозиторијум Рударско-геолошког факултета Универзитета у Београду

[ДР РГФ]

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Article

Bioleaching Process for Copper Extraction from Waste in Alkaline and Acid Medium

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Abstract: Flotation wastes are becoming a valuable secondary raw material and source of many metals and semimetals worldwide with the possibilities of industrial recycling. The flotation tailings contain oxide and sulfide minerals that have not been sufficiently stabilized and form acidic mine waters, which in turn contaminate groundwater, rivers, and reservoirs sediments. An effective way to recycle these mine wastes is to recover the metals through leaching. While the focus is on acid bioleaching by iron- and sulfur-oxidizing bacteria, alkaline leaching, and the removal of iron-containing surface coatings on sulfide minerals contribute significantly to the overall environmental efficiency of leaching. For this study, static and percolate bioleaching of copper from flotation waste at the Bor copper mine in Serbia was investigated in alkaline and then acidic environments. The aim of the study was to verify the effect of alkaline pH and nutrient stimulation on the bioleaching process and element extraction. A sample was taken from a mine waste site, which was characterized by XRF analyses. The concentration of leached copper was increased when copper oxide minerals dissolved during alkaline bioleaching. The highest copper yield during alkaline bioleaching was achieved after 9 days and reached 67%. The addition of nutrients in acidic medium enhanced the degradation of sulfide minerals and increased Cu recovery to 74%, while Fe and Ag recoveries were not significantly affected. Combined bioleaching with alkaline media and iron- and sulfur-oxidizing bacteria in acidic media should be a good reference for ecological Cu recovery from copper oxide and sulfide wastes.

Keywords: copper; alkaline; acidic bioleaching; secondary raw materials



Citation: Ristović, I.; Štyriaková, D.; Štyriaková, I.; Šuba, J.; Širadović, E. Bioleaching Process for Copper Extraction from Waste in Alkaline and Acid Medium. *Minerals* **2022**, *12*, 100. <https://doi.org/10.3390/min12010100>

Academic Editors: Carlos Hoffmann Sampaio, Wesley Monteiro Ambros and Bogdan Grigore Cazacliu

Received: 27 December 2021

Accepted: 12 January 2022

Published: 15 January 2022

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1. Introduction

The copper and precious metal deposits in the territory of the town of Bor in the Republic of Serbia were discovered at the beginning of the 20th century. Since then, there have been two world wars, changes in ore processing technologies, and the Bor mine has been closed and reopened several times. Due to its importance, the entire radius around the town of Bor was the focus of investors for the exploitation of copper and gold. Since its establishment, the local company RTB Bor (Bor Mining and Metallurgical Basin) was state-owned. Constant mismanagement led to the sale of part of the company (63%) to the Chinese partner Serbia Zijin Copper DOO Bor. The main activity of the company is mining and processing of copper ore, and the production of cathode copper, copper wire, gold and silver, sulfuric acid, copper sulfate, selenium, platinum, palladium, brass, bronze, and precious metal salts.

In addition to primary reserves, Bor mines have large quantities of secondary raw materials derived from centuries of mining and processing copper ores.

The total amount of flotation waste in Bor is 588 million tonnes (Table 1). The old flotation waste (Figure 1) contains the most valuable source of copper for the industry. The

old Bor flotation dump was active from 1934 to 1980 and includes two fields (fields 1 and 2). The area of these two fields is about 1.6 km². The total volume of old flotation waste from Zijin Bor Copper is 28,000,000 tonnes with an average grade of 0.245% copper, 0.395 g gold per tonne, and 1.7 g silver per tonne. The recovery of a significant amount of copper, together with other metals, can reduce the environmental impact of the waste [1].

Table 1. Secondary raw materials in flotation tailings Zijin Bor Copper (old RTB Bor and Majdanpek).

Location	Tailings (t)	Cu (%)	Au(g/t)	Ag (g/t)
Old Flotation Tailing Dump	28,000,000	0.245	0.395	1.700
Tailing Dump RTH	50,000,000	0.200	0.100	0.500
Veliki Krivelj	160,000,000	0.100	0.050	0.500
Majdanpek	350,000,000	0.079	0.154	1.071
Σ All flotation tailings	588,000,000	0.103	0.133	0.897



Figure 1. Location of exploratory field.

Table 1 shows the approximate amounts of secondary raw materials in flotation wastes of Zijin Bor Copper mine (old RTB Bor) [1].

The analysis of a sample of cathode copper from the ore mined in 2012 at the Veliki Krivelj open pit mine, which belongs to the RTB Bor Group, provided an overview of the amounts of copper lost in the technological phases of mining, flotation concentration, and metallurgical concentration.

The results of the analysis show that of the total amount of copper in the deposit (100%), a larger part remains immediately unused, namely with the overburden (55%), and a smaller part is utilized in the final product—copper in the form of cathodes (45%). The losses described are the result of the state of the art and the processes used for copper production [2].

A previous detailed analysis of the mineralogical and chemical composition of the old flotation waste samples showed that the copper content in the tailings can reach 0.5%. The qualitative and quantitative mineralogical analysis of the waste samples showed that the sulfide mineral content in the sample is 21% and the pyrite mineral content is 20.81%. The remaining minerals (78%) in the sample consist of silicates, quartz, and carbonates, while oxides account for about 1%. In terms of composition, covellite (0.081%), chalcopyrite (0.049%), enargite (0.037%) and chalcocite (0.018%) were discovered as copper sulfide minerals [3].

The flotation residues consist mainly of a fine-grained fraction, as 55.3% of the particles are smaller than 0.074 mm [4]. In the presence of water and ambient oxygen, the typical release of pyrite is about 91%, which represents a significant potential for the mobilisation of toxic metal ions and the formation of acid mine drainage over many years [5].

Obviously, there are still obstacles to the acceptance and implementation of bioleaching in the processing of chalcopyrite, which are not necessarily limited to biological aspects.

According to Holmes and Debus (1991), biological refining must have more than 20 advantages over conventional processing to attract the interest of the mining industry [6]. The main disadvantage of sulfide (bio)leaching is that the process is considered slow compared to the pyrometallurgical process and other high-intensity hydrometallurgical processes [6].

Although several new alkaline leaching techniques, including two-stage mineral leaching and metal recovery from tailings ore [7], have been proposed, and recent extraction experiments have been conducted, there are very few studies on copper extraction from solid wastes such as copper sulfide residues associated with an alkaline leaching environment colonized by bacteria. These studies are mainly concerned with the isolation of strains [8], the biological leaching process of flotation wastes, and the alkaline leaching mechanism of chalcopyrite [9].

The alkaline leaching mechanisms were briefly discussed by authors Yin et al. (2018) [10]. The comparison of leaching with alkaline bacteria, ammonia, or their combination undoubtedly shows that copper recovery is significantly higher when combined with an increase in Cu extraction yield of 4.42% (48 days, ammonia leaching), or about 20% (30 days, alkaline bacteria leaching) at 29.57% (30 days, combined leaching). The alkaline microorganisms and the ammonia solution had a synergistic effect, especially at low ammonia levels (30 g/L to 60 g/L). Furthermore, high doses of ammonia (>60 g/L) reduced bacterial activity to a minimum or even destroyed the microorganisms.

Sulfide minerals are often biodegraded in acidic environments, carrying unwanted amounts of iron, alumina, and silica, which then overlay the surface of the sulfide. If ecological alkaline biochemical pre-treatment is carried out, the environmental hazard of acid mineral dissolution can be reduced, and this is the aim of this study. Although acid bioleaching by iron- and sulfur-oxidizing bacteria is the main focus, alkaline leaching and the removal of iron surface coatings on sulfide minerals contribute significantly to the overall leaching performance.

