Abstract: With a presumed steady decline of gold ore grade in mineral resources, mining applications enabling efficient metal extraction from low-grade ores are of increasing interest to the minerals industry. Microbial processes may provide one such solution since they can participate in the biogeochemical cycling of gold in many direct and indirect ways. This review examines current literature on the role of microorganisms in gold processing and recovery. The review covers aspects such as the biotechnical pretreatment of gold ores and concentrates, microbially catalysed permeability enhancement of ore bodies, gold solubilisation through biooxidation and complexation with biogenic lixiviants, and microbially mediated gold recovery and loss from leach liquors.

Key words: gold extraction, ores, deposit, bioleaching, review.

Introduction

The role of biological agents in the mining industry is currently limited to the use of microorganisms in bioleaching and bioremediation. However, there are a number of ways in which biotechnology will be used in the near future to aid the mining industry. This review focuses on the development of novel biotechnologies and the role they will play in gold exploration, processing and remediation. The development of these biotechnologies has been enabled by advances in our molecular-level understanding of the role microorganisms play in the solubilisation, dispersion and precipitation of gold, brought upon by the rapid development of molecular genetic techniques over the past decade. This fundamental knowledge is now being used to develop new methods for gold exploration, processing and remediation.

An understanding of the distribution of microbial species in soils overlying mineralization can be utilized to develop bioindicator systems that assist with gold exploration. An in-depth knowledge of how microorganisms interact with gold complexes is being used to develop biosensors, further supporting exploration. Processing technologies are being improved based upon advances in our understanding of the interactions between microorganisms, cyanide and gold. For instance, cyanide-producing microorganisms are being investigated for use in situ leaching of gold. In turn, the use of cyanide-utilizing microorganisms for the degradation of cyanide is being explored. Combined the implementation of biotechnologies in the gold mining sector is set to revolutionize the industry, leading to the greener, more efficient extraction of gold [1, 2].

Chemical leaching and biooxidation stages were examined in a two-stage biooxidation process of an auriferous sulfide concentrate containing pyrrhotite, arsenopyrite and pyrite. Chemical leaching of the concentrate (slurry density at 200 g/L) by ferric sulfate biosolvent (initial concentration at 35.6 g/L), which was obtained by microbial oxidation of ferrous sulfate for 2 hours at 70°C at pH 1.4, was allowed to oxidize 20.4% of arsenopyrite and 52.1% of sulfur. The most effective biooxidation of chemically leached concentrate was observed at 45°C in the presence of yeast extract. Oxidation of the sulfide concentrate in a two-step process proceeded more efficiently than in one-step. In a two-step mode, gold extraction from the precipitate was 10% higher and the content of elemental sulfur was two times lower than in a one-step process [3].

Plant design parameters for gold extraction, leach residence time and cyanide consumption are generally determined from standard bench-scale bottle roll or agitation leach tests. The application of laboratory data to process design has essentially evolved from the testing of oxide or low cyanide-consuming ores. Such scale-up factors may not be appropriate for ores that deplete cyanide and oxygen since this interferes...
with gold extraction kinetics. High cyanide-consuming ores also exacerbate the differences between the conditions applicable to small batch tests and large continuous operations. Testing a reactive high-sulfide South American ore deposit necessitated the re-evaluation of conventional bench-scale cyanidation procedures. Pulp density can be a major factor affecting the results for plant design purposes and should be treated as a test work variable. As with high clay-bearing ore, where pulp viscosity can hinder mass transfer, minimizing the influence of test equipment on gold extraction kinetics may require low pulp densities. This introduces interpretative complexity since cyanide consumption changes as a function of pulp density. Activated carbon also consumes cyanide, so its effect has to be accounted for in the case of carbon-in-leach (CIL) or carbon-in-pulp (CIP) plant flowsheets. Since cyanide is an expensive reagent, often a major component of plant operating costs, it is important that the scale-up of cyanide consumption from laboratory to plant be representative. The traditional method of reporting cyanide consumption in bench-scale testing is questioned since it takes credit for residual free cyanide in leach tails. The projected plant consumption may be better estimated by using the total cyanide addition in a laboratory test. We propose a test work protocol that categorizes the cyanide consumption into its components but requires more extensive testing to generate design data. The ultimate validation of scale-up factors used by project engineers requires rigorous bench-scale versus commercial plant comparative data. Such data are not readily available [4].

