

Copper leaching from copper-bearing ore after pretreatment with a microbial consortium of bacteria and filamentous fungi

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Abstract

Purpose. To evaluate the effect of pretreating copper-bearing ore with a microbial consortium comprising iron- and sulfur-oxidizing bacteria and filamentous fungi on the efficiency of subsequent sulfuric acid percolation leaching of copper.

Methods. The initial ore was characterized using chemical, X-ray fluorescence, X-ray diffraction, and mineralogical analyses. The thermodynamic stability of copper-bearing compounds was evaluated using an Eh – pH diagram for the Cu – Fe – S system constructed in HSC Chemistry 8. Iron- and sulfur-oxidizing microorganisms and filamentous fungi were isolated and cultivated on selective culture media. Percolation leaching was performed using H₂SO₄ solutions with concentrations of 5, 10, and 15 g/dm³. In 145-day column experiments, conventional sulfuric acid leaching was compared with leaching preceded by biological pretreatment of ore with particle sizes of less than 10 and less than 200 mm.

Findings. The initial ore contained 0.71 wt.% total copper, 0.57-0.58 wt.% oxidized copper, 6.9 wt.% iron, and 2.57 wt.% sulfate ions. Copper was found to occur in both oxide and sulfide forms, including chalcopyrite. The microbial consortium contained iron- and sulfur-oxidizing bacteria, heterotrophic microorganisms, and filamentous fungi. Increasing the H₂SO₄ concentration from 5 to 15 g/dm³ increased copper recovery from 42.42 to 46.69%, while a concentration of 10 g/dm³ was selected for further experiments. Biological pretreatment increased copper recovery from ore with a particle size of less than 10 mm from 56.75 to 72.0% and from ore with a particle size of less than 200 mm from 54.45 to 66.3%.

Originality. The effect of ore pretreatment with a consortium of iron- and sulfur-oxidizing bacteria and filamentous fungi on the subsequent sulfuric acid leaching of copper from ore with different particle sizes was experimentally established. The combined action of bacterial oxidation of iron- and sulfur-bearing compounds and metabolites produced by filamentous fungi was shown to increase the accessibility of copper-bearing mineral phases to acid dissolution.

Practical implications. The results can be used to substantiate the parameters of combined biological pretreatment and sulfuric acid leaching of low-grade and mineralogically complex copper ores, as well as to develop heap-leaching flowsheets for copper-bearing mineral resources.

Keywords: copper-bearing ore; percolation leaching; column leaching; sulfuric acid; biological oxidation; bacteria; filamentous fungi; copper recovery

1. Introduction

The depletion of high-grade copper ore reserves and the gradual decline in valuable component contents in mined mineral resources necessitate the processing of low-grade, off-balance, and mineralogically complex ores. Conventional beneficiation and pyrometallurgical processing methods do not always provide the required technological and economic efficiency for such raw materials. Consequently, hydrometallurgical methods are becoming increasingly important, as they enable copper recovery from low-grade and refractory mineral resources, as well as from copper-bearing industrial waste materials [1]-[5]. An additional advantage of these methods is that they allow raw materials to be processed without high-temperature operations and enable technologi-

cal flowsheets to be adapted to different ore types and geological conditions.

When selecting a mineral-processing technology, consideration should be given not only to the recovery of the target component but also to the resource efficiency, environmental performance, and operational effectiveness of the entire technological chain. Pyrometallurgical methods remain important for processing complex ores and concentrates, whereas hydrometallurgical flowsheets allow leaching to be integrated with the subsequent treatment of pregnant leach solutions, including membrane-based and electrochemical separation processes. The industrial implementation of such processes requires efficient water use and recycling, the mitigation of environmental and occupational health risks,

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and, in the case of borehole-based geotechnologies, reliable isolation of the treatment intervals and effective management of technical risks [6]-[11].

The efficiency of hydrometallurgical processing of copper ores is largely governed by the modes of copper occurrence and the mineralogical composition of the feed material. Copper oxide minerals dissolve relatively readily in acidic media, whereas copper recovery from sulfide minerals, particularly chalcopyrite, is hindered by slow dissolution kinetics and the formation of passivating surface layers. When processing mixed oxide-sulfide ores, the selection of leaching parameters should account for the relative proportions of the mineral phases, ore particle size, degree of liberation of copper-bearing minerals, acid-consuming capacity of the gangue, and the redox conditions of the process [4], [12]-[15].

Combined process flowsheets are frequently used to treat complex and refractory mineral raw materials. Such flowsheets involve preliminary modification of the phase composition or reactivity of the mineral matrix. Depending on the feed composition, pretreatment may include thermal processing, roasting with chemical reagents, electrochemical oxidation, or microwave irradiation followed by leaching [16]-[19]. In this context, biological pre-oxidation can be regarded as a distinct pretreatment method for enhancing the susceptibility of sulfide minerals to subsequent hydrometallurgical metal recovery.