2. Materials and Methods

2.1. Materials

Within the RIS-Cure project “Zero waste recovery of copper wastes in the ESEE region (East & South-East Europe)”—EIT RawMaterials, in August 2020, the geological surveys were done—exploratory drilling on old Bor tailing dump. Drilling was performed with drilling rig Mustang 4 F1, PQ3 diameter (continual coring) with wire line method. In total it has been drilled 103.50 m:

- Well B—1 21.50 m;
- Well B—2 18.50 m;
- Well B—3 30.00 m;
- Well B—4 33.50 m.

Figure 2 shows the position of wells. Position of wells are marked with black points.



Figure 2. The position of wells on old Bor tailing dump (edged with red line—total surface area of dump).

After drilling, samples were transported to the laboratory of Technical faculty of Bor for preparation. The drilling cores were packed and stored in the boxes provided and the boxes were labelled. Samples were taken from each meter of drilling, and approximately 100 samples were packed in 100 bags, each with a weight of approximately 2.5 kg, resulting in a total of approximately 250 kg of material.

Of all the samples, a total of 30 kg was prepared and sent to Slovakia for leaching tests. At each meter of sample, the material was divided into halves longitudinally, meaning that in total, 1 m of sample was divided into halves along the longitudinal axis into two sub-samples: left and right. One part of the sample was kept as reserve until the end of the project, and the other part was to be sent to the laboratory of ekolive.

Sample preparation for the laboratory of ekolive in Slovakia was carried out using a quartering method (manually). This method is applied to samples that have large amounts and mass but are sufficiently dry, so that mixing and quartering can be done. The process of applying this method involves distribution of samples on a flat surface (inert foil), mixing of samples, and formation of a cone with a spoon. The cone is then stretched into a cake and separated into four parts. Two parts are intended for continuation of quartering, and the other two are packed into reserve packaging. Quartering was done several times until the required amount of 300 g by sample was accomplished. From every sample (from 100 m drilled, at every meter one sample was taken), 100 sample packages weighting 300 g were prepared, one from every meter of the exploratory well.

The packing and disposal of the drill core and samples are shown in Figure 3.

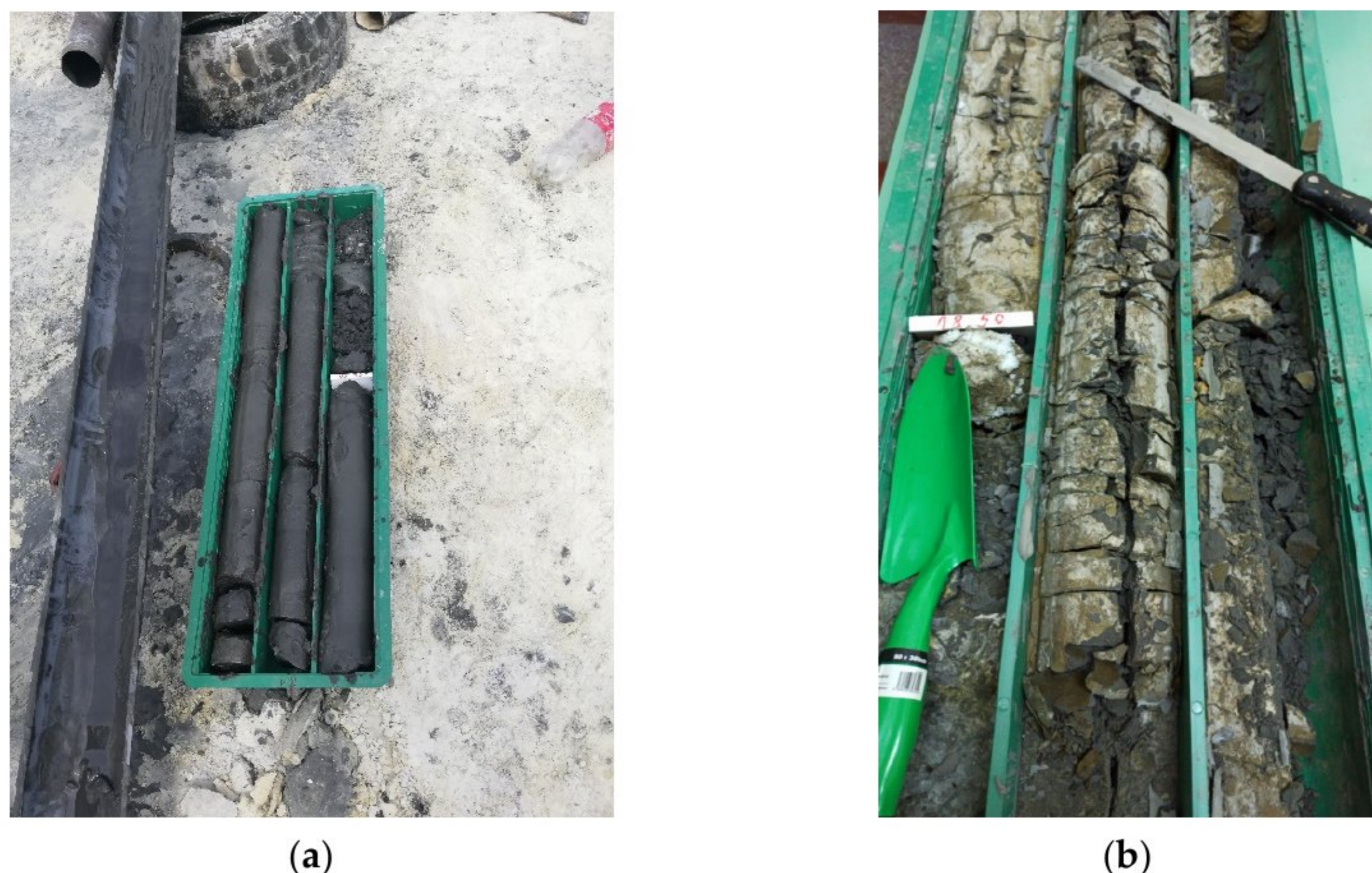


Figure 3. Packing and disposing of the drilled core and sampling. (a) packing of samples on site; (b) preparing samples by longitudinal division.

The sample was used for the experimental static laboratory and percolate leaching tests in the alkaline and acidic range. Chemical analyses were performed using a portable Vanta X-ray fluorescence (Olympus, Parkway, Center Valley, PA, USA) instrument for rapid, accurate, laboratory-quality elemental analyses of solid and liquid phases (Tables 2–4), and the concentration of Zn, As, Ag, Hg, and Pb was measured using a 30 Varian atomic absorption spectrometer (Varian Techtron Pty. Ltd., Mulgrave, Australia). The mineralogical composition of a flotation waste sample is described in many publications, and it was found that the most abundant sulfide mineral was pyrite, and the most abundant barren minerals were quartz and other silicate minerals, with concentrations of 22% and 77%, respectively. The following copper sulfide minerals were found: covellite (0.21%), chalcopyrite (0.16%), enargite (0.14%) and chalcocite (0.04%) [3,5]. The waste sample was analyzed using a Philips XPert SW binary diffractometer (PANalytical B.V., Almelo, Netherlands) with $\text{CuK}\alpha$ radiation (40 kV, 50 mA) equipped with an automatic divergence slit, a sample spinner, and a graphite secondary monochromator. Data were collected for $2\text{--}60^\circ$ 2θ with a step size of 0.05° and a counting time of 30 s per 0.05° . The analysis confirmed the mineralogical composition (wt%): quartz (42%), dominant over dickite (18%) and amorphous glass (10%). The alunite phase was also present in a concentration of 4%, while albite and muscovite were below 2%. The sulfide minerals were pyrite (15%), chalcopyrite (0.14%), enargite (0.08%), covellite (0.02%) and chalcocite (0.04%).