Rapid technological advancement and the relatively short life time of electronic goods have resulted in an alarming growth rate of electronic waste which often contains significant quantities of toxic and precious metals. Compared to conventional chemical recovery methods, bioleaching has been shown to be an environmentally friendly process for metal extraction. In this work, gold bioleaching from electronic scrap material (ESM) was examined using batch cultures of the bacterium Chromobacterium violaceum which produces cyanide as a secondary metabolite. Gold was bioleached via gold cyanide complexation. The ESM was pretreated using nitric acid to dissolve the base metals (mainly copper) in order to reduce competition for the cyanide ion from other metals present in ESM. ESM was added to the bacterial culture after it reached maximum cyanide production during early stationary phase. Leaching with spent medium using bacterial cell-free metabolites showed a higher gold recovery of 18%, compared to that of two-step bio-

leaching of 11% at 0.5% w/v pulp density of ESM. Gold bioleaching was further enhanced to 30% when the pH of the spent medium was increased to shift the equilibrium in favor of cyanide ions production. Spent medium bioleaching of pretreated ESM yield a higher gold recovery compared to two-step bioleaching at a pulp density of 0.5% w/v [5].

Some of refractory gold ores represent one of the difficult processable ores due to fine dissemination and interlocking of the gold grains with the associated sulphide minerals. This makes it impossible to recover precious metals from sulphide matrices by direct cyanide leaching even at high consumption of cyanide solution. Research to solve this problem is numerous. Application of bacteria shows that, some types of bacteria have great affect on sulphides bio-oxidation and consequently facilitate the leaching process. In this paper, leaching of Saudi gold ore, from Alhura area, containing sulphides before cyanidation is studied to recover gold from such ores applying bacteria. The process is investigated using stirred reactor bio-leaching rather than heap bio-

leaching. Using statistical analysis the main affecting variables under studied conditions were identified. The design results indicated that the dose of bacteria, retention time and nutrition K\textsubscript{2}SO\textsubscript{4} are the most significant parameters. The higher the bacterial dose and the bacterial nutrition, the better is the concentrate grade. Results show that the method is technically effective in gold recovery. A gold concentrate containing >100 g/t gold was obtained at optimum conditions, from an ore containing < 2 g/t gold i. e., 10 ml bacterial dose, 6 days retention time, and 6.5 kg/t K\textsubscript{2}SO\textsubscript{4} as bacteria nutrition [6].

The beneficiation of mineral substances through biotechnology forms the objective and basis of the present study of the pyrite bacterial leaching (bioleaching) process. The use of ferrous and sulfide oxidizing bacteria in bioleaching processes is a recent technique that has found industrial applications in copper, uranium, cobalt and gold extraction. To pursue its application and extend it to the upgrading of other ores, its technical and economic viability must be continually demonstrated and optimized through credible technological innovations in terms of scale-up and better control of biochemical activity. This is what this work aims to achieve by improving our knowledge of the intimate mechanisms governing the action of microorganisms during pyrite bacterial oxidative dissolution. The substrate used is formed of finely ground pyrite. The cultures (classical nutritive medium without ferrous iron) are batch-prepared and kept at a temperature of 35°C. agitated and aerated.
The bacterial population used comprises three species: *Thiobacillus ferrooxidans*, *Thiobacillus thiooxidans* and *Leptospirillum ferrooxidans*. Specific equipment was developed and adapted for the study, including the design of special pyrite electrodes and the use of electrochemical methods for corrosion and interface investigations. These tools served to identify and monitor the electrochemical reactions occurring during the bioleaching, both in solution and on the surface of the pyrite electrodes. The work consisted in relating the observations of the changes in certain key electrochemical parameters to the presence of Fe$^{2+}$ and Fe$^{3+}$ ions in solution. The electrochemical behavior of pyrite during bioleaching was studied by continuous measurement of certain electrochemical parameters in different situations, both natural and induced. The overall chemical process of pyrite bioleaching was determined and subdivided into distinct elementary stages. The key factors of each elementary stage and their respective roles were identified. This made it possible, for each stage, to differentiate the electrochemical reactions occurring in solution and at the interfaces, which, when combined, lead to overall reactions that advance the bioleaching of pyrite. The results completely contradict the theory of direct dissolution of the pyrite by the bacteria and indicate that (i) the ferric ion hence remains the only powerful oxidant of pyrite during its bacterial leaching, (ii) the only role of the bacteria is (re)generation of ferric ions in solution, and (iii) the process of bacterial adhesion or contact or attack no longer has the same meaning as was hitherto attributed [7].