Heap leaching is one of the technically feasible methods for processing low-grade copper ores. In this process, the leaching solution percolates through a bed of previously prepared mineral material [20]. Laboratory-scale percolation tests make it possible to reproduce specific features of this process and evaluate the effects of ore particle size, leaching reagent concentration, irrigation duration, and feed pretreatment conditions [21]. Increasing the degree of ore comminution promotes the liberation of valuable minerals and enlarges the surface area available for contact with the leaching solution. At the same time, however, finer comminution may increase acid consumption because of enhanced interaction between the acid and gangue minerals.

In addition to the chemical activity of the leaching reagent, the efficiency of geotechnological processes is governed by the conditions of solution transport through the ore mass. Filtration uniformity, permeability of the mineral bed, and the physicochemical properties and activation state of the solution affect both the duration of contact with copper-bearing minerals and the intensity of mass transfer [22]-[24]. In borehole leaching systems, the process is additionally affected by clogging of the pore and filtration space, which reduces the injectivity of the ore-bearing rock mass. Chemical, hydrodynamic, and mechanical stimulation methods are therefore employed to restore permeability [25]-[28]. Although these studies primarily address other metals and technological schemes, the identified patterns confirm that hydrodynamic conditions must be considered when interpreting the results of percolation leaching tests.

One promising approach to improving copper recovery from sulfide and mixed ores is bioleaching. Bio-oxidative pretreatment can increase the accessibility of metals contained in sulfide minerals for subsequent hydrometallurgical recovery [29], [30]. In such processes, acidophilic iron- and sulfur-oxidizing microorganisms catalyze the transformation of iron- and sulfur-bearing compounds. *Acidithiobacillus*

ferrooxidans oxidizes Fe^{2+} to Fe^{3+} , which acts as an oxidizing agent for sulfide minerals, and also contributes to the oxidation of reduced sulfur compounds. The regeneration of Fe^{3+} and formation of sulfuric acid maintain conditions favorable for the transfer of copper from the mineral phase into solution [4], [12], [14], [31], [32]. Under acidic conditions, the oxidative dissolution of copper sulfides may proceed through the action of dissolved oxygen and Fe^{3+} ions, while microbial activity continuously regenerates the oxidizing capacity of the leaching solution [33].

The practical applicability of bacterial Fe^{2+} oxidation has also been demonstrated in other hydrometallurgical systems. Studies on in situ uranium leaching have investigated the bioactivation of circulating solutions, the industrial application of bacterial iron oxidation, and the use of flow-through bioreactors containing immobilized *A. ferrooxidans* cells [34]-[36]. Despite differences in the target metal and mineral feed composition, these studies confirm the universal technological function of iron-oxidizing bacteria, namely the regeneration of Fe^{3+} and maintenance of the oxidizing capacity of acidic leaching media.

Published studies indicate that mixed cultures comprising iron- and sulfur-oxidizing microorganisms can promote more effective chalcopyrite dissolution than individual pure cultures. This improvement is attributed to the functional complementarity of microorganisms involved in Fe^{3+} regeneration, elemental sulfur oxidation, and the removal of sulfur-bearing products from mineral surfaces [37]. In the bioleaching of mixed oxide-sulfide copper ores, process efficiency depends not only on microbial activity but also on the ability of microorganisms to colonize mineral particle surfaces. Sulfur-oxidizing microorganisms may be particularly important because they maintain solution acidity and oxidize sulfur and polysulfide compounds formed on mineral surfaces [38].

The use of microscopic fungi represents another promising direction in biohydrometallurgy. Species belonging to the genera *Aspergillus*, *Penicillium*, and *Trichoderma* are capable of producing organic acids and other metabolites that decrease the pH of the medium, interact with the mineral matrix, and form soluble complexes with metals. Particular attention has been given to *Aspergillus niger*, whose applicability to the bioleaching of metals from low-grade ores and sulfide-bearing materials has been demonstrated experimentally [32], [39], [40]. The activity of filamentous fungi may complement the oxidative action of iron- and sulfur-oxidizing bacteria by increasing the accessibility of copper-bearing phases to subsequent acid leaching.

The combined use of different groups of microorganisms may integrate several mechanisms of interaction with mineral raw materials. Iron-oxidizing bacteria promote Fe^{3+} regeneration, sulfur-oxidizing microorganisms maintain the acidity of the medium and prevent the accumulation of certain sulfur-bearing products, whereas filamentous fungi affect the mineral matrix through the production of organic acids and complexing agents. The effectiveness of such a microbial consortium depends on the mineralogical and chemical composition of the ore, particle size, accessibility of mineral surfaces, medium acidity, and treatment duration.

Despite the considerable number of studies devoted to the bacterial leaching of copper sulfides and the separate application of filamentous fungi, the combined use of bacteria and fungi during the prolonged percolation leaching of complex

copper ores remains insufficiently investigated. Data on the effect of such biological pretreatment on copper recovery from ores of different particle sizes under conditions simulating heap leaching are still limited. Addressing this issue is particularly relevant for ores from the Balkhash region because of the heterogeneous distribution of copper, the presence of both oxide and sulfide mineral phases, and the high proportion of silicate minerals.

The purpose of this study is to evaluate the effect of ore pretreatment with a microbial consortium comprising iron- and sulfur-oxidizing bacteria and filamentous fungi on the efficiency of subsequent sulfuric acid percolation leaching of copper from ore originating from the Balkhash region.