The grain surfaces were examined using the FE MIRA 3 scanning electron microscope (Tescan, Czech Republic) equipped with an energy dispersive XRD (EDX) chemical composition analyzer (Oxford Instruments, Abingdon, UK).

2.2. Experimental Procedures

The leaching experiments were conducted parallel in flasks and plastic columns, each containing 750 g of waste sample and a total medium volume of 6900 mL, which were used for discontinuous static and percolate leaching (Figure 4). The medium for alkaline bioleaching consisted of 4.0 g/L K_2HPO_4 , 0.8 g/L $\text{MgSO}_4\cdot 7\text{H}_2\text{O}$, 0.4 g/L NH_2Cl , and 0.75 g/L Na_2EDTA with a pH of 8.5, and the medium for iron-oxidizing bacteria with a pH of 1.5 consisted of (g/L) 0.5 g K_2HPO_4 , 0.5 g $\text{MgSO}_4\cdot 7\text{H}_2\text{O}$, 1.0 g $(\text{NH}_4)_2\text{SO}_4$, 0.1 g KCl,

and 10 g $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, and for sulfur-oxidizing bacteria with a pH of 4.5 consisted of (g/L) 0.5 g K_2HPO_4 , 0.5 g $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 1.0 g $(\text{NH}_4)_2\text{SO}_4$, 0.1 g KCl , and $\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$.

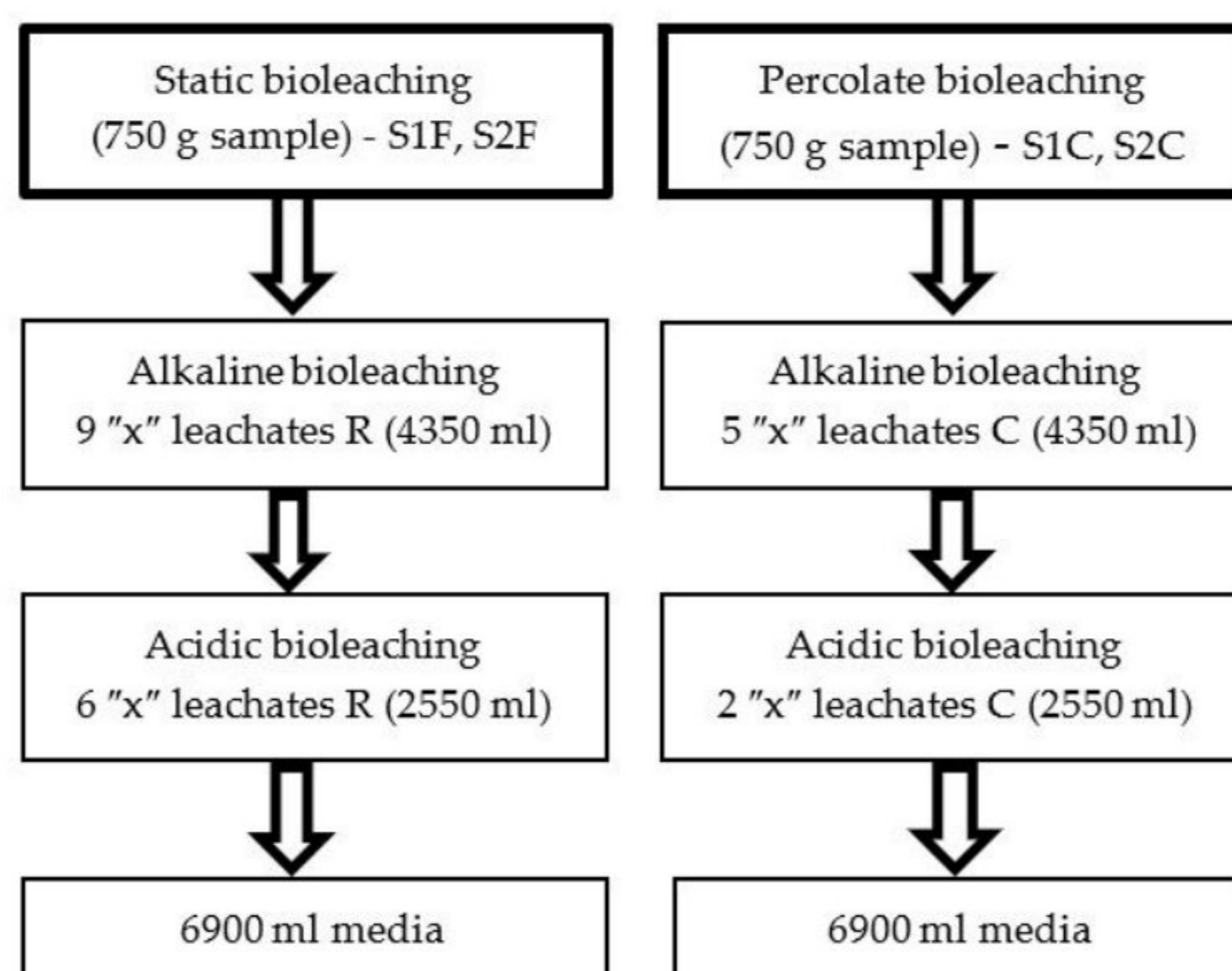


Figure 4. Method of the static and percolate bioleaching process of the waste sample with the replacement or collection leachates (R/C).

Based on the chemical analyses of the leachates, the analyzed concentrations of the extracted elements were compared. The effects of bioleaching with alkaline media and bioleaching with different acidic media were evaluated based on the concentration of the extracted elements (Al, Si, S, Fe, Cu, Zn) after the exchange or collection of the measured media quantities and after mass balance analysis of the main element Cu in the leaching solutions. The consistent effect of bioleaching was also confirmed by chemical analyses of the solid phase after alkaline and acid leaching.

The abiotic control also contained stimulant nutrients, but with 50 mM NaN_3 (sodium azide) to strongly inhibit bacterial growth under natural and resource-rich conditions [11].

In discontinuous static leaching and percolate leaching, alkaline bioleaching pre-treatment was applied nine times by replacing 450–600 mL of solution (total solution—4350 mL) for 9 days. Samples were treated with different mixtures of alkaline (*Bacillus megaterium*, *Bacillus cereus*, *Bacillus mycoides*, *Bacillus pumilus*, *Corynebacterium diphtheriae*, *Pseudomonas* sp. and *Enterobacter* sp.) and then acidic bacterial consortia (*Acidithiobacillus ferrooxidans*, *Leptospirillum ferrooxidans*, *Acidiphilium acidophilum* and *Acidithiobacillus thiooxidans*) *microlive*[®] (7.5 mL with 10^8 CFU/mL) at different pH values. Bacteria were identified using the BBL Crystal Identification System (Becton, Dickinson and Co., Franklin Lakes, NJ, USA).