A quick, simple and reproducible method for the indirect determination of the population of the absorbed and suspended bacteria in the cultivation media is the Bradford’s method. It has been used to evaluate the fraction of bacteria adhered to the mineral during the bioleaching process and the suspended fraction. It was observed that a significant fraction of the bacteria is adhered to the mineral surface. This proportion varies with the environmental conditions, mainly pH, the presence of ferric ions and the way to supply the mineral sulphide. It has also been observed that the fraction adhered to the solid shows a progressive loss of its oxidative activity. The results can help demonstrate that the sulphides leaching is due to the combination of the direct bacterial action on the sulphide mineral surface with the indirect leaching by the ferric ion produced by the bacteria into the solution [8].

The bacterial oxidation of sulfide ores can be exploited in the mining industry to leach and extract heavy metals from metal sulfide deposits. Conventional methods for the extraction of precious metals from such complex sulfidic matrices are highly energy consuming and notorious for environmental pollution problems. Precious metals in the form of grains are very finely distributed and encapsulated in the sulfide matrices and direct cyanidation of the refractory sulfide ores usually gives unsatisfactory recovery of such metals. Bio-oxidation of a gold-bearing pyrite concentrate has been considered as an alternative process for the pretreatment of such a refractory ores prior to cyanidation. In this research, mixed cultures of *Thiobacillus ferrooxidans*, *Thiobacillus thiooxidans*, and *Leptospirillum ferrooxidans* strains of DSM culture collection were examined batchwise for their ability to oxidize iron portion of gold-bearing pyrite concentrate from Mouteh mine in Isfahan. Effects of initial levels (i.e. ratio of the bacterial cell numbers in the inoculum) and ore adaptation of mixed cultures of the above named bacterial species on bioleaching of the sulfide pyrite has been studied. Time course of changes in iron concentration was determined. Iron dissolution during bioleaching could be used as an index for release of gold, which has been intergrown in the sulfidic matrix. Mixed cultures of these strains of three bacterial species acted efficiently in leaching iron and the variables indicated above had positive influence on the rate of iron dissolution. In order to characterize kinetics of the bio-oxidation in terms of the logistic equation, the data obtained were fitted to this equation. A linear regression was performed on the data in their logarithmic form and logistic model parameters were calculated. The logistic equation was found to be a good fit to all of the data obtained in this study [9].

A proposal for a modified in situ leaching method for extracting gold from oxidized gold ores using a non-cyanide lixiviant is described. A non-cyanide lixiviant is suggested because of the obvious concerns posed by injecting cyanide-bearing solutions into the subsurface. Oxidized gold ores were chosen as a focus because earlier research on the use of sodium thiosulphatic as a lixiviant under anaerobic conditions indicated that the presence of pyrite led to rapid thiosulphatic breakdown. A reconnaissance research program involving ore characterization and hydrometallurgical test work on samples from four Australian ore deposits and preliminary reactive transport modeling studies was carried out. This work showed that lixiviant-oxidant combinations of sodium thiosulphatic and ferric EDTA and iodide and iodine are both capable of extracting high percentages of accessible gold from the selected samples in bottle roll tests under anaerobic conditions. The ore
characterization and reactive transport studies suggested that both physical and chemical methods of permeability enhancement may be required to lift bulk permeability and the availability of gold for dissolution to sufficiently high levels to obtain adequate gold recoveries. Assuming that such methods prove to be both necessary and economically viable, the mining method would no longer be regarded as simple in situ leaching. Therefore, the term «in-place leaching» has been adopted for the proposed gold extraction system [10].

The intensification of the thiosulphatic leaching of silver, gold and bismuth from sulphide concentrates using mechanical activation and mechanochemical pretreatment step was investigated. The physico-chemical changes in a complex sulphide concentrate (Casapalca, Peru) as a consequence of mechano-chemical pretreatment had a pronounced influence on the subsequent silver extraction. The optimum results from mechanochemical pretreatment and subsequent leaching of the concentrate with ammonium thiosulphatic were achieved with 99% recovery of Ag after only 3 min of leaching. The leaching of gold from a mechanically activated complex sulphide concentrate (Banska Hodrusa, Slovakia) using ammonium thiosulphatic was studied as follows. Physico-chemical transformations in the concentrate due to mechanical activation have an influence on the rate of extraction and the recovery of gold. It was possible to achieve 99% Au recovery within 45 min for a sample mechanically activated. Mechanical activation proved to be an appropriate pretreatment for this concentrate before extraction of gold into thiosulphatic leaching solution. The selective leaching of bismuth from the lead concentrate (Atacocha, Peru) by using of sodium thiosulphatic and mechanical activation as the pretreatment step was examined as the last example. It is possible to achieve more than 90% recovery of bismuth in leachate even in three minutes for mechanically activated samples [11].