To achieve this purpose, the chemical, phase, and mineralogical compositions of the initial ore were characterized. Preliminary tests were conducted using different sulfuric acid concentrations, followed by comparative column experiments involving conventional sulfuric acid leaching and leaching after the biological pretreatment of ore fractions with different particle sizes.

2. Methods

Copper-bearing ore from the Balkhash region was used as the study material. The initial sample consisted of consolidated ore fragments and clay-rich material (Fig. 1).



Figure 1. Preparation of the initial copper ore sample

Characterization of the ore included multielement X-ray fluorescence analysis, X-ray diffraction analysis, mineralogical examination, and chemical analysis. Copper content was determined both in the initial ore and in the solid residues obtained after leaching. Pregnant leach solutions were analyzed by atomic absorption spectrometry.

Semiquantitative X-ray fluorescence analysis was performed using an Axios 1 kW wavelength-dispersive X-ray fluorescence spectrometer manufactured by PANalytical, the Netherlands. The analytical data were processed using SuperQ software and the Omnia 37 software package based on the fundamental parameters method. The uncertainty of the semiquantitative analysis was $\pm 20\%$.

The microstructure and local elemental composition of the ore minerals were examined using a JXA-8230 electron probe microanalyzer manufactured by JEOL Ltd., Tokyo, Japan.

To assess the thermodynamic stability of copper-bearing phases, a Pourbaix diagram for the Cu-S-Fe system was constructed at 25°C using HSC Chemistry 8 software.

Culture media were selected and modified for the isolation and cultivation of microorganisms, taking into account

the growth requirements of acidophilic iron- and sulfur-oxidizing microorganisms [14], [41]. Acidophilic iron-oxidizing microorganisms were isolated primarily using specialized culture media containing ferrous sulfate at pH values ranging from 1.8 to 3.0 [41]. No organic carbon source was added because chemolithotrophic microorganisms utilize atmospheric carbon dioxide as their carbon source. When sulfide ore samples were examined, the potential presence and growth of chemoorganoheterotrophic bacteria and filamentous fungi were also considered [14], [32], [39], [40].

Laboratory percolation tests were conducted to evaluate the effects of ore particle size, sulfuric acid concentration, and biological pretreatment on copper recovery. Before the preliminary tests, the ore was crushed in a jaw crusher to a particle size of less than 25 mm and screened through a sieve with 25 mm apertures. The masses of the resulting particle-size fractions were as follows: $-25 + 15$ mm, 3.0 kg; $-15 + 10$ mm, 19.5 kg; and $-10 + 2.5$ mm, 7.5 kg.

The prepared ore samples were loaded into three percolation columns, with 10 kg of material placed in each column. Sulfuric acid solutions with concentrations of 5, 10, and 15 g/dm³ were used for irrigation. Before leaching commenced, the ore in each column was pre-wetted to saturation using a sulfuric acid solution of the corresponding concentration.

The pregnant leach solutions were analyzed daily for copper, iron, and free sulfuric acid concentrations. The solution volume and pH were also measured. A schematic diagram of the percolation leaching setup is presented in Figure 2.

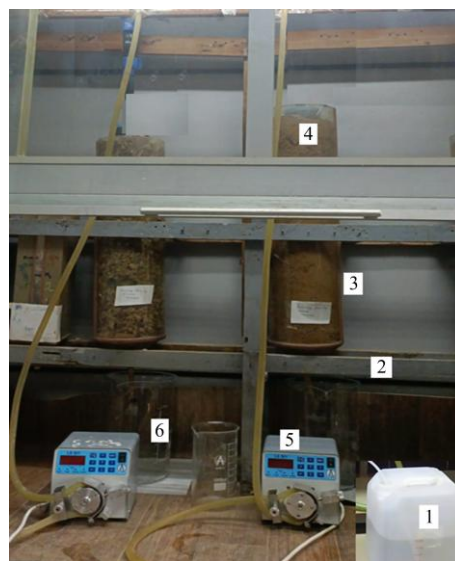


Figure 2. Experimental setup for ore percolation leaching: 1 – irrigation solution tank; 2 – supporting frame; 3 – percolation columns; 4 – ore; 5 – pump; 6 – collection tank

The main column tests were conducted using crushed and uncrushed ore samples with different particle-size distributions:

– variant A – crushed ore with a particle size of less than 10 mm: $-10 + 5$ mm – 47 kg; $-5 + 2.5$ mm – 47 kg; and $-2.5 + 0$ mm – 46 kg;

– variant B – uncrushed ore with a particle size of less than 200 mm: $-200 + 100$ mm – 50 kg; $-100 + 50$ mm – 50 kg; and $-50 + 10$ mm – 56 kg;

– variant C – crushed ore with a particle size of less than 10 mm: $-10 + 5$ mm – 47 kg; $-5 + 2.5$ mm – 47 kg; and

-2.5 + 0 mm – 46 kg, pretreated with a microbial consortium comprising bacteria and filamentous fungi.