The flasks and columns were kept under steady-state acidic conditions without aeration for one month, then under aeration with iron-oxidizing bacteria for one month, and with sulfur-oxidizing bacteria at 21 °C for one month. Before sampling, the sample was simply mixed to homogenize the solution. After sedimentation for one minute, the culture (20 mL) was separated from the sample by centrifugation at 6000 rpm for 15 min and the liquid supernatant was collected for elemental analysis. Dissolved elements were monitored six times when the sample medium in the bottles (425 mL) was changed. The solution concentration data for each leachate sample is the average of three measurements.

The parallel samples from static leaching are labelled S1F and S2F, and from percolate leaching S1C and S2C. All column leaching tests were performed with perforated plastic columns with a height of 170 mm and a radius of 80 mm. The height of the 750 g sample bed was about 90 mm. The bottom and top of the columns were lined with silicate filter paper. The surface of the sample was flooded with 30 mL of the medium daily for 85 days.

During the three-month bioleaching test at 21 °C, the leachate was removed five times from the collection container. The volume of medium that flowed through the column (2550 mL, applied continuously) was the same as the volume of medium used in a batch test during the three-month bioleaching.

3. Results and Discussion

Acid mine water containing Cu, Fe and other metals and metalloids contaminates the environment (ground and surface water, sediments, and soils) caused by numerous mining activities, including tailings. The average copper ore content in the 21st century is less than 0.6% copper. High copper grades are found in flotation wastes such as the Bor mine tailings, which produced acidic effluents in the lakes of the Bor mine area. Lake Robule is a highly acidic lake located near the town of Bor at the foot of an open pit mine. With the developed solution for recycling various copper wastes, the extracted copper could be of high value and a substitute for mining primary raw materials and, together with other metals extracted in the process, could significantly reduce pollution of the mine and the surrounding area. The economic extraction of copper from waste requires low-cost processing techniques such as in-situ, ex-situ landfill and heap leaching.

Therefore, in this study, a new mixture of alkaline bacteria was inoculated, and the test of alkaline bioleaching pre-treatment of sulfide waste to remove Cu oxide minerals was carried out, and its main leachable properties were investigated along with Cu and Fe content in leachate and pH effects. The combined leaching of *microlive*[®] in an alkaline medium and iron and sulfur-oxidizing bacteria *microlive*[®] in an acidic medium was also explored experimentally. The results of this study should serve as a suitable benchmark for ecological copper recovery from copper sulfide waste.

3.1. Alkaline Bioleaching

The waste samples (input) contain a high Fe content (8%) and a useful amount of 0.4% Cu, which is why they were used for various leaching experiments to check the leaching effect when extracting the elements. The main disadvantage of bioleaching of sulfides is that it is a slow process. The slow dissolution is attributed to both sulfur- and iron-containing reaction products in the case of chalcopyrite. Natural colonization by indigenous microbial strains that are accustomed to the metals/metalloids in the waste is commonly used in the bioleaching of copper. During the bio-leaching process, solid precipitates may cover the surface of the chalcopyrite.

However, as efforts continue to extract as many metals as possible in an even shorter time, alkaline pre-treatment of bioleaching has been considered to remove metal layers and expose the mineral surface to acidic bacteria.

According to Dew et al. (1999), dissolution of enargite requires a high redox potential. Prior to bacterial inoculation, they pre-leach the heterogeneous sulfide concentrates with ferric sulfate to remove easily oxidizable minerals such as chalcocite. During subsequent bioleaching, redox potentials remained high, favoring the decomposition of enargite but not chalcopyrite [12].

Semiquantitative mineralogical examination of the flotation waste revealed a considerable concentration of carbonate minerals (about 20–25%), indicating a high capacity for acid neutralization, and about 70% of the copper is contained in the oxide mineral cuprite (Cu₂O) [13]. Acid leaching usually produces alumina and silica gels, which cause serious technical and environmental problems, especially in solvent extraction. The local market for sulfuric acid may be limited, and large-scale storage poses a significant safety and environmental problem. Considering the limitations of acid leaching and the functional, health, safety and environmental limitations of other acidic methods, various more environmentally friendly options for leaching Cu residues in the alkaline pH range are being investigated [9].

The collected waste from the Bor mine contained carbonate minerals below 1%, but the layered silicate clay minerals and aluminum potassium sulfate minerals present can

form alumina and silica gels during acid leaching. SEM-EDS analysis confirmed an undesirable concentration of iron, alumina and silica covering the chalcopyrite surface. Alkaline bioleaching decomposed and removed the iron surface coatings of Si and Al on the chalcopyrite surface, which contributed significantly to the overall leaching efficiency (Figure 5).

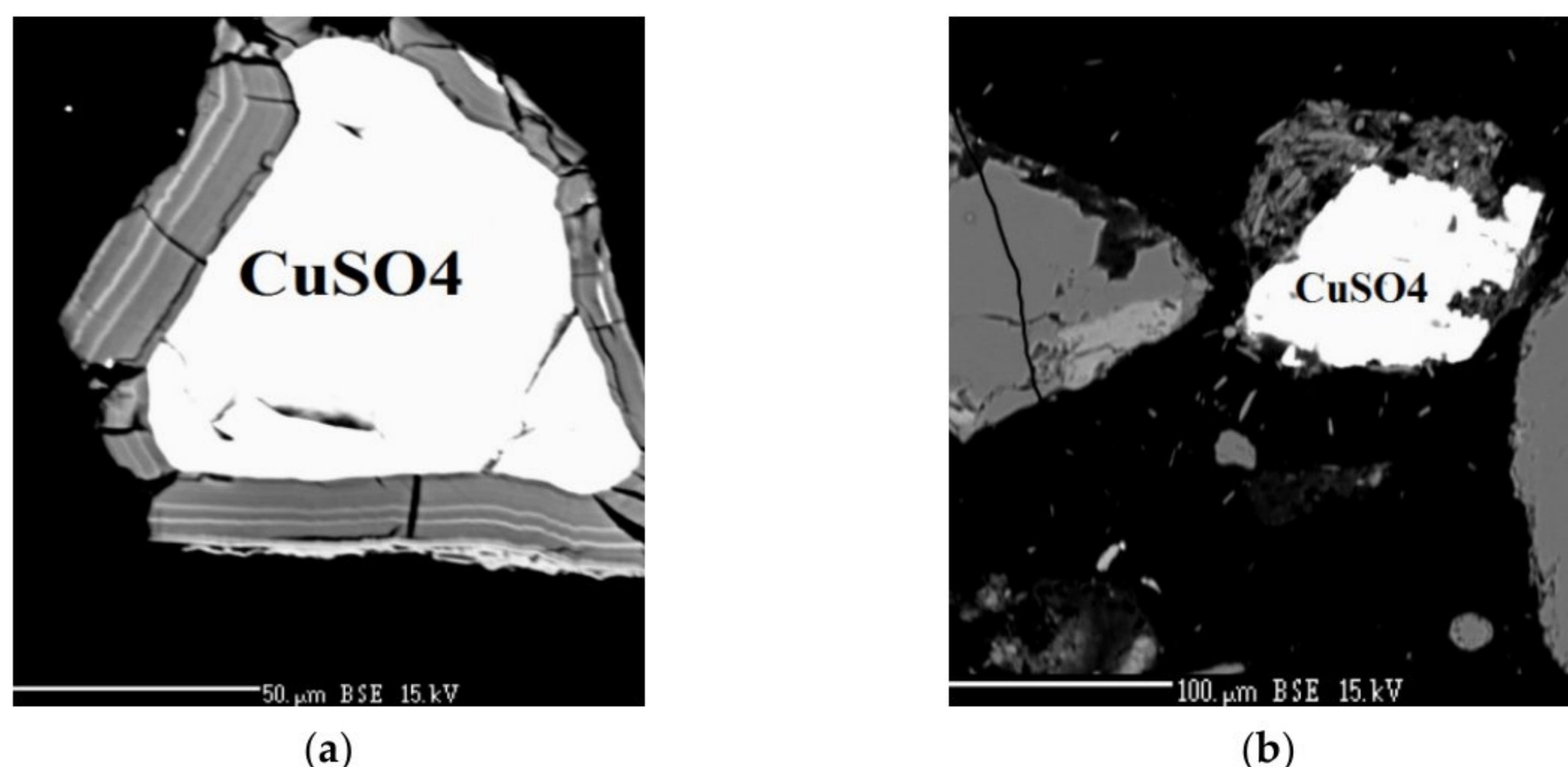


Figure 5. The SEM images chalcopyrite before (a) and after alkaline bioleaching (b).