Heap leaching with thiosulphatic is only used in one location for the treatment of a refractory preg-robbed gold ore initially pre-treated with bio-oxidation. The chemistry of thiosulphatic leaching is complex. Further work is required to develop a better control of the system. A mild refractory gold ore containing pyrite and chalcopyrite was used in this study to investigate the use of thiosulphatic as an alternative heap leaching technology. Preliminary bottle rolls tests indicated that similar gold extraction was obtained with cyanide and thiosulphatic. In the column leach test, effects of thiosulphatic, copper and ammonia concentrations, and their ratio on both gold extraction and reagent consumption, were assessed. The range of reagent concentration were: 0.1 – 1.0M (NH₄)₂S₂O₃, 0.03 – 0.10M CuSO₄·5H₂O, 1.0 – 6.0M NH₄OH. The solid-liquid ratio was in the range of 0.8:1 to 5.1. Best results showed that 72% of gold was extracted in 50 days with 0.3M (NH₄)₂S₂O₃, 0.05M CuSO₄·5H₂O and 6M NH₄OH or a combination of 1.0M (NH₄)₂S₂O₃, 0.03M CuSO₄·5H₂O and 3M NH₄OH. The reagent consumption at solid/liquid ratio of 3.1 was 37 – 48.6 kg/t (NH₄)₂S₂O₃ and 0 – 0.62 kg/t CuSO₄·5H₂O. The results of thiosulphatic leaching were also compared with cyanide leaching [12].

The leaching of gold using alkaline amino acids-hydrogen peroxide solutions at low concentrations has been studied. The application of alkaline amino acid-hydrogen peroxide system may offer an alternative and environmentally benign process for gold leaching, particularly in the context of leaching low grade gold ores in an in-situ or in heap leach processes. In the presence of an oxidant or oxidants, it was found that amino acids can dissolve gold at alkaline condition at low and moderate temperature. Heating the leach solution between 40 and 60 degrees C was found to enhance the gold dissolution significantly in alkaline amino acid-peroxide solutions. It was also found that gold dissolution increases by increasing amino acid concentration, peroxide and pH. Amino acids acts synergistically to dissolve gold. Although glycine showed the highest gold dissolution as a single amino acid compared to histidine and alanine, histidine was found to enhance gold dissolution when used in equimolar amounts with glycine. The presence of Cu²⁺ ion enhances gold dissolution in the glycine-peroxide solutions. The process will propose an environmentally benign process for gold treatment in order to replace the use of cyanide in heap or in-situ leaching. In the presence of pyrite, the amount of gold leached was lower due to the peroxide consumption in sulphide oxidation [13].

Bioleaching is one of advanced technologies of processing of gold-consisting ores with such advantages as low-wasteness and ecological purity since gas and dust are not emitted into the atmosphere [14]. The technology of bioleaching is simple in application and highly effective, especially for processing of ores with low content of precious metals. It allows to save materials and energy and in the future can replace such ways of processing of mineral raw materials as roasting, autoclave leaching, metallurgical melting which pollute environment with poisonous gases and toxic chemicals. In practice of bioleaching there are used various microorganisms depending.
on aims. Acidophile thionone bacteria from the sort Acidithiobacillus ferrooxidans are the most popular. In the USA 20% of production of copper is the share of its receiving from dumps of off-balance ores by means of the above bacteria. In China and Mexico there are skilled installations for bacterial leaching of copper concentrates. The assessment of the similar enterprises shows that at capital expenditure for building of installation about 900 thousand dollars prime cost of 1t of the received product makes less than 50 dollars, time of payback of installation – 18 months. That is, in a year after installation development the profit makes 375 thousand dollars [15].

The Pacific Ore Technology company at the enterprise «Radio Hill» (Australia) operates trial installation of compact bacterial leaching of copper-nickel ore, this technology, according to experts, can be applied to bacterial obduction of persistent gold-sulphide ores. Sulphidic minerals make about 15% of total amount of ore. Ores split up to a class of fineness minus 7,5 mm and are put on the waterproofing basis in stacks of 5 m high, with a lump of 1000 t. Process continues for about a year, so non-ferrous metals are extracted in concentrates for 70-90% [16].