The tests were conducted in titanium percolation columns with a diameter of 0.5 m and a height of 1.38 m. The height of the ore bed in the columns ranged from 0.48 to 0.49 m. The column radius was 0.258 m, while the irrigation area, calculated using the equation ($S = \pi R^2$), was 0.209 m².

An overall view of the laboratory setup used for the column experiments is shown in Figure 3.



Figure 3. Laboratory setup for ore leaching tests conducted in titanium percolation columns

Leaching was performed using sulfuric acid solutions. The solution acidity was maintained within a pH range of 1.2-1.5, corresponding to a sulfuric acid concentration of approximately 5 g/dm³. Biological pretreatment of the ore was carried out using a microbial consortium comprising bacteria and filamentous fungi at pH 1.8-2.0.

A filter cloth with a diameter matching the internal diameter of the column was placed on the ore surface to ensure uniform distribution of the irrigation solution. The solution was supplied using a peristaltic pump.

Each column experiment lasted 145 days. Irrigation was performed continuously without operational interruptions. During leaching, the pH, oxidation-reduction potential (ORP), and copper and iron concentrations in the pregnant leach solutions were monitored.

Upon completion of the leaching experiments, the ore was washed with water until the residual concentrations of copper, iron, and sulfuric acid in the wash solutions decreased to sufficiently low levels. The pH was measured using a Consort 930 meter. Copper and iron concentrations were determined by atomic absorption spectrometry, with parallel verification by titrimetric analysis. The residual acid concentration was determined titrimetrically.

3. Results and discussion

3.1. Thermodynamic assessment of copper-bearing mineral behavior

The dissolution of oxide copper minerals proceeds considerably more readily than that of sulfide minerals. The Pourbaix diagram for the Cu – S – Fe system at 25°C shows that the stability fields of individual copper-bearing phases are governed by the pH and oxidation-reduction potential of the medium (Fig. 4).

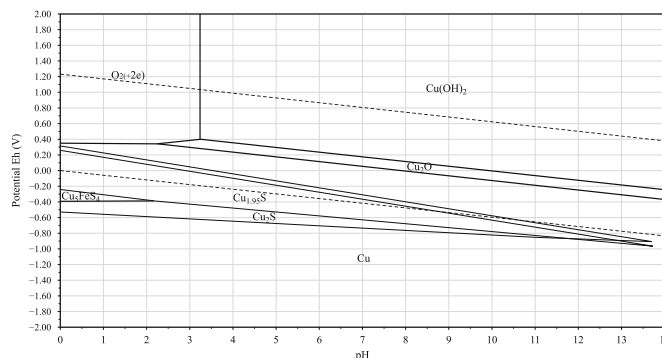


Figure 4. Pourbaix diagram for the Cu – S – Fe system at 25°C

Within the pH range of 0-1 and at Eh values from -0.40 to 0.20 V, bornite (Cu_5FeS_4) occurs within its thermodynamic stability field. The stability fields of the copper sulfide phases $Cu_{1.95}S$ and Cu_2S are located at lower oxidation-reduction potentials. The results indicate that decreasing the pH creates favorable conditions for the dissolution of oxide copper minerals and the transfer of copper into solution. In contrast, the dissolution of sulfide minerals requires not only an acidic medium but also the maintenance of an appropriate oxidation-reduction potential and the presence of an oxidizing agent.

3.2. Chemical, phase, and mineralogical composition of the ore

According to the chemical analysis, the contents of the major components in the investigated ore were as follows, wt.%: total copper, 0.71; oxidized copper, 0.57-0.58; SO_4 , 2.57; and Fe, 6.9. Based on its oxidized copper content, the investigated material is classified as an oxidized ore. However, the phase and mineralogical analyses revealed the presence of copper sulfide minerals, indicating the complex mineralogical composition of the ore. The results of the X-ray diffraction analysis are presented in Table 1.

Table 1. Results of X-ray diffraction analysis of the ore

Mineral	Formula	Content, wt. %
Albite	$Na(AlSi_3O_8)$	24.6
Quartz	SiO_2	21.2
Chalcopyrite	$CuFeS_2$	18.4
Clinocllore	$Al_2Mg_5Si_3O_{10}(OH)_8$	8.6
Gypsum	$CaSO_4 \cdot 2H_2O$	6.9
Tremolite	$(Ca,Na,Fe)_2Mg_5Si_8O_{22}(OH)_2$	6.6
Muscovite	$KAl_2(AlSi_3O_{10})(OH)_2$	5.6
Kaolinite	$Al_2Si_2O_5(OH)_4$	4.0
Laumontite	$CaAl_2Si_4O_{12} \cdot 4H_2O$	3.1
Rutile	TiO_2	1.0

The principal mineral phases identified in the investigated ore were albite, quartz, and chalcopyrite. According to the X-ray diffraction results, the chalcopyrite content was 18.4 wt.%. The gangue mineral assemblage also contained clinocllore, gypsum, tremolite, muscovite, kaolinite, laumontite, and rutile.

According to the X-ray fluorescence analysis, the ore is characterized by a predominantly silicate composition, as indicated by its high oxygen and silicon contents. Relatively high concentrations of aluminum, calcium, and iron were also detected. The results of the multielement analysis are presented in Table 2.