In our static and percolation leaching experiments of alkaline pre-treatment, the alkaline bacteria were used to remove slightly oxidized minerals at pH 8.5. Nevertheless, pulp density has a great influence on bioleaching performance; numerous experiments have shown that increasing pulp density by more than 15% greatly reduces bioleaching performance [14,15]. This is due to the destruction of bacterial cells by contact with mineral raw materials and the concentration of harmful chemical components in the solution [16]. Discontinuous static bioleaching with media change or percolate bioleaching with removal of extractable toxic elements may be a suitable method for metal toxicity removal.

The effect of alkaline static pre-treatment in the flasks proved the extraction of Cu in considerably high concentrations into the leachate, already during the first media change, with a visible green–blue coloration of the leachate. This Cu concentration (1369 mg/L) gradually decreased with further media changes (Figure 6). High concentrations of Fe (2829 mg/L) and lower concentrations of other elements Zn (9.6 mg/L), As (1.5 mg/L), Sr (9.9 mg/L), Zr (5.2 mg/L), Mo (3.1 mg/L), and Hg (11.6 mg/L) were measured in the leachates. The medium can be recycled after precipitation of the metals in a way that depends on the Cu concentration obtained in the leachates. In abiotic controls with 50 mM NaN_3 added to inhibit bacterial growth, the highest Cu extraction in the leachate was 550 mg/L, which is about half of the concentration obtained during the bioleaching process (data not shown).

Concentrations of Cu above 1000 mg/L in PLS suitable for copper processing by SX/EW technology can be achieved by multistage leaching in a series of chemical reactors [17] or by the proposed alkaline bioleaching. The first to fourth change of medium with a Cu concentration above 500 mg/L corresponded to 1850 mL of alkaline medium with a bioleaching of 750 g of the sample. Further medium changes were not as effective for Cu extraction (Figure 6).

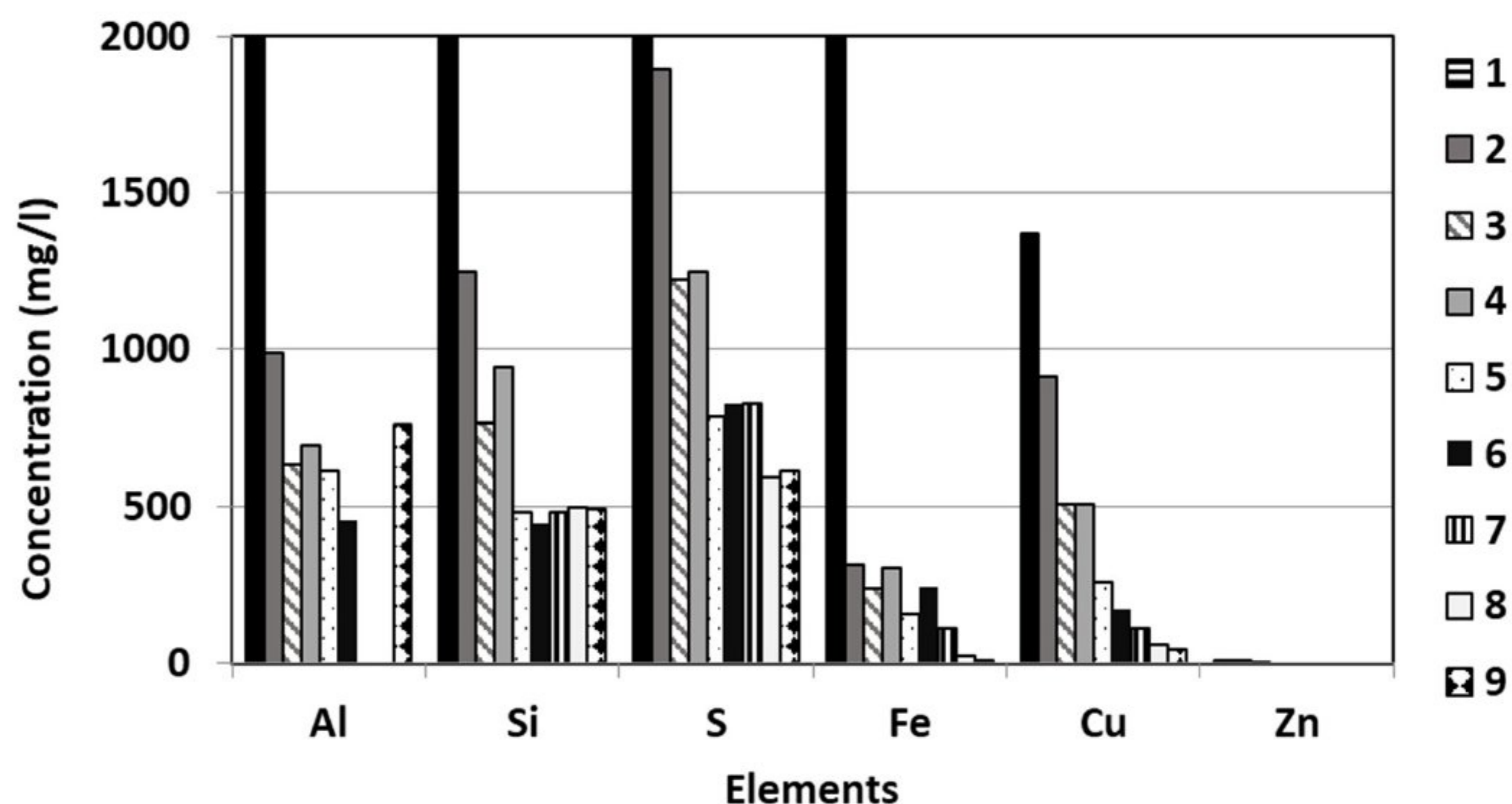


Figure 6. The selected elements extraction during discontinuous static alkaline bioleaching in the replacement 9 leachates (1–9).

Alkaline bioleaching under percolation conditions mimics the leaching of waste in a landfill and shows the significant effect of the first percolation of the medium to obtain an even higher Cu concentration in the leachate of 2200 mg/L (Figure 7) than static bottle leaching. The first chemical analyses were carried out after 1015 mL percolation volume of solution per 750 g sample. Other percolation procedures were not as efficient in Cu extraction as the first percolation cycle. For the abiotic controls, Cu extraction was again half of the bioleached concentrations (data not shown).