It is known that leaching is transfer to solution (usually water) of one or several components of strong substance by means of water or organic solvent, it is frequent with the participation of gases of oxidizers or reducers [17].

The work purpose – research of regularities and the technological scheme of gold extraction from ores of Bakyrchik deposit, the East Kazakhstan region, by percolation bioleaching.

**Materials and methods**

There are several some ways of leaching, in this experiment there was applied a percolation leaching by infiltration (percolation) of liquid reagent of solution through a motionless layer of solid material. The percolation way is imitation of a compact way of leaching. Generally percolation leaching is applied, for example, for processing gold-consisting porous structure with particles of 0.2-1.0 mm in special round sand of porous structure with particles of 0.2-1.0 mm in special round (height of 2-4 m, diameter of 12-14 m) and rectangular (length of 25 m, width of 15 m) tubs capacity on sand of 800 – 900 t; duration of full processing of one sand loading is 4-8 days. Percolation leaching is carried out by consecutive supply of solutions of cyanides alkaline alkaline-agrarian metals of the decreasing concentration from 0.1 – 0.2 to 0.03 – 0.05%.

In the administrative relation the Bakyrchik mining enterprise is located in the Zharminsky Area of East Kazakhstan region at distance of 90 km to the southwest from the regional center of Ust-Kamenogorsk. In close proximity to the enterprise on the southwest there is an Auezov working settlement, in 4 km to the West – the settlement of Shalobai (Figure 1). Transport system of the enterprise and settlements with the regional center and Semipalatinsk which is in 170 km to the northwest.

**Results and their discussion**

Gold leaching by solutions of cyanide of sodium was carried out in the following mode: at the 1 stage (to 20% of extraction of gold) concentration of cyanide of sodium is 0.6 g/dm³, density of an irrigation is 25 dm³/t of ore, without pauses in an irrigation; at the 2nd stage (20-40% of extraction of Au) concentration of NaCN is 0.4 g/dm³, density of an irrigation is 15 dm³/t of ore, a pause in an irrigation – 1 day; at the 3rd stage (extraction of gold 40%) concentration of NaCN is 0.2 g/dm³, density of an irrigation is 5 dm³/t of ore, a pause in an irrigation of 2 days, Figure 2. Stage-by-stage decrease in density of irrigation allows maintaining concentration of gold in solution at higher level, in this case 2 mg/dm³, рН of initial solution of cyanide of sodium in all cases was equal to 10.

After each irrigation the whole solution was removed from a leaching cycle, i.e. the ore irrigation in each cycle was carried out by fresh alkaline solution of cyanide of sodium. The gold-consisting solution removed from a turn was analyzed on the content of gold, residual cyanide, alkaline, the content of iron and non-ferrous metals was periodically defined: cobalt, nickel, zinc, copper.

![Figure 1 – Geographical location of Bakyrchik gold-arsenous deposits](image-url)
The general duration of leaching made 162 days, the maximum concentration of gold in solution is 5.63 mg/dm³. Extent of extraction of gold in solution for the entire period of leaching is 75.17%. In process of increase in duration of leaching, on each of three stages concentration of gold increased in solution in the beginning, and then evenly decreased.

Dynamics of gold extraction at percolation ore leaching of the Bakyrchik deposit. II – stage was given on Figure 3.

The average content of gold in solution at 1st stage of leaching (duration of 16 days) is 1.93 mg/dm³, at the 2nd stage (22 days) – 3.13 mg/dm³, at...
the 3rd stage (124 days) – 4.08 mg/dm³, extraction of the 1 irrigation 1.40, 1.81, 0.78% respectively. For the entire period of leaching average concentration of gold in solution made 2.50 mg/dm³, extraction of the 1 irrigation – 1.09%. The most noticeable decrease of gold concentration was observed at the final stage of leaching. In the rage from 65th to 69th irrigation, concentration of gold in solution decreased from 2.54 to 1.54 mg/dm³. For receiving the solutions (2mg/dm³), which were more concentrated on gold at the final stage of leaching it is recommended to periodically carry out an irrigation without removal of solutions. Dynamics of gold extraction at percolation ore leaching of the Bakyrchik deposit. III – stage, Figure 4.

It is revealed that cyanation of gold-arsenic concentrates of Bakyrchik deposits after bacterial leaching leads to gold extraction increase, and the best results are received when procedures of thiosulphatic and sulphatic leaching are preceded by A. ferrooxidans culture processing.

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