Table 2. Results of automated multielement X-ray fluorescence analysis of the initial ore samples from the deposit

Element	Content, wt.%	Element	Content, wt.%
O	36.248	Cr	0.014
Na	1.293	Mn	0.112
Mg	1.782	Fe	6.916
Al	5.092	Co	0.015
Si	19.049	Cu	1.059
P	0.172	Zn	0.041
S	2.021	Rb	0.006
Cl	0.145	Sr	0.044
K	1.384	Zr	0.021
Ca	5.739	Nb	0.003
Ti	0.795	Mo	0.011
Y	0.010		

Mineralogical analysis showed that the investigated samples consist predominantly of silicate rocks, mainly quartzized and K-feldspar-altered granodiorites affected by metasomatic alteration to varying degrees. The rocks exhibit mottled, massive, and fractured textures. Their color ranges from white and light gray to pink and black. The average rock density is approximately 2.7 g/cm³, while the Mohs hardness ranges from 5 to 6.

In most samples, iron oxides and hydroxides are well developed, imparting brownish or yellowish hues to the rock. The presence of chlorite and copper carbonates gives some areas a greenish coloration.

Copper in the investigated ore occurs predominantly as chalcopyrite, together with secondary sulfides such as chalcocite, covellite, and bornite. Chalcopyrite is present mainly as disseminations within the gangue matrix or as irregular aggregates measuring 0.1-0.3 mm. Many chalcopyrite grains are oxidized and are rimmed by chalcocite, covellite, and bornite. Individual chalcocite and covellite grains range in size from 20 to 60 μm.

Copper sulfides are mainly associated with quartz veinlets, magnetite nests, and zones enriched in iron oxides. The ore is characterized by disseminated, interstitial, and veinlet textures (Figs. 5 and 6). Thin malachite veinlets, 10-20 μm thick, were identified in individual samples. Less commonly, zones were observed in which malachite occupied up to 20% of the polished-section area. Fine disseminations of native copper were detected within widened portions of the malachite veinlets. Chrysocolla was also present in minor amounts.

Magnetite or titanomagnetite was identified in nearly all of the investigated samples. Magnetite occurs as isometric grains and aggregates ranging from 0.3 to 1.0-1.5 mm in size. Along their margins, magnetite grains are partially or completely replaced by iron hydroxides, predominantly goethite. Ilmenite lamellae were also observed within titanomagnetite, forming lattice and mosaic textures. Pyrite occurs as cubic grains 20-30 μm in size and as isolated aggregates located mainly along quartz veinlets.

Scanning electron microscopy confirmed the presence of copper-bearing inclusions within sulfide minerals and the silicate matrix. Electron images and the results of local elemental analysis are presented in Figure 7.

The results confirm the heterogeneous distribution of copper within the investigated ore. A portion of the copper is associated with sulfide minerals and occurs as fine inclusions within the silicate matrix.

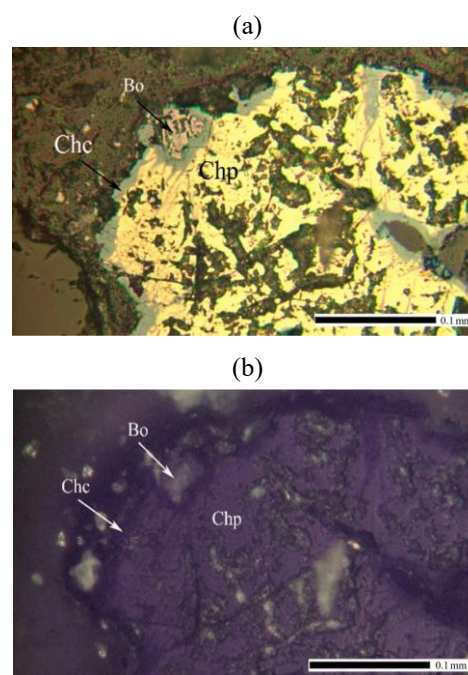


Figure 5. Copper ore sample from the Balkhash region (polished section 1 – chalcopyrite, chalcocite, and bornite; magnification 40×): (a) plane-polarized light; (b) crossed polars

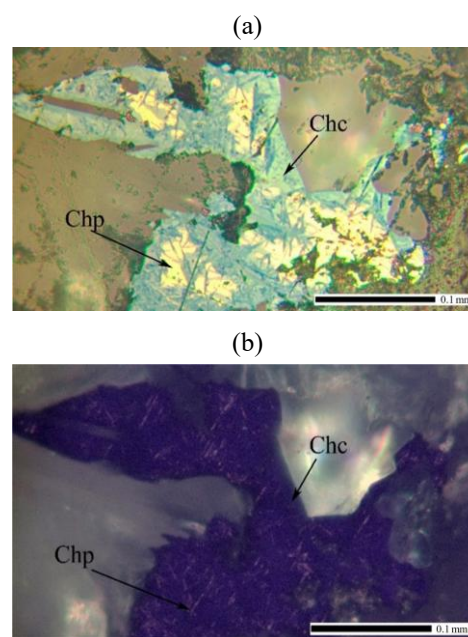


Figure 6. Copper ore sample from the Balkhash region (polished section 2 – chalcocite and chalcopyrite; magnification 40×): (a) plane-polarized light; (b) crossed polars

These mineralogical features may hinder direct sulfuric acid leaching because the enclosing mineral matrix and surface coatings restrict contact between the leaching solution and the copper-bearing phases [42].