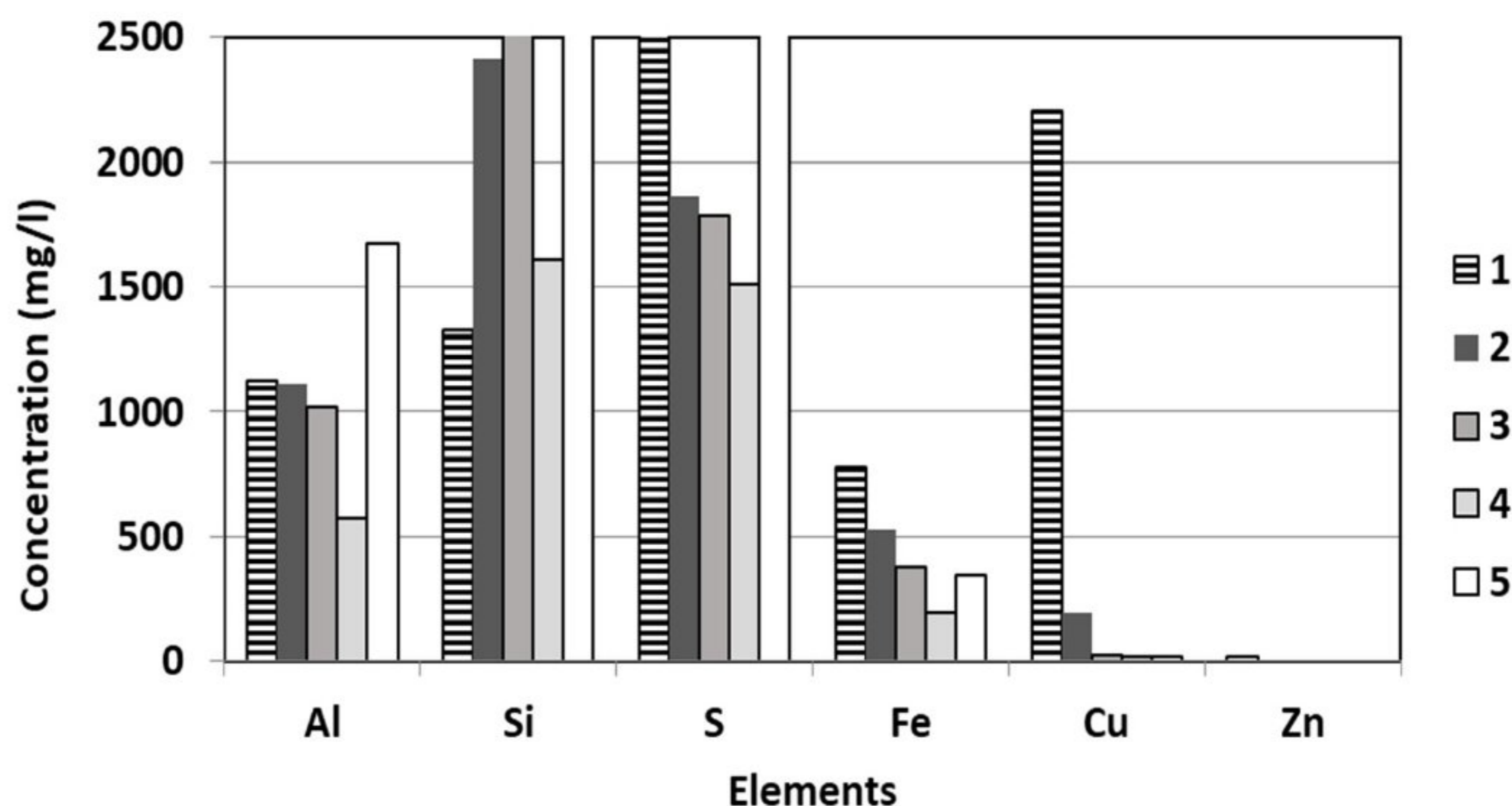


Figure 7. The elements extraction during discontinuous percolate alkaline bioleaching in the replacement 5 leachates (1–5).

The percolation and static tests show that a medium volume of 1015 mL to 1850 mL per 750 g of waste was sufficient to remove extractable Cu in the alkaline bioleach. After the alkaline bioleaching and the removal of extractable Cu, a biological static acidophilic leaching was performed without and with aeration and stimulation of the iron-oxidizing bacteria and then of the sulfur-oxidizing bacteria by nutrients in the medium, which can degrade resistant Cu sulfides.

The dissolution of elements (Al, Si, S, Fe, Cu, Zn) during the alkaline demand tests is shown in Figures 6 and 7 at pH 8.5, and a mass balance analysis of the main element Cu

with leachate replacement was calculated to compare the recovery of this useful element in the leaching solutions. According to the chemical analyses of the leachates, 63% ($\pm 5\%$) of the Cu was dissolved by percolate and static leaching. However, there was a difference in media consumption, as percolate bioleaching required 1015 mL of media for 750 g of sample and static bioleaching required 1850 mL of media for 750 g of sample.

In the parallel bioleaching tests, 63–67% of Cu was extracted, followed by 50% of Zn and 13–25% of Pb. The extent of Fe and S extraction was further influenced by the contribution of the media. A higher Cu extraction rate tends to occur at the beginning of alkaline bioleaching with very high Cu concentrations in the leachates. From the chemical analyses (Table 2) of the solid samples, it appears that the samples contained other metals and metalloids such as As, Hg, or Ag, which were not affected by the bioleaching. These concentrations increased slightly as a function of the decrease in Cu. In alkaline bioleaching, mainly Cu was found under both static and percolate conditions. Silver and gold can be selectively bioleached from the waste after Cu recovery, which will be a next step.

Table 2. The comparison of chemical analyses of the input waste sample and alkaline bioleached samples.

Concentration (%)	Al	Si	S	Mn	Fe	Cu	Zn	As	Ag	Hg	Pb
Input	4.73	21.57	9.6	0.01	8.19	0.43	0.004	0.012	0.015	0.0008	0.008
S1 F	4.54	25.35	10.82	0.01	8.52	0.14	0.002	0.011	0.016	0.0007	0.006
S2 F	5.04	24.98	10.79	0.01	8.38	0.16	0.002	0.014	0.016	0.0011	0.006
S1 C	5.71	24.95	10.32	0.09	9.14	0.14	0.002	0.013	0.017	0.0009	0.007
S2 C	5.79	25.17	10.02	0.09	8.98	0.15	0.002	0.013	0.017	0.0009	0.007

Recently, Han et al. (2018) used an autoclave to study copper leaching from old Bor copper mine flotation waste. The authors found that a H_2SO_4 concentration of 0.5 M, a pressure of 2 MPa, a temperature of 180 °C, a leaching time of 60 min, and a solid phase content of 400 g/L resulted in a leaching efficiency of 98% of the copper [18].

Scientists Antonijevic et al. (2008) [3] reported an average copper leaching of 60% at pH 1 with H_2SO_4 and minimum Fe dissolution values. The extraction of copper and iron increases linearly as the leaching temperature increases from 20 to 60 °C. Copper extraction was 80% after 2 h at 60 °C with the addition of the oxidant ferric sulfate at a concentration of 5 g/L Fe^{3+} ions. The highest degree of copper leaching was observed at a stock consistency of 33%. The effect of stirring speed was investigated at 300, 600, and 900 rpm. The time required to achieve the maximum degree of copper leaching was reduced by increasing the stirring speed from 30 min at 300 rpm to 10 min at 900 rpm. The results showed a maximum copper extraction of 60% after leaching with 0.1 M sulfuric acid. The results suggest a linear relationship between copper dissolution and temperature and a chemically regulated dissolution rate of the copper minerals. Other authors have similarly found rapid dissolution of Cu (85% of the total) from Cu oxide minerals within the first 5 min of sulfuric acid leaching [13].

However, the experiments of these authors required mixing of the samples, a higher temperature, and a very acidic pH, which was not necessary in our alkaline experiments. The experiment proved the possibility of using a simple percolation system with media recycling and obtaining a similar yield of Cu to acid leaching with sulfuric acid, where the handling requires many safety and operational procedures.

3.2. Acidic Bioleaching

Most microbiological studies in the field of bioleaching are carried out with recognized iron- and sulfur-oxidizing bacteria, either as single strains or in mixed cultures. In some cases, however, ore-specific bacteria are not avoided and may even support bioleaching. Furthermore, it is often shown that indigenous microorganisms adapted to high levels

of certain metals in their ecosystem are more efficient as bioleaching catalysts. Adapting microorganisms to a particular mineral system by gradually exposing them to higher concentrations of the major elements present is a common procedure in laboratory experiments. In addition, many significant studies have been conducted on the tolerance of individual strains and cultures to base metal ions [19,20]. Similarly, microbial development in heap farms can be enhanced by adding nutrients to leaching fluids and creating conditions that lead to higher iron concentrations [21].

In our experiments, the first four media changes with a pH of 1.5 were expected to promote the activity of iron-oxidizing bacteria (Figures 6 and 7), but the Cu content only ranged from 44 to 299 mg/L. When a stimulating medium for sulfur-oxidizing bacteria was used at pH 4.5 in 5 and 6 media changes, the Cu content increased significantly above 500 mg/L. The Fe content in the leachate also increased continuously (Figure 8).

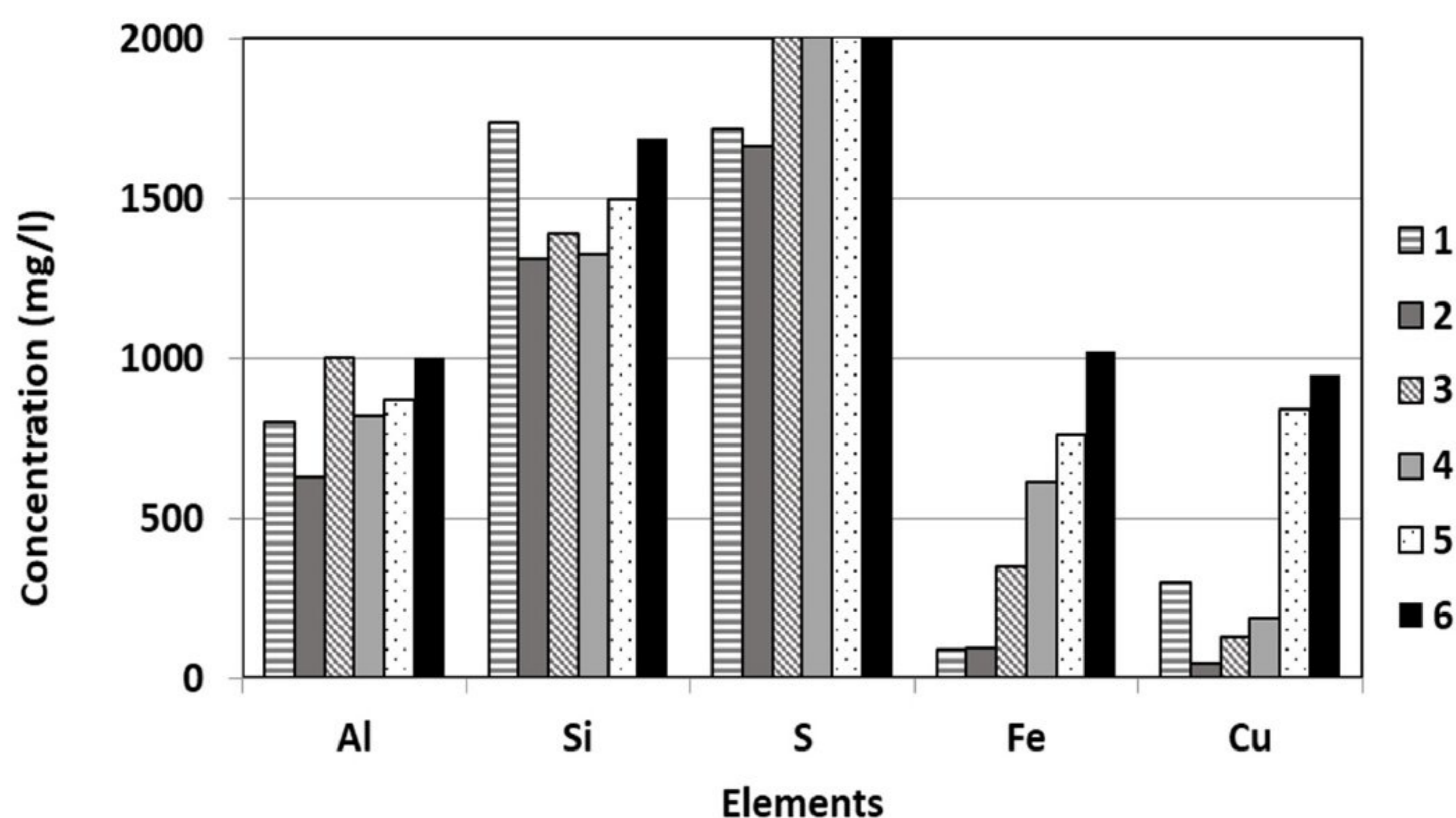


Figure 8. The selected elements extraction during discontinuous static acidic bioleaching in the replacement 6 leachates (1–6).

Static acid bioleaching in aerated bottles can gradually degrade the sulfide minerals, but it takes a month to achieve visible and measurable extraction of Cu and Fe, which would mean further recovery of Cu from the waste (Figure 8). A controlled process of biodegradation and oxidation of sulfide minerals could lend itself to multiple leaching steps and subsequent alkaline bioleaching, intensifying the process of Cu recovery.

During the percolation test of the columns, the acidic leachate (pH 2.4) was collected in the collection tank, which stimulated the formation of iron precipitates due to biological activity, so that the measured leachate analysis misrepresented the Cu and Fe concentrations extracted from the waste sample. The Cu concentration did not exceed 251 mg/L and the Fe concentration reached 716 mg/L (Figure 9), as these precipitates in the collection tank contained Cu (0.2%) and Fe (15%) (Table 3).