3.3. Results of microbiological investigations

Chemolithotrophic microorganisms were detected in the ore samples, including the iron- and sulfur-oxidizing bacteria *Acidithiobacillus ferrooxidans* and *Acidithiobacillus thiooxidans*, representatives of the genera *Leptospirillum* and *Sulfobacillus*, as well as archaea. Chemoorganoheterotrophic bacteria and filamentous fungi were also identified.

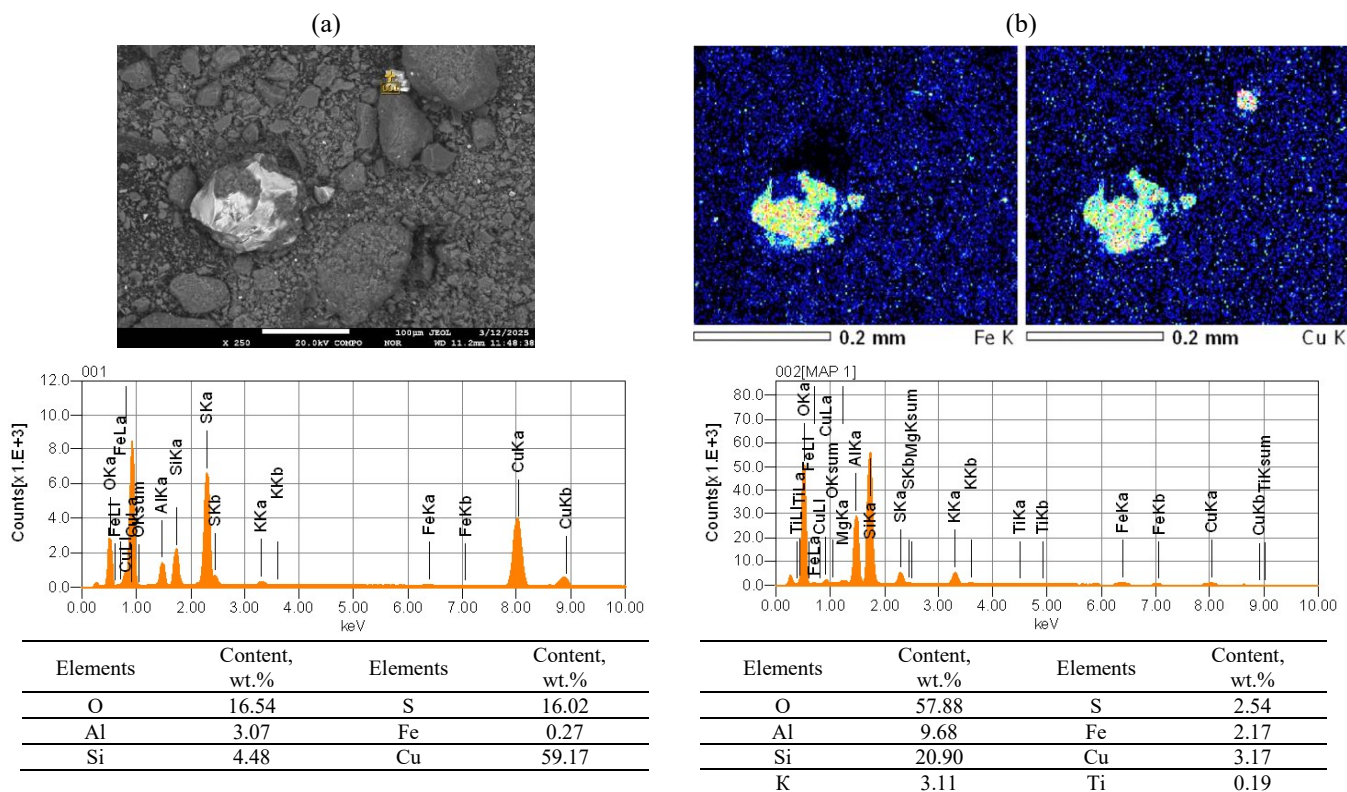


Figure 7. Electron images of copper-bearing mineral particles: (a) chalcopyrite; (b) sulfide inclusions within silicate fragments

Under conditions favorable for the growth of iron- and sulfur-oxidizing bacteria, active proliferation of the fungal microbiota was observed. This made it possible to use a microbial consortium comprising bacteria and filamentous fungi for ore pretreatment prior to sulfuric acid leaching.

The presence of different microbial groups may provide a combined effect on the mineral matrix. Iron- and sulfur-oxidizing bacteria contribute to Fe³⁺ regeneration and the oxidation of reduced sulfur compounds, whereas filamentous fungi are capable of producing organic acids and increasing metal accessibility for subsequent dissolution [14], [39], [40], [43].