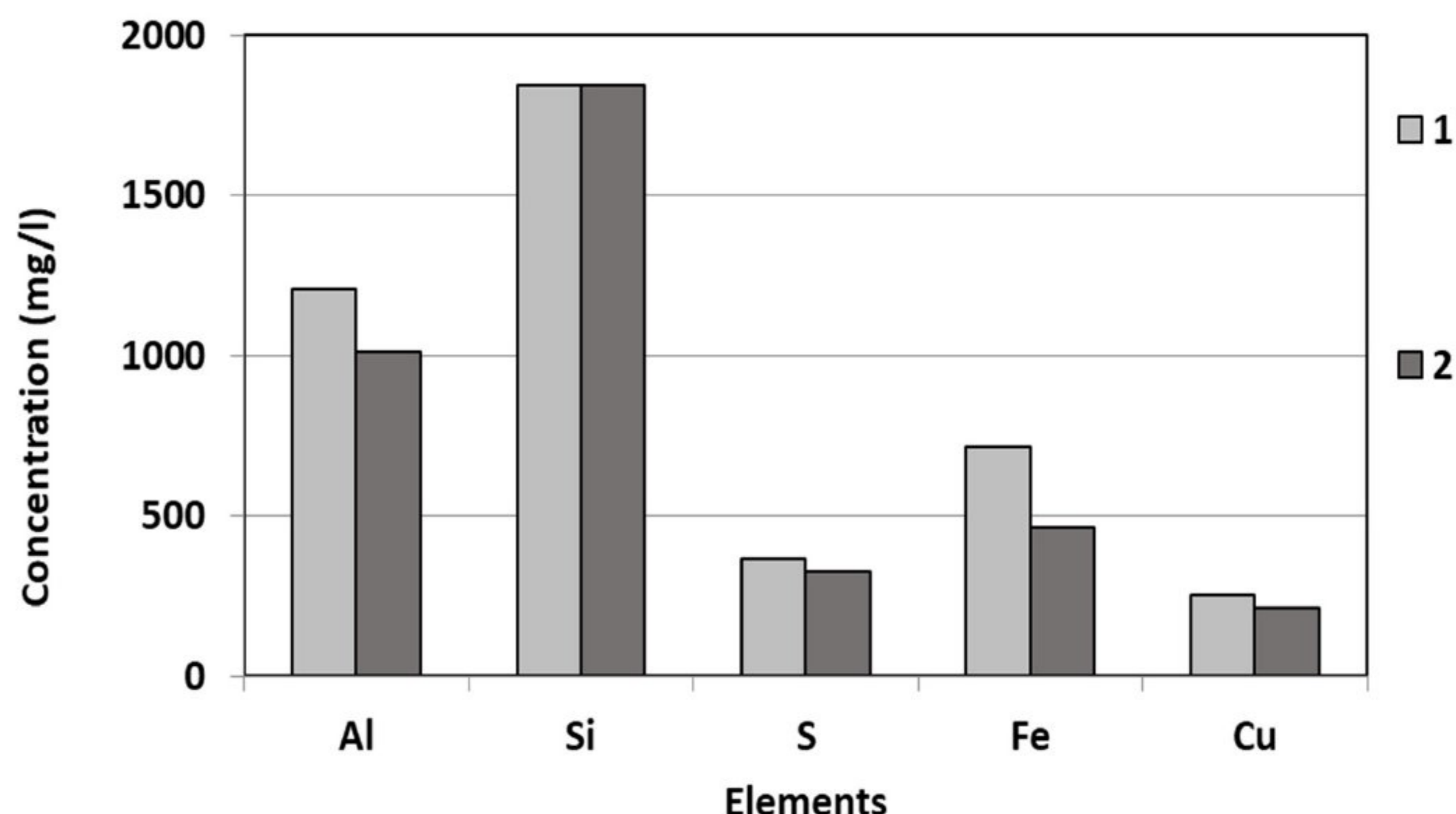


Figure 9. The elements extraction during percolate acidic bioleaching in the replacement 2 leachates (1–2).

Table 3. The chemical analyses of the iron precipitates in leachates after percolation bioleaching media.

Concentration (%)	Al	Si	S	Mn	Fe	Cu	Zn	As	Ag	Hg	Pb
Fe precipitates	1.75	8.36	9.28	0.002	15.41	0.21	0.002	0.02	0.02	0	0.019

Comparing the results of the chemical analyses of the samples after the alkaline bioleaching and the subsequent acidic biological leaching with the input sample, one can confirm the increase of the efficiency of Cu recovery from 67% to 74% by the subsequent acidic biological leaching. The Pb, As, and partially Zn concentrations were increased in the solid sample after percolation of the acidic medium during bioleaching (S1C, S2C), which is associated with a significant decrease of S by 12–23% and a higher decrease of Si than under static conditions (S1F, S2F, Table 4).

Table 4. The comparison of chemical analyses of the input waste sample and bio-chemical leached samples.

Concentration (%)	Al	Si	S	Mn	Fe	Cu	Zn	As	Ag	Hg	Pb
Input	4.7	21.6	9.60	0.01	8.19	0.43	0.004	0.012	0.015	0.0008	0.008
S1 F	4.2	22.9	10.28	0.008	7.21	0.11	0.002	0.012	0.013	0.0009	0.008
S2 F	4.6	25.8	10.92	0.01	8.03	0.12	0.002	0.016	0.017	0.0011	0.014
S1 C	4.5	19.4	7.40	0.009	8.12	0.10	0.003	0.022	0.014	0.0008	0.027
S2 C	4.2	19.5	8.43	0.008	9.40	0.13	0.005	0.027	0.017	0.0009	0.034

Comparing the results from the analysis of the solid phases after alkaline leaching and subsequent acid leaching (Tables 2 and 4), it was found that alkaline leaching increased the concentration of Si > Al > S > Fe especially under percolation conditions with the same yield of Cu and Zn extraction. However, a significant decrease in the enriched concentration of aluminosilicate phases and a decrease in the concentration of Al and Si occurred mainly after the subsequent acidic bioleaching under percolation conditions. The enriched Fe concentration in the alkaline leach decreased more under percolation conditions of the acid

bioleach than in the static bioleach. With the same increase in yield of 7% Cu, it is possible to enrich Pb, As, and partially Zn under acid percolation conditions and selectively recover Cu and S from the solid waste in solution.

4. Conclusions

The flotation waste consists of fine-grained particles and, in the presence of biogeochemical substances (water, atmospheric oxygen, microorganisms), represents a dangerous reactive potential for the extraction of heavy metal ions and the formation of acid mine drainage. In nature, weathering processes convert the sulfide minerals in this deposit to copper sulfate or oxides and make the minerals available for metal mobilization. In our static and percolation leaching experiments of alkaline pre-treatment, the alkaline bacteria easily removed oxidized minerals such as copper oxide and sulfate minerals and metals that covered the surface of the mineral sulfides, and the acidic biological leaching subsequently decomposed the remaining sulfide minerals. The highest copper yield of 67% for alkaline bioleaching was achieved after 9 days, and the subsequent addition of nutrients in acidic medium improved the copper yield to 74%. The alkaline pre-treatment experiment proved the possibility of using a simple percolation system with a lower amount of medium (1015 mL) than static leaching (1850 mL) for 750 g of the sample and achieving a similar Cu yield. The Cu concentration increased significantly above 500 mg/L with sulfur-oxidizing bacteria used at 5 and 6 medium changes with a pH of 4.5 followed by acid leaching. Although further testing is required to determine the combined leaching process, the initial results of our exploratory studies could lead to a cost-effective and environmentally friendly alkaline bioleaching process for selective Cu recovery from tailings.

Author Contributions: Conceptualization: I.R. and D.Š.; funding acquisition: all participants; investigation: all participants; exploratory drilling: I.R. and E.Š.; sampling: I.R. and E.Š.; methodology: D.Š., I.Š. and J.Š.; visualization: all participants; writing—original draft: all participants; writing—review and editing: I.R. and D.Š. All authors have read and agreed to the published version of the manuscript.

Funding: This work was financially supported by EIT RawMaterials in the frame of the RIS CuRE project “Zero waste recovery of copper wastes in the ESEE region”, Grant number–18248.

Data Availability Statement: Not applicable.

Acknowledgments: The authors thank EIT RawMaterials in the frame of the RIS CuRE project for financial support.

Conflicts of Interest: The authors declare no conflict of interest.

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