3.4. Preliminary percolation tests

The preliminary tests were conducted for 25 days using sulfuric acid irrigation solutions with concentrations of 5, 10, and 15 g/dm³. The relationship between copper recovery and leaching duration is shown in Figure 8.

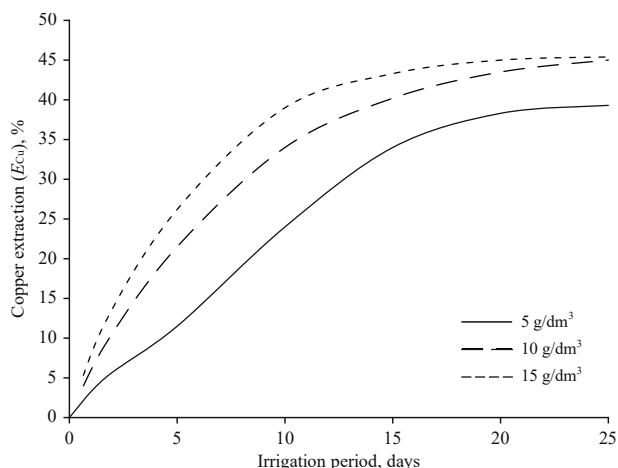


Figure 8. Copper recovery as a function of preliminary percolation leaching duration

When a sulfuric acid solution with a concentration of 5 g/dm³ was used, copper recovery reached 42.42%. Increasing the acid concentration to 10 g/dm³ raised copper recovery to 45.17%, while a concentration of 15 g/dm³ resulted in a recovery of 46.69%.

Copper recovery increased with increasing sulfuric acid concentration. However, the difference between the results obtained at concentrations of 10 and 15 g/dm³ was relatively small. Considering sulfuric acid consumption and the requirements for subsequent treatment of the pregnant leach solutions, an H₂SO₄ concentration of 10 g/dm³ was selected for further investigation. Based on the preliminary test results, the -25 + 15 mm fraction was identified as the preferred ore particle-size fraction.

3.5. Results of the main column tests

The main column tests compared conventional sulfuric acid leaching with leaching preceded by biological pretreatment of the ore using a microbial consortium comprising bacteria and filamentous fungi. The tests were conducted for 145 days (Table 3).

Table 3. Copper ore leaching performance after 145 days

Parameter	Ore particle size, mm			
	< 10 mm		< 200 mm	
	Conventional leaching	Biological pretreatment	Conventional leaching	Biological pretreatment
Cu recovery, %	56.75	72.0	54.45	66.3

For crushed ore with a particle size of less than 10 mm, biological pretreatment increased copper recovery from 56.75 to 72.0%, corresponding to an increase of 15.25 percentage points. For coarse ore with a particle size of less than

200 mm, copper recovery increased from 54.45 to 66.3%, representing an increase of 11.85 percentage points.

For both particle-size classes, pretreatment with bacteria and filamentous fungi resulted in higher copper recovery than conventional sulfuric acid leaching. The highest copper recovery, 72.0%, was obtained for crushed ore with a particle size of less than 10 mm.

Coarse ore particles have a lower specific surface area; therefore, the acid reacts primarily with their external surfaces, while minerals enclosed within the particles remain less accessible to the leaching solution. Nevertheless, biological pretreatment also increased copper recovery from the coarse ore.

The observed effect may be attributed to the preliminary biological oxidation of sulfide minerals, the regeneration of Fe^{3+} ions by iron-oxidizing bacteria, and the action of organic acids produced by filamentous fungi. This combined effect promotes alteration of sulfide mineral surfaces and increases copper accessibility during subsequent sulfuric acid leaching [14], [32], [39], [40], [43].

Thus, the column test results demonstrated the advantage of the combined process involving biological pretreatment of the ore with a microbial consortium comprising bacteria and filamentous fungi, followed by sulfuric acid leaching. Process efficiency is governed not only by the application of biological pretreatment but also by the phase composition, structural characteristics, and particle size of the initial ore.

The obtained results are consistent with findings reported in international studies on the biological enhancement of copper-bearing material leaching. Fu et al. [37] found that mixed cultures of iron- and sulfur-oxidizing microorganisms promoted more effective chalcopyrite dissolution than individual pure cultures. Zhao et al. [38] showed that *Acidithiobacillus ferrooxidans* promotes chalcopyrite oxidation, attaches preferentially to defective areas of the mineral surface, and induces structural and chemical changes in the mineral. In a study of mixed oxide-sulfide copper ore, Ma et al. [44] demonstrated the important role of sulfur-oxidizing microorganisms capable of oxidizing sulfur and polysulfide compounds formed on mineral surfaces while maintaining the acidity of the leaching medium. In column experiments conducted by Velásquez-Yévenes et al. [45], increasing the microbial concentration raised copper recovery from low-grade chalcopyrite ore from 32 to 44% in 0.45 m high columns and from 30 to 40% in 1.0 m high columns. The applicability of filamentous fungi to the bioleaching of sulfide-bearing materials was demonstrated by Ilyas et al. [39], who investigated metal recovery from sulfide ore using *Aspergillus niger*.

4. Conclusions

Physicochemical, mineralogical, and microbiological investigations were conducted on copper-bearing ore from the Balkhash region. The ore was characterized by a complex mineralogical composition, with copper occurring in both oxide and sulfide forms. Preliminary percolation tests showed that increasing the sulfuric acid concentration from 5 to 15 g/dm³ increased copper recovery from 42.42 to 46.69%. Since only a minor difference was observed between the results obtained at H_2SO_4 concentrations of 10 and 15 g/dm³, a concentration of 10 g/dm³ was selected as the rational value for further investigation.

Pretreatment of the ore with a microbial consortium comprising iron- and sulfur-oxidizing bacteria and filamentous

fungi was found to improve the efficiency of subsequent sulfuric acid leaching. For crushed ore with a particle size of less than 10 mm, copper recovery increased from 56.75 to 72.0%, corresponding to an increase of 15.25 percentage points. For coarse ore with a particle size of less than 200 mm, copper recovery increased from 54.45 to 66.3%, corresponding to an increase of 11.85 percentage points. The highest copper recovery of 72.0% was achieved for crushed ore with a particle size of less than 10 mm.

The improvement in copper recovery following biological pretreatment may be attributed to the oxidation of sulfide minerals, the regeneration of Fe^{3+} ions by iron-oxidizing bacteria, and the action of organic acids produced by filamentous fungi. The results demonstrate the potential of a combined process involving biological pretreatment followed by sulfuric acid leaching for the treatment of low-grade and mineralogically complex copper ores. The proposed approach may provide a basis for the further optimization and substantiation of heap-leaching parameters for copper-bearing mineral resources.

Author contributions

Conceptualization: AK, TS; DM; Data curation: MY, DM, AB; Formal analysis: MY, DM; Funding acquisition: BK; Investigation: AK, TS, MY, DM, AB; Methodology: AK, TS; Project administration: BK; Resources: BK; Software: DK; Supervision: AK; Validation: TS, DK; Visualization: MY, DM; Writing – original draft: AK, BK, TS, MY, DM, AB, DK; Writing – review & editing: AK, BK, MY, AB, DK. All authors have read and agreed to the published version of the manuscript.

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Conflicts of interest

The authors declare no conflict of interest.

Data availability statement

The original contributions presented in the study are included in the article, further inquiries can be directed to the corresponding author.

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Дослідження процесу вилугування міді з мідевмісної руди із застосуванням біологічної комбінації бактерій та мікрогрибів

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Мета. Оцінювання впливу попередньої обробки мідевмісної руди біологічною комбінацією залізо- та сіркоокиснювальних бактерій та мікрогрибів на ефективність подальшого сірчаноокислотного перколяційного вилугування міді.

Методика. Вихідну руду досліджували методами хімічного, рентгенофлуоресцентного, рентгенофазового та мінералогічного аналізів. Термодинамічні умови існування сполук міді оцінювали за діаграмою Eh – рН системи Cu – Fe – S, побудованою у програмному комплексі HSC Chemistry 8. Залізо- та сіркоокиснювальні мікроорганізми та мікрогриби виділяли і культивували на селективних живильних середовищах. Перколяційне вилугування проводили розчинами H₂SO₄ концентрацією 5, 10 і 15 г/дм³. У колонних експериментах тривалістю 145 діб порівнювали стандартне сірчаноокислотне вилугування та вилугування після попередньої біологічної обробки руди крупністю менше 10 і 200 мм.

Результати. Вміст загальної міді у вихідній руді становив 0.71%, окисненої міді – 0.57-0.58%, заліза – 6.9%, сульфат-іона – 2.57%. Встановлено наявність оксидних і сульфідних форм міді, зокрема халькопїриту. У складі мікробної асоціації виявлено залізо- та сіркоокиснювальні бактерії, гетеротрофні мікроорганізми і мікрогриби. При підвищенні концентрації H₂SO₄ від 5 до 15 г/дм³ вилучення міді зросло з 42.42 до 46.69%, а концентрацію 10 г/дм³ прийнято для подальших випробувань. Попередня біологічна обробка підвищила вилучення міді з руди крупністю менше 10 мм з 56.75 до 72.0%, а з руди крупністю менше 200 мм – з 54.45 до 66.3%.

Наукова новизна. Експериментально встановлено вплив попередньої обробки комбінацією залізо- та сіркоокиснювальних бактерій і мікрогрибів на подальше сірчаноокислотне вилугування міді з руди різної крупності. Показано, що поєднання бактеріального окиснення сполук заліза та сірки з дією метаболітів мікрогрибів сприяє підвищенню доступності мідевмісних мінеральних фаз для кислотного розчинення.

Практична значимість. Отримані результати можуть бути використані під час обґрунтування параметрів комбінованої біологічної та сірчаноокислотної обробки бідних і складних за складом мідних руд, а також під час розроблення технологічних схем купчастого вилугування мідевмісної мінеральної сировини.

Ключові слова: мідевмісна руда; перколяційне вилугування; колонне вилугування; сірчана кислота; біологічне окиснення; бактерії; мікрогриби; вилучення міді